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What regulates decomposition in agroecosystems? Insights from reading the tea leaves

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Litter decomposition is a critical Earth process, recycling nutrients and setting a portion of plant tissue on a path toward soil organic matter. Despite this importance, we still lack a good understanding of local factors that regulate decomposition, especially in agroecosystems where management plays an outsized role. Using a narrow range of climate and soils, we buried 1,308 pre-manufactured "litter bags" of differing residue quality (i.e., green and rooibos tea leaves) in 109 plots across several management practices to (1) explore the local controls on decomposition in agroecosystems and (2) test the robustness of the Tea Bag Index (TBI). We found that management practices intended to increase soil ecosystem services, that is, soil health, altered the decomposition of both teas. For example, adding nitrogen fertilizer and implementing perennial cropping decreased the extent of green tea decomposition (carbon-to-nitrogen ratio, or C:N = 12.8). No-tillage increased, but perennial cropping decreased, the rate of rooibos tea decomposition (C:N = 50.1). Cropped prairie accelerated green tea decomposition and increased the extent of red tea decomposition. A random forest regression model showed that soil temperature was the strongest predictor of green tea decomposition, but a soil health score also played a significant role in predicting the mass remaining. Soil texture and nutrient availability best predicted rooibos tea decomposition. Finer textured soils seemed to decelerate rooibos decomposition but increased the extent of decomposition. Furthermore, we demonstrated that the TBI metrics correlated somewhat well with empirically derived decomposition constants and were similarly sensitive to the effects of management. Still, the green tea stabilization factor had a substantial prediction bias. Our study increased our basic understanding of what regulates decomposition in agroecosystems. It also showed that the TBI can be a scientifically rigorous citizen science approach to monitoring changes in soil health.

machine learning, nutrient cycling, regenerative agriculture, random forest regression, soil organic carbon, Tea Bag Index (TBI)

1 Introduction

Litter decomposition is a critical function of ecosystems around the world. As a snapshot, approximately 286 petagrams (Pg), or 10¹⁵ grams, of carbon (C) and 13 Pg of nitrogen (N) are stored in global plant litter stocks (Potter and Klooster, 1997). This massive stock of decomposing plant material plays a vital role in terrestrial carbon (C) sequestration, regulating soil fertility, and providing habitat for biota (Jenkinson et al., 1991; Freschet et al., 2013). It is no wonder that researchers have spent decades decomposing litter in many environments and under a plethora of experimental conditions, all to better understand decomposition and improve predictions in biogeochemical models.

Currently, we know that litter decomposition is a function of three major factors: litter quality (usually expressed as C-to-N ratio), climate, and decomposer organisms (Swift et al., 1979; Bradford et al., 2016). Regional- to global-scale studies are critical toward improving our understanding of temperature and litter quality as primary drivers of decomposition (Parton et al., 2007; Adair et al., 2008; Bontti et al., 2009; Tuomi et al., 2009; Djukic et al., 2018; Ball et al., 2022); however, these large-scale studies miss the mark on more nuanced, local factors (<1 km distances) that regulate decomposition where climate may be similar. Agroecosystems are a prime example, where farmer decisionmaking at the field-scale will change many regulating factors such as soil microclimate, legacy residue quality, nutrient availability, and decomposer organisms. How agroecosystem management affects decomposition has received less attention compared to natural ecosystem counterparts, despite the strong influence that management decisions have on litter (or residue) decomposition (Beare et al., 1992; Wickings et al., 2012; McDaniel et al., 2023).

Most decomposition studies, whether in natural or managed ecosystems, use the "litter bag" method to estimate a decomposition rate and study nutrient composition/release of remaining litter (Wider and Lang, 1982; Karberg et al., 2008). The litter bag method involves placing plant litter in a plastic mesh bag, deploying several "sacrificial samples" for removal at specific time intervals, and measuring the mass remaining at each time point. Many methodological variations will affect the final modeled decomposition rate and mass remaining, including time intervals of retrieval and total time of decomposition, decomposition model selection, changing the size of holes in the mesh, whether litter bags are buried or placed on the surface of the soil, and size and quality of the litter (Santos et al., 1984; Bradford et al., 2002; Karberg et al., 2008; Bokhorst and Wardle, 2013; Cornwell and Weedon, 2014). These decisions should be tailored to best answer specific research questions.

Although burying manufactured materials as common, decomposable substrates has been done for decades—cotton strips probably being the most familiar (Latter and Howson, 1977; Harrison et al., 1988)—a seminal 2013 study introduced new "pre-made" litter bags to the world. These researchers introduced the concept of decomposing tea bags of green (*Camellia sinensis*) and rooibos tea (*Aspalathus linearis*) to better understand what factors are regulating decomposition in soils (Keuskamp et al., 2013). Decomposing both teas of differing quality allowed for the calculation of a Tea Bag Index (TBI) that estimated the

decomposition rate and a "stabilization" factor without the onerous task of deploying several sacrificial samples (Keuskamp et al., 2013; Sarneel et al., 2024). Both the pre-manufacture teas and TBI made measuring decomposition more accessible—even to citizen scientists. The TBI method was groundbreaking for improving our understanding of decomposition, as evidenced by the many studies following the original 2013 study (Houben et al., 2018; Petraglia et al., 2019; Duddigan et al., 2020a; Pino et al., 2021), even including some aquatic decomposition studies (Marley et al., 2019; Mori et al., 2023).

Agroecosystems are an excellent venue for using tea bags and the TBI method to better understand decomposition for a few reasons. First, agroecosystems are highly managed by humans, and the emerging soil health movement has increased interest in onfarm, citizen science monitoring after a change in management practice (Karlen et al., 2021). Indeed, decomposing tea, among other inexpensive and accessible materials (e.g., cotton and birch tong depressors), proved to be comparable in their ability to detect differences among soil health-promoting practices from long-term experiments in Iowa, USA (Middleton et al., 2021). Second, the two teas are at opposite ends of the residue quality spectrum, with green being "high" and similar to leguminous crop residue (C:N = 12-14) and rooibos being "low" and more similar to the chemical composition of high-N-demanding crop residues (C:N =43-78) (Keuskamp et al., 2013; Marley et al., 2019; Duddigan et al., 2020b; Middleton et al., 2021). Finally, from a fundamental science perspective, agroecosystems are also ideal because it is the norm for plant residues to be mixed with disturbed soil. This is unlike many natural ecosystems, where it is atypical for aboveground plant material to get mixed with disturbed soil.

Given the importance of residue decomposition in agroecosystems, the intimate contact that residue has with agricultural soils, and our lack of understanding of local controls on decomposition, we set up a study decomposing green and rooibos teas across a variety of management practices in the USA Corn Belt. Notably, these management practices all occurred on similar soils and under a relatively identical climate. Our primary research questions were as follows:

- 1. Do the easy-to-deploy TBI decomposition metrics correlate with empirically measured decomposition metrics, and are management treatment effects consistent across both methods?
- 2. How do management practices alter empirically derived decomposition kinetics of both teas (green and rooibos)?
- 3. What are the overarching regulators, at a local scale, on decomposition of a high- and low-quality residue?

We predict that the TBI index ($k_{\rm TBI}$) estimated with just one 68-day mass loss measurement of both teas will correlate with observed decomposition rates measured the traditional way of using seven sacrificial samples. We hypothesized that increasing the soil health-promoting practices—minimizing disturbance, increasing plant inputs, increasing plant diversity, and adding animal manure—will increase decomposition rate (k) but especially of rooibos tea, which is poorer "quality" (i.e., lower C:N and more lignin) because of enhanced biological activity and increased labile resources in the soil matrix (McDaniel et al., 2014). To answer the third question, we measured 67 variables across nine long-term experiments and

used a machine learning random forest regression (RFR) model to determine what soil and management properties best predict decomposition under similar climate and inherent soil conditions.

2 Materials and methods

2.1 Site characteristics and experimental design

We carried out this study in 2018 in Iowa, USA, at nine long-term agroecosystem experiments. Iowa has a humid continental climate with hot summers and cold winters. Soils under these nine long-term experiments were developed under glacial deposits and/or a loess cap. All soils were classified under the Mollisol soil order under the United States Department of Agriculture (USDA) classification (Soil Survey Staff, 2024). Basic soil properties (mean \pm standard error) include: 29 \pm 4% clay, 3.8 \pm 0.9% soil organic matter, 6.6 \pm 0.8 pH, and 24.3 \pm 7.8 meq 100 g $^{-1}$ cation exchange capacity. See Table 1 for more details on soil properties.

We grouped nine long-term experiments into categories based on eight management interventions (Table 2; Supplementary Table S1; Supplementary Figure S1). The nine long-term experiments making up these management interventions were all randomized complete block design experiments with three to six randomized, blocked replications (Supplementary Table S1). Most experiments had four replications, and in total, there were 109 individual plots. At the time we buried tea and sampled soils, these experiments were established at different times, ranging from three (Tejera et al., 2019) up to 20 years since establishment (Liebman et al., 2008). Soil properties and tea decomposition were both measured at the plot level.

A snapshot of how these management practices affect tea, birch tongue depressors, and cotton decomposition at isolated points in time was previously published in Middleton et al. (2021). Here, we focus on the decomposition dynamics of the tea only, including seven time points to model the decomposition rate and mass remaining. Most importantly, here, we explore mechanisms that regulate the decomposition of the two teas with machine learning.

2.2 Decomposition of green and rooibos teas

We measured mass loss of green (*C. sinensis*) and rooibos (*A. linearis*) tea leaves during the 2018 growing season (Supplementary Figure S2A). Chemical composition and moisture content of the off-the-shelf Lipton[®] teas were analyzed previously (Supplementary Table S2; Middleton et al., 2021), and confirmed similarity with other TBI studies (Keuskamp et al., 2013; Marley et al., 2019; Duddigan et al., 2020b).

The pre-manufactured tea bags (mesh = 0.25 mm) were reinforced with nylon string and labels with electrical tape to make them easier to find. We buried 756 tea bags to a depth of 8 cm in the soil after any spring tillage and/or spraying between 12 June and 20 July (Supplementary Figure S2). The tea bags were buried in the plot to get good coverage and randomly assigned a retrieval date. All tea bags were buried equidistant from the center of the crop

row and the interrow to equally assess both more root-influenced and less root-influenced soils. In other words, for maize, which was the majority of plots, crops are 76 cm apart, and tea bags are buried $\frac{1}{2}$ of the distance (19 cm) in either direction.

The tea bags were retrieved at 4, 7 ± 1 , 14 ± 1 , 30 ± 2 , 68 ± 2 , and 130 ± 10 days after burial, depending on the weather conditions (Supplementary Figure S2). We used a trowel and a soil knife to gently excavate the tea bags from their location. Only 15 tea bags were lost out of all deployed, mostly due to wildlife damage. In the field, tea bags were placed in a Ziploc bag and stored in a cooler with icepacks. Back at the laboratory, tea bags were temporarily stored, 3–7 days, at 4 °C until processing to prevent further mass loss.

To process, we gently cleaned the soil off the tea bags. The entire tea bag was then dried in a forced air oven at 50 $^{\circ}$ C for 3 days to stabilize mass for storage until weighing. We weighed the tea bag and reinforcement materials, then gently cut them, and weighed the contents (the tea and any soil particle contamination) alone. The contents were transferred to a crucible and heated in a muffle furnace at 530 $^{\circ}$ C for 8 h to measure combustible organic material by subtracting the remaining ash content.

2.3 Soil and crop measurements

Soil temperature and moisture were measured within every plot during every site visit, from burial to final retrieval (n=7 through time). Volumetric water content (VWC) was measured at 0–4.5 cm using an ML3 ThetaProbe Soil Moisture Sensor (DeltaT Devices; Cambridge, UK). Soil temperature was measured with a digital stem thermometer to a depth of 5 cm. We collected three measurements in close proximity to the tea bags per plot for a plot average at each sampling event.

We collected two soil samples from each plot—at burial and final tea bag retrieval (18 October to 20 November)—by compositing 10–15 soil cores (0–15 cm deep and 2 cm diameter). From here on, these two separate sampling events will be referred to as spring and autumn sampling, respectively. After collecting, soil samples were stored on ice until they could be transported to the laboratory, where they were refrigerated at 4 °C until they could be processed and analyzed. Soils were sieved fresh through a 2-mm sieve. Then, only a few analyses were conducted on fresh soils (e.g., microbial biomass, inorganic nitrogen, and gravimetric water content). The remainder of the analyses were conducted on soils that were air dried for 1 month at room temperature (~25 °C, Table 1).

Microbial biomass C and N (MBC and MBN) were analyzed using the chloroform-fumigation, extraction method with 5 g soil and 20 ml $0.5\,\mathrm{M}$ K₂SO₄ extractant [Vance et al., 1987; modified according to Middleton et al. (2021)]. Extracts were stored frozen at $-20\,^{\circ}\mathrm{C}$ until analysis. Extracts were analyzed for non-purgeable organic C and total N (TN) via combustion on a Shimadzu TOC-L analyzer with TN capabilities (Shimadzu Corporation, Kyoto, Japan). MBC and MBN were calculated by the differences between fumigated and non-fumigated samples and corrected by the extraction efficiency factors of 0.45 (MBC) and 0.54 (MBN) (Brookes et al., 1985; Joergensen, 1996). Soil inorganic nitrate

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TABLE 1 Summary statistics of select soil properties and crop yield used as random forest regression predictor variables.

Soil property (units)	Abbreviation [‡]	Sample number	Minimum	25th quantile	Median	75th quantile	Maximum	Mean	Standard deviation
Bulk density (g cm ⁻³)	BD	109	0.75	1.16	1.25	1.32	1.49	1.23	0.12
Maximum water holding capacity (%)	MWHC	109	49.4	64.0	68.2	71.6	78.8	67.1	6.6
Fine clay (%)	FClay	109	7.8	12.3	12.9	13.9	16.3	12.9	1.4
Clay (%)	Clay	109	17.4	26.7	29.2	31.6	36.1	28.8	3.7
Sand (%)	Sand	109	32.1	39.5	42.3	44.2	52.8	42.1	4.1
Silt (%)	Silt	109	13.5	25.1	29.1	32.9	50.5	29.1	6.7
Particle size mode (µm)	PS _{mode}	109	0.7	15.3	17.8	19.8	103.0	18.3	11.6
Particle size mean-weighted diameter (μm)	PS _{mean}	109	29.9	64.0	79.3	93.6	138.9	79.5	22.0
Particle size kurtosis	PS _{kurt}	109	3.4	7.2	9.2	12.1	29.7	10.6	5.1
Cation exchange capacity (meq $100~\mathrm{g}^{-1}$)	CEC	109	13	19	22	28	57	24	8
Total organic carbon (%)	TOC	109	1.35	2.13	2.44	3.01	7.36	2.73	1.07
Total organic nitrogen (%)	TN	109	0.14	0.19	0.22	0.25	0.45	0.23	0.06
$Ammonium\ (mg\ N\ kg^{-1})$	NH ₄	218	0.1	0.6	0.9	2.8	96.9	4.3	8.6
Nitrate (mg N kg ⁻¹)	NO ₃	218	0.1	2.9	6.9	14.2	111.1	10.0	9.3
Soil pH in 1:1 (w:w) 0.01 M CaCl	pН	218	4.7	6.4	6.9	7.4	7.8	6.8	0.8
Permanganate oxidizable carbon (mg C kg $^{-1}$)	POXC	218	266	488	550	636	891	561	118
Soil test phosphorus via Mehlich-III extract (mg P ${\rm kg^{-1}}$)	STP	218	1	17	25	35	81	27	15
Soil test potassium via Mehlich-III extract (mg K ${\rm kg}^{-1}$)	STK	218	74	137	158	186	278	159	38
Soil test magnesium via Mehlich-III (mg Mg ${\rm kg^{-1}}$)	STmg	218	99	204	259	369	617	287	102
Soil test sulfur via CaPO $_4$ extract (mg S kg $^{-1}$)	STS	218	3	4	5	6	13	5	1
Soil test calcium via DTPA † extract (mg Ca kg $^{-1}$)	STCa	218	1,448	2,416	3,029	4,248	10,500	3,535	1,714
Soil test iron via DTPA † extract (mg Fe kg $^{-1}$)	STFe	218	8	18	30	43	147	36	26
Soil test manganese via DTPA † extract (mg Mn kg $^{-1}$)	STMn	218	3.0	5.4	7.7	10.5	34.7	9.1	5.4
Soil test copper via DTPA † extract (mg Cu kg $^{-1}$)	STCu	218	0.7	1.2	2.1	3.4	24.3	2.9	2.8
Soil test zinc via DTPA [†] extract (mg Zn kg ⁻¹)	STZn	218	0.4	0.8	1.2	1.7	6.9	1.4	0.8
Soil test boron via DTPA † , sorbitol extract (mg B kg $^{-1}$)	STB	218	0.1	0.2	0.3	0.4	0.9	0.3	0.2
Salt concentration (mmhos cm ⁻¹)	Salt	218	0.1	0.2	0.3	0.5	6.2	0.5	0.6
Microbial biomass carbon (mg C kg ⁻¹)	MBC	218	99	188	229	298	593	258	105
Microbial biomass nitrogen (mg N kg ⁻¹)	MBN	218	2	24	32	43	590	41	47
Microbial biomass C:N ratio (unitless)	MBC:N	218	<1	6	8	9	41	8	4
Potentially mineralizable carbon (mg C kg^{-1})	PMC	218	7	51	65	83	183	68	27
Potentially mineralizable nitrogen (mg N kg^{-1})	PMN	218	<1	18	21	26	110	22	9
Potentially mineralizable C:N ratio (unitless)	PMC:N	218	<1	2	3	4	22	3	2
Maize yield (Mg ha ⁻¹)	MaizeYield	92	1.4	10.5	12.6	15.1	18.6	11.7	4.2

 $^{^\}dagger \text{Diethylenetriaminepenta} \text{acetic acid;} ^{\ddagger} \text{Abbreviations used in Figure 4.}$

 (NO_3^-) and ammonium (NH_4^+) N were analyzed using colorimetric microplate methods with same non-chloroformed extracts using the vanadium (III) chloride method read at a wavelength of 540 nm (Doane and Horwáth, 2003), while NH_4^+ used the cyanurate and salicylate method at 595 nm (Sinsabaugh et al., 2000), and both colorimetric reactions were measured on a Biotek SynergyHTXTM plate reader (BioTek Winooski, Vermont).

Other indicators of soil biological activity or labile pools of C and N were analyzed on air-dried soils. Permanganate oxidizable C (POXC) was measured using a modified method of Weil et al. (2003) with 2.5 g of soil and 18 ml of deionized (DI) water, as well as 2 ml of 0.2 M KMnO₄, and the end reaction absorbance was measured on the microplate reader (Middleton et al., 2021). Potentially mineralizable C and N (PMC, PMN) were calculated by measuring carbon dioxide (CO₂) and inorganic N produced during an incubation (Middleton et al., 2021). Briefly, 5 g of soil brought to 50% maximum water holding capacity (MWHC) was incubated in 50 ml conical centrifuge test tubes for 14 days, and in addition to nitrate and ammonium produced at the end of the incubation, we measured CO₂ production in a test tube headspace on a LI-830 CO₂ gas analyzer (LI-COR, Lincoln, NE) (sensu McDaniel and Grandy, 2016).

Dried soils were analyzed for bioavailable nutrients using extracts commonly used in agroecosystems for predicting plantavailable nutrients. We assume that measuring these labile pools of macro/micronutrients will also reflect availability to soil microorganisms decomposing plant materials. Soil test phosphorus (STP), soil test potassium (STK), calcium (STCa), and magnesium (STMg) were all extracted using 2 g per 20 ml Mehlich-III extractant (Mehlich, 1984). Sulfur (STS) was extracted with 10 g of soil using 25 ml of monocalcium phosphate extractant (Combs et al., 1998). Soil test iron (STFe), manganese (STMn), zinc (STZn), copper (STCu), and boron (STB) were all measured using 10 g of soil to 20 ml of diethylenetriaminepentaacetic acid extractant, with sorbitol included in the STB extract to complex with B (Lindsay and Norvell, 1978; Miller et al., 2000). All bioavailable plant extracts were then analyzed on an ICP-OES 7300 (Perkin Elmer, Waltham, MA, USA) using calibration curves and check standards. Soil pH was measured using 0.01 M CaCl in a 1:1 (w:w) ratio with a HQ440D Hach pH meter with IntelliCAL PHC281 probe (Hach; Loveland, CO, USA).

We also analyzed dried soils for more slow-to-change variables such as soil texture, soil organic matter (SOM), MWHC, soil organic C (SOC), and total N (TN). Soil texture was analyzed using laser diffractometry (Miller and Schaetzl, 2012) with a Malvern Mastersizer 2000E laser particle size analyzer and a Hydro 2000MU pump accessory (Malvern Instruments Ltd., Worcestershire, UK). MWHC was measured using the "filter-funnel paper and drainage" method described by Nelson et al. (2023). Soil organic matter was measured using loss on ignition for 12 h at 450 °C. Soil samples were oven-dried, fumigated with HCl if pH >7.2, and rolled in tin and analyzed for SOC and TN on a Vario Max elemental analyzer (Elementar Americas, Inc., Ronkonkoma, NY, USA).

Maize yield was generally measured for the four center rows of each plot using combines equipped with yield monitors. At the Boyd Farm, plots were exceptionally small (5 rows) and required a 2-row combine. For the Sorenson Long-term Assessment of Miscanthus Productivity and Sustainability (LAMPS) experiment,

maize yield was determined by hand harvest. Maize yields were adjusted to $155\,\mathrm{g~kg^{-1}}$ moisture content and reported as Mg of grain per hectare.

2.4 Calculations

We used the three-parameter exponential decay model for modeling decomposition and calculating decomposition metrics in SigmaPlot v15 (Grafiti LLC; Palo Alto, CA). Decomposition metrics—rate (k), predicted final mass loss (Y_0) , and "initial mass" of decomposable material (a)—were calculated at the plot level to allow us to do statistics on these metrics. The equation is presented as follows:

$$M(t) = Y_0 + (a)e^{-kt} \tag{1}$$

where M(t) is the ash-free dry mass at any given day, Y_0 is the predicted final mass remaining or asymptote of the exponential decay curve, a is the final mass loss from the initial of decomposable material when t = 0, k is the decomposition rate in % mass loss per day, and t is time measured in the unit of day.

We also calculated the TBI metrics using only the 68-day time point for all plots for comparison (Keuskamp et al., 2013). The TBI stabilization factor (S_{TBI}) is calculated as follows:

$$S_{TBI} = 1 - \frac{a_g}{H_\sigma} \tag{2}$$

where a_g is the decomposable fraction and H_g is the hydrolysable fraction of the green tea (Keuskamp et al., 2013). The S_{TBI} is similar to the mass remaining of the green tea, but is thus estimated from the hydrolysable fraction and the extent of decomposition. Accordingly, the TBI decomposition rate (k_{TBI}) is calculated the same as Equation 1, but with 68-day mass remaining, and Y_0 is estimated from the decomposable fraction of the rooibos tea.

To provide a cumulative measure of soil temperature and moisture, we used eight measurements at burial (0 days) to final retrieval (130 days). The area under the curve between measurements was used to create a 130-day cumulative temperature and soil moisture for each plot. For soil temperature, this ranged from 1,749 to 2,329 $^{\circ}$ C. For volumetric soil moisture, this ranged from 1.947 to 3.634 cm³ cm⁻³.

We used a novel index of the coverage or adoption of the USDA Natural Resources Conservation Service's (NRCS) five soil health principles—maximize presence of living roots, minimize disturbance, maximize soil cover, and maximize biodiversity—as a predictive factor in the RFR model. This novel index, called the Soil Health Principle Score (or SHPS), was calculated on a scale from 0 to 5 (Supplementary Table S3; McDaniel and Middleton, 2024).

2.5 Data handling and statistical analyses

Statistical analyses were conducted in R (version 4.3.1). Data were checked for normality and heterogeneity of variances using

the ggResidpanel package (version 0.3.0). All data, before further processing, were checked for normality and heterogeneity of variances (Zuur et al., 2010). Outliers (>2 × standard deviation) were removed, and transformations were made to any nonconforming datasets. No transformations were needed, and fewer than five of our outlier values were removed per site. Data were visualized using SigmaPlot version 15.0 (Inpixon; Palo Alto, CA, USA).

We used a simple, one-way ANOVA using the *aov* function in R to determine differences in management practices on tea decomposition and set $\alpha=0.1$. For each management intervention or practice (Table 2), we compared the business-as-usual, or control treatment, vs. the experimental treatment. Management practices occurred at multiple sites, sometimes crossed with other management practices. For example, notillage and cereal rye cover crops were both individual but also combined factorially at the Gilmore City, IA experiment (Qi et al., 2011). The conventional tillage practice of disc tillage, both with and without cover crops, was aggregated with those disc tilled experiments near Boone, IA, USA, increasing the sample number for comparisons. This is reasonable for sites that had the same or similar soil series (McDaniel and Middleton, 2024), and a somewhat narrow range of soil properties (Table 1).

To compare the applicability of TBI to agroecosystem soils and a wide range of management practices, we used TBI calculations from Keuskamp et al. (2013). The TBI allows greater citizen scientist involvement by making decomposition measurement easier and more accessible. Using TBI, one can calculate a decay rate (k_{TBI}) and stabilization factor (S_{TBI}) using mass loss of green and rooibos teas at only one time point. We used a 68-day time point because the green tea mass remaining begins to plateau at 20-40 days at moderate temperatures (Keuskamp et al., 2013; Duddigan et al., 2020b). We used the cor function in R to analyze linear correlations, specifically examining the relationship between TBI and empirically derived values to determine the Pearson correlation coefficient and corresponding p-value. Bias was calculated by subtracting the predicted TBI value from the observed value, using a full decomposition curve. Relative bias was calculated by dividing the observed value by the predicted value and multiplying

Random forest regression is an ensemble learning algorithm that develops several individual regression trees with training and test datasets to provide an overall prediction using the average of their outcomes (Breiman, 2001, 2017). In this study, we used four RFR models to predict k and Y_0 for green and rooibos teas individually using NumPy, Pandas, and Scikit-learn in Python. For each of these four models, the data included 110 observations and 67 predictor variables (Supplementary Table S4). We split the observations into 75% for the training set and 25% for the testing set. Then, the set of RFR hyperparameters was tuned using Bayesian Optimization in the training phase. The Bayesian function uses the training set to find the best hyperparameters for the RFR model. We minimized the MSE in the objective function. The number of iterations was set to 100, and we used five-fold cross-validation to estimate RFR's performance.

3 Results

3.1 Soil microclimate and predictor variables

Compared to the past 50 years of precipitation at these sites in Iowa, 2018 was on the wetter end. For example, in 2018, precipitation ranged from 1,033 to 1,328 mm across all sites compared to the 50-year mean annual rainfall of 850–900 mm (+15 to +56%; Mesonet, 2024). Soil temperature and moisture varied over the length of the experiment (Supplementary Figure S2). Soil temperature ranged from 0.0 to 32.3 °C, with a low at the end of November and a peak in late June. The soil temperature mean \pm standard deviation was 20.4 \pm 5.3 °C. Soil volumetric water content ranged from 0.045 to 0.479 cm³ cm⁻³ with a low at the end of July and a peak in early July. The VWC mean \pm standard deviation was 0.261 \pm 0.090 cm³ cm⁻³.

Some soil properties varied greatly even though this study was conducted in a narrow geographic range (Table 1). There were some soil properties with CVs greater than 100% (e.g., ammonium, soil pH, and microbial biomass N), while other soil properties did not vary as much with CVs less than 25% (e.g., clay content, pH, STS, and STK). The treatment effects on select soil properties can be found in Middleton et al. (2021). Briefly, restored prairie had the greatest effect on MBC both in spring and autumn, increasing it by 116 and 104%, respectively, compared to annual cropping systems on average. Interestingly, adding more crop residue decreased PMC (-33%), POXC (-13%), and MBN (-23%) during the spring only, possibly because the increased crop residues cooled the soil. There were some other, smaller, inconsistent treatment effects on soil properties (Middleton et al., 2021).

3.2 Decomposition dynamics measured empirically and Tea Bag Index values

Decomposition rates varied across soils, but green tea decomposed much more quickly than rooibos tea and reached a more discernible plateau across all treatments during our 130-day study (Figure 1; Table 3). Green tea decomposed at a mean rate of 0.19% $\rm d^{-1}$ and reached Y_0 of 24%. Rooibos tea, on the other hand, decomposed at a mean rate of 0.028% $\rm d^{-1}$ and Y_0 of 53%. Green tea was better predicted by the three-parameter, exponential decay model (Equation 1) with a range in R^2 from 0.87 to 1.00 (mean R^2 = 0.96), which compares to rooibos tea that had an R^2 range from 0.63 to 0.99 (mean R^2 = 0.90; Table 3).

Due to interest in using the TBI as an easy and more accessible way to monitor management effects on decomposition, we correlated modeled coefficients (from Equation 1) to the $k_{\rm TBI}$ and $S_{\rm TBI}$ at 68 days (Figure 2). The $S_{\rm TBI}$ is supposed to predict the asymptote, or Y_0 , of the green tea and did correlate (r=0.65), but there was a systematic bias of -13% with measured Y_0 (Figure 2A, -56% relative bias). The $k_{\rm TBI}$ is supposed to predict the decomposition rate of the rooibos tea. This correlation with rooibos k was closer to 1:1, albeit weaker than $S_{\rm TBI}$, but smaller relative bias (Figure 2B, 25% relative bias).

TABLE 2 Description of long-term agricultural experiments in Iowa, USA, used for management intervention contrasts.

Management intervention	Experiment(s) name	Control treatment	Experimental treatment	Pertinent details	Reference(s) for more experimental information	Paired comparisons (n)	Years since established
+Biochar	Fields 70/71	No biochar	18.4 tons biochar per ha	All in continuous maize (across residue removal rates). Located near Boone, IA, USA.	Rogovska et al., 2016	8	11
+Cover crop	Comparison of Biofuel Systems (COBS)	Continuous maize	Continuous maize + cereal rye winter cover crop	All no-tillage. Located near Boone, IA, USA.	Nichols et al., 2014	4	17
	Agricultural Drainage Water Quality-Research and Demonstration Site (ADWQ-RDS)	Maize-soybean	Maize-Soybean + cereal rye winter cover crop	Combination of tilled $(n = 4)$ and no-tilled plots $(n = 4)$. Located near Gilmore City, IA, USA.	Qi et al., 2011	8	14
+Nitrogen	COBS	No N fertilizer	107 and 192 kg N ha ⁻¹ y ⁻¹	Both continuous maize and prairie treatments were included. Located near Slater, IA, USA.	Nichols et al., 2014	8	10
	Long-term Assessment of Miscanthus Productivity and Sustainability (LAMPS)	No N fertilizer	224 kg N ha ⁻¹ y ⁻¹	Both continuous maize and <i>miscanthus</i> × <i>giganteus</i> are included. Located near Boone, IA, USA.	Tejera et al., 2019	8	3
+Residue	Fields 70/71	100% aboveground residue removal	All aboveground residue remains	Continuous maize (across two biochar rates).	Rogovska et al., 2016	8	11
Diversified rotation	Neely-Kinyon	Maize-soybean rotation with synthetic fertilizer	Alfalfa and small grains integrated in rotation, fertilized primarily with manure	Synthetic N added as UAN (or 32%). Animal manure, either chicken or composted cattle manure. Located near Greenfield, IA, USA.	Delate and Cambardella, 2004	4	16
	Marsden Agricultural Diversification Experiment	Maize-soybean rotation with synthetic fertilizer	Alfalfa and small grains integrated in rotation, fertilized primarily with manure	Synthetic N added as UAN (or 32%). Animal manure added as composted cattle manure. Located near Boone, IA, USA.	Liebman et al., 2008	4	20
No-tillage	Long-term Tillage Trials	Chisel plow tillage	No-tillage	Maize-soybean rotation. Located near Boone, IA, USA.	Al-Kaisi et al., 2015	4	16
	ADWQ-RDS	Chisel plow tillage	No-tillage	Maize-soybean rotation. A combination of two treatments, one with and one without a rye cover crop. Located near Gilmore City, IA, USA.	Qi et al., 2011	8	14
Perennial cropping	LAMPS	Annual crop (i.e., maize)	Perennial biomass crop	Miscanthus x giganteus was established in 2015.	Tejera et al., 2019	8	4
Cropped prairie	COBS	Maize-soybean rotation	Restored prairie with 26 native species seed mix	Prairie stands are harvested for biomass yearly.	Nichols et al., 2014	4	10

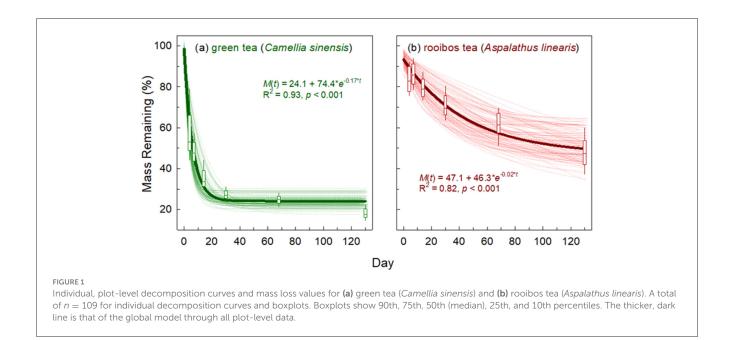


TABLE 3 Summary statistics of green and rooibos tea decomposition.

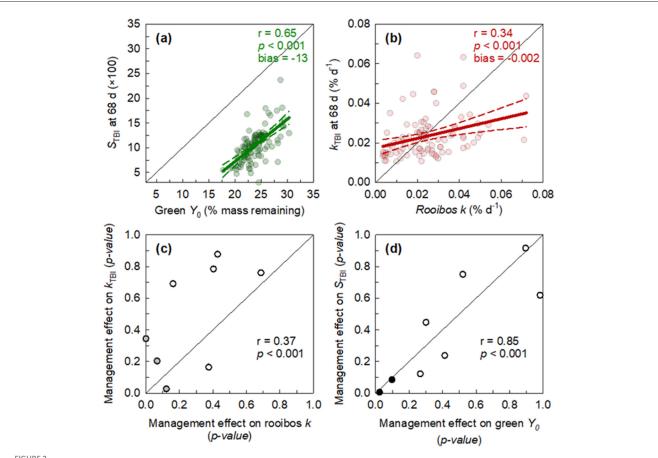
Three-parameter, exponential decay metric	Minimum	25th quantile	Median	75th quantile	Maximum	Mean	Standard deviation	
Green Tea (Camellia sinensis)								
Coefficient of determination or R ² (unitless)	0.87	0.96	0.97	0.98	1.00	0.96	0.02	
Final mass remaining or Y_0 (% mass remaining)	17.7	22.3	23.8	25.3	30.4	23.9	2.5	
Total mass loss from $t = 0$ or a (% mass loss)	68.7	73.8	75.1	76.7	80.9	75.1	2.6	
Decomposition rate or k (% mass loss d^{-1})	0.072	0.137	0.201	0.243	0.309	0.193	0.064	
Rooibos Tea (Aspalathus linearis)								
Coefficient of determination or R ² (unitless)	0.63	0.87	0.91	0.95	0.99	0.90	0.06	
Final mass remaining or Y ₀ (% mass remaining)	23.3	40.9	47.2	53.8	64.3	47.0	10.0	
Total mass loss from $t = 0$ or a (% mass loss)	32.7	41.5	47.4	55.5	97.2	53.4	18.1	
Decomposition rate or k (% mass loss d^{-1})	0.004	0.015	0.024	0.032	0.130	0.028	0.023	

Management had little effect on individual treatment contrasts at an $\alpha < 0.1$ (Figure 3; Supplementary Table S5). There were some exceptions for particular management practices and decomposition metrics. Adding synthetic N fertilizer, for example, slightly increased green Y_0 by 7% compared to conventional tilled soils (p < 0.001). Adding perennial plants—like perennial cropping and restoring prairie—strongly altered decomposition dynamics (Figure 3; Supplementary Table S5). Perennial cropping with *miscanthus* × *giganteus* both lowered green and rooibos k (-24 and -27%) and also increased green Y_0 (+11%) compared to annual crops (p < 0.067). Restoring native prairie both increased green k (+31%) and decreased rooibos Y_0 (-19%) compared to annual cropping (p < 0.071). By far, the strongest treatment effect was no-tillage (based on magnitude), which increased rooibos k by 75% compared to conventional tilled soils (p < 0.001).

To further examine the utility of the TBI, from the aspect of detecting treatment effects, we compared the management effect on empirically derived decomposition metrics (k and Y_0) with those derived from TBI ($k_{\rm TBI}$ and $S_{\rm TBI}$; Figures 2C, D; Supplementary Table S6). The one-way ANOVA p-values from both methods correlated relatively well (r>0.37), especially between the green Y_0 and $S_{\rm TBI}$ (r=0.85; Figure 2D). However, there was some inconsistency on which management intervention had significant treatment effects on both empirically and TBI-derived metrics (Supplementary Tables S5, S6).

3.3 Predicting tea decomposition with random forest regression

Using RFR, we were able to predict decomposition metrics moderately well (Supplementary Table S7). The lower root mean squared error (RMSE) for green tea indicates that, on the whole, it was better predicted with our 67 variables than rooibos, both



Correlation between 3-parameter, exponential decay model constants and Tea Bag Index (TBI) metrics from one timepoint (68 d). (a) Green tea final mass remaining (Y_0) correlated with Tea Bag Index stabilization factor (S_{TBi}) multiplied by 100. (b) Rooibos tea decomposition rates (k) correlated with that estimated from TBI (K_{TBV}). Correlation of p-values from one-way ANOVAs across management practices between (c) K_{TBi} and k and (d) Y_0 and S_{TBi} (see Table 2 for management interventions). Data points are color coded where white for both metrics are insignificant (p-value >0.1), gray when one of the two is significant, and black for when both are significant. Pearson correlation coefficients (r), p-value, and 1:1 line are shown.

in the training and test datasets. We chose to highlight the top 10 predictor variables for both teas' k and Y_0 (Figure 4). Generally speaking, for the dynamic soil properties, more autumn measurements ended up on the top 10 list compared to spring. Temperature was a strong predictor of green tea decomposition, but barely made the top 10 list for rooibos tea. Maize yields showed up on all four variables' top 10 lists—indicating the factors regulating maize yield may also regulate decomposition. Particle size features—clay, fine clay (FClay), particle size kurtosis (PS_{kurt}), mean particle size (PS_{mean}), particle size mode (PS_{mode}), sand—were much more important in predicting rooibos tea decomposition compared to green tea.

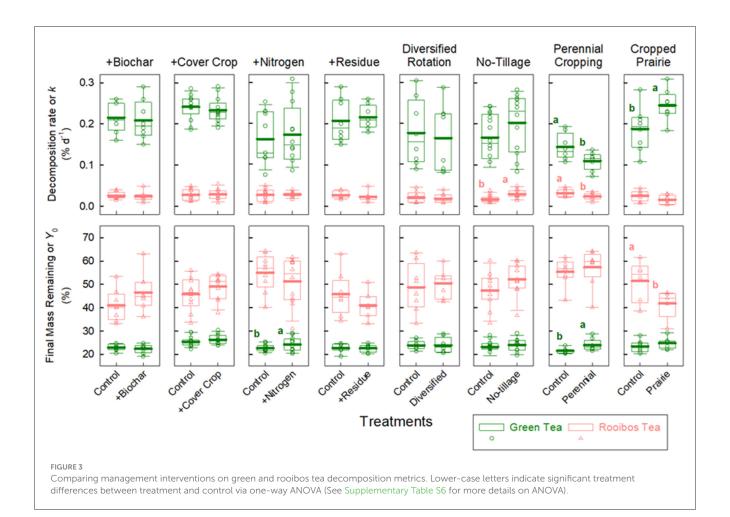
We explored the top five predictors in further depth with partial dependency plots to evaluate their relationship with decomposition metrics (Figure 5). Soil temperature was the first and strongest predictor of green k and Y_0 , with a positive relationship between the variables. In other words, greater temperatures increase both the decomposition rate and the amount remaining at 130 days. Maize yield also positively correlated with green k and Y_0 , and was the second and fourth predictors, respectively. Interestingly, the combined index for soil health principles (SHPS) was the second-best predictor of green Y_0 , with a positive relationship, indicating

that the more soil health principles included in the management, the greater the mass remaining of the rapidly decomposing tea.

Rooibos tea decomposition was strongly predicted by particle size, but some other interesting variables emerged from the RFR and partial dependence plot (Figure 5). Microbial biomass N, collected in the autumn, was the first predictor of rooibos k, indicating that more microbial biomass predicts greater rooibos tea decomposition. Smaller particle sizes tended to predict decreased rooibos k, and vice versa for large particle sizes and mean particle size in general. Fine clay was the first predictor of rooibos Y_0 , with negative correlation or dependence, indicating that an increase in fine clays decreases the final % mass remaining within the timeframe of our study (<1 year).

4 Discussion

We used a variety of agroecosystem management practices, while controlling for climate, to elicit localized effects on decomposition of common substrates (well-characterized teas in this case). The accessibility of these common substrates can also facilitate citizen scientists' engagement in monitoring changes in



decomposition that may occur after a shift in agroecosystem management. This engagement by farmers, as citizen scientists, can help bolster a soil health movement intent on managing agroecosystems to maximize multiple ecosystem services (Cooper et al., 2007; Grudens-Schuck and Sirajuddin, 2019; Karlen et al., 2021). The following sub-sections will answer our three research questions.

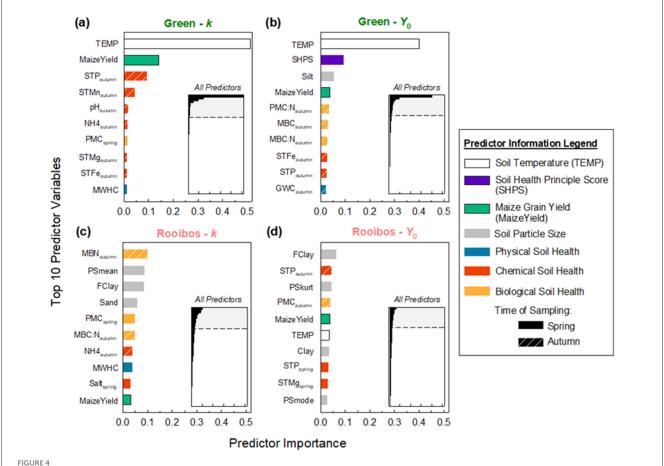
4.1 Does the Tea Bag Index correlate with empirically measured decomposition metrics?

The TBI is a tool for community scientists to derive an index of decomposition by measuring mass loss at just one time point (Keuskamp et al., 2013). It requires using both a high-and low-quality substrate, green and rooibos teas, respectively, and has been used in dozens of studies (Petraglia et al., 2019; Duddigan et al., 2020a; Dossou-Yovo et al., 2021; Pino et al., 2021; Mori, 2022a). Using the original nylon mesh Lipton® tea bags, we found good evidence that the $k_{\rm TBI}$ and $S_{\rm TBI}$ do correlate well with three out of four empirically measured decomposition metrics with six time points (Figure 2). The correlations were stronger with green tea, but the decomposition rates were closer to the actual value for rooibos tea (Figure 2B). Furthermore, and

maybe most importantly for community science monitoring of soil biological activity after management change, the management practices we tested had similar effects on both empirically measured and TBI-estimated decomposition metrics (Figure 2; Supplementary Tables S6, S7).

The TBI approach, while accounting for substrate quality by using two substrates pre-packaged in Lipton tea bags, provides an index of decomposition rate and stabilization that can be easily measured at global scales (Keuskamp et al., 2013; Sarneel et al., 2024). However, unfortunately, in 2016, Lipton changed its tea bags from a 0.25-mm nylon mesh to a non-uniform <0.2 mm polypropylene mesh that is also more decomposable. This change has hindered the use of the TBI approach, as the new "non-woven" tea bag materials are not as feasible for calculating $k_{\rm TBI}$ or $S_{\rm TBI}$ (Mori, 2022a). There are alternative tea brands, however, that still use uniform nylon mesh tea bags that could be used by citizen scientists. These new teas need to be rigorously tested as the original green and red Lipton teas. This raises the question of whether a TBI is necessary for assessing agroecosystem management in terms of decomposition.

In addition to challenges getting the woven mesh, there has been recent criticism of the TBI method and assumptions (Mori, 2022b, 2025). Future studies should focus on robust measurements of decomposition that are still easy and accessible, allowing engagement by citizen scientists. For example, researchers recently used a simple decomposition ratio that was sensitive to



Random forest regression predictor importance for variables regulating green and rooibos tea decomposition. Top 10 predictors and their relative importance are shown for: (a) green tea decomposition rate (k), (b) green tea final, modeled mass remaining (Y_0), (c) rooibos tea k, and (d) rooibos tea Y_0 . Inset graphs show distribution of top 10 (shaded region) relative importance within entire 67 predictors.

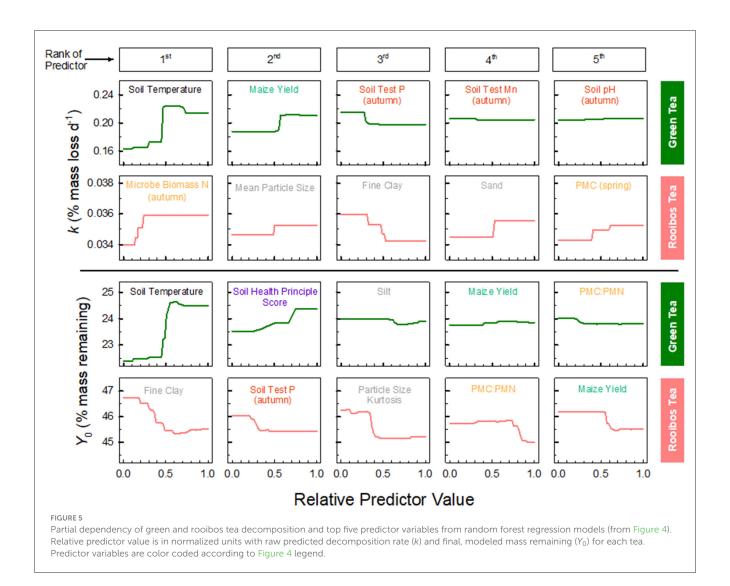
management practices in Canada, similar to the TBI metrics (Hayes et al., 2024). Alternatively, citizen scientists could simply monitor management effects on mass loss after a given duration of any, or a few, common pre-packaged substrates (i.e., teas, tongue depressors, or cotton strips). The "soil your undies" campaign, which involves decomposing bleached cotton underwear, is a good example and has the added benefit of being humorous.

4.2 Do soil health-promoting practices consistently alter decomposition kinetics?

While not consistent across management practices, there was some evidence for management altering decomposition (Figure 3). Duddigan et al. (2020a), using the same TBI approach with 511 samples within the UK, found that adding soil amendments greatly increased $k_{\rm TBI}$, so much so that it overrode any geographical (i.e., soil and climate) effects. We did not find as strong effects of management on decomposition, even with our soil amendment practices (+Biochar and +Nitrogen). However, some salient management effects on decomposition were apparent in our study.

First, adding N fertilizer increased green Y_0 by 7% (Figure 3), or decreased green tea mass loss. Because green tea has greater

N content and a narrow C:N of ~13 (Supplementary Table S2), it is a rapidly available net source of N for soil microbes. One might reasonably expect this finding, and that adding supplemental fertilizer N could alleviate N demand and thus slow decomposition of this high-quality residue. But this generalization is complicated by previous findings. A meta-analysis on mostly forest litter showed that the effect of supplemental N on decomposition depended on both the rate and litter quality (Knorr et al., 2005). Overall, according to this meta-analysis, adding N can stimulate decomposition of high-quality residues but inhibit decomposition of low-quality residues (a finding opposite to ours). However, when the meta-analysis results were split into three N fertilizer rate categories (<75, 75-125, and >125), the low and high N rates (<75, >125) had inhibitory effects on decomposition regardless of litter quality. More than 50% of our research plots received N rates in these two categories, but even those plots that received some in the intermediate category were close to these somewhat arbitrary cut-offs. In agroecosystems, researchers have also found mixed results with fertilizer N inputs inhibiting decomposition (Le Guillou et al., 2011), some finding no effect (Grandy et al., 2013), and others finding that both management and residue quality are key determinants of regulating decomposition (McDaniel et al., 2014). These studies collectively illustrate that the rate and source



of N fertilizer, the quality of residue, and other management factors will all regulate whether and to what extent decomposition is affected by N fertilizer.

Second, we found that no-tillage increased rooibos k by 75% compared to conventional tillage, and this was the largest magnitude of a management effect that we observed (Figure 3). This was also the only management practice that confirmed our hypothesis that soil health-promoting practices would increase the decomposition of poorer quality litter (i.e., rooibos tea). Although no-tillage has been shown to alter soil microclimatic conditions, namely increasing soil moisture and decreasing temperature (Licht and Al-Kaisi, 2005; Potter et al., 1985), it is not likely a factor explaining increased decomposition since neither microclimatic variable emerged to predict rooibos decomposition (Figure 4). Instead, other mechanisms may explain the strong effect of increasing the decomposition rate of this relatively low-quality tea.

Perhaps the most parsimonious explanation is the positive effects of no-tillage on soil microbiota, as documented in a meta-analysis (Zuber and Villamil, 2016), and their indirect effect on the decomposition of more-difficult-to-decompose substrates. In other words, enhanced microbial biomass and activity in the no-tilled

soils increased rooibos k. To add additional evidence for this hypothesis, microbial biomass N was the best predictor of rooibos k (Figure 5). However, the direct effects of no-tillage on either soil microbial biomass or potential mineralization were not as strong. We found that no-tillage generally increases these measures of soil biological activity, but the difference is not statistically significant (Middleton et al., 2021). It could be that one-way ANOVAs are a blunt tool that is not as sensitive to measuring decomposition and using RFR to tease apart treatment effects.

Third, and finally, perennializing agroecosystems altered the decomposition of both substrates in interesting and countervailing ways. Using perennial crops, for example, $miscanthus \times giganteus$, for just 3 years decreased green and rooibos k, and also increased green Y_0 (Figure 3). In contrast, 10 years of cropping native prairie biomass had opposite effects on the teas—increasing green k but decreasing rooibos Y_0 . Both perennial cropping and restored prairie treatments have year-round cover; however, we lack a clear explanation for the interesting, unexpected, and opposing effects observed between the two practices. Perhaps it may be due to differences in the effects of these perennial plantings on either soil microclimate via plant—water demand, C, and nutrient cycling, or

both (Daigh et al., 2014; Ye and Hall, 2020; Studt et al., 2021). These divergent effects on decomposition may be ultimately due to the inherent differences in rooting structure, morphology, phenology, and even diversity under miscanthus and prairie.

A mature stand of miscanthus can decrease soil temperatures by 16% on average during the growing season, but has a variable impact on water content, sometimes increasing and decreasing depending on the time of year (Studt et al., 2021). The decrease in both tea's decomposition rate, and perhaps an increase in green Y_0 , is likely due to the perennial crops' effect on soil microclimate. However, miscanthus does also alter other soil properties that may further explain our findings—like increasing proportion of amino compounds (Khaleel, 2023), increasing water holding capacity (Studt, 2019), and generally improving efficiency of N cycling compared to annual crops (Davis et al., 2013; Smith et al., 2013; Studt et al., 2021). Native, perennial prairie, in contrast, increased green k and decreased Y_0 . Previous studies from this site show that restored prairie increased soil microbial biomass and altered extracellular enzyme activities depending on the N fertilizer rate (Bach and Hofmockel, 2015; Middleton et al., 2021).

4.3 What are the overarching regulators, at a local scale, on decomposition of highand low-quality residue?

By isolating local factors from broader factors, such as climate and litter quality, we can improve our mechanistic understanding of what regulates decomposition. While we showed that some management practices affect decomposition, it does not provide mechanistic explanations to the previous question—do soil health-promoting practices consistently alter decomposition kinetics? This is why we chose to also use RFR with our large data set to better tease apart mechanisms within and across management practices in this relatively localized network of long-term experiments in Iowa, USA.

Despite this study's local approach, we still found soil temperature, or at least the cumulative thermal units, to be the primary driver in green tea decomposition (Figure 4). The magnitude of this first predictor's importance was 3.6× and 4.3× greater than that of the second-best predictors for green k and Y_0 , respectively; and greater soil temperatures increased both green k and Y_0 (Figure 5). This upholds the primacy of temperature in driving decomposition rate, at least for this highquality, narrow-C:N litter. We think there are local, unaccountedfor factors affecting soil temperature that are not entirely explained by our one-way ANOVA or RF approaches. This could range from something as subtle as farm-level nuances in residue management, differences in maize hybrids—varying in canopy closure speed, or even nearby tree lines acting as windbreaks. In contrast to strong control on green tea, temperature barely made an appearance in regulating rooibos tea decomposition (Figure 4). The reason soil moisture may not have played an important factor is that 2018 was one of the wetter years in recent Iowa history across all sites (Mesonet, 2024).

It is quite remarkable that soil particle size played such a strong role in rooibos tea decomposition. All particle size predictors

pointed to decreased k and Y_0 when rooibos tea is buried in soils with finer particles (Figure 5). Or in other words, more clayey soils tend to decrease the rate but increase the extent of rooibos tea decomposition. This contrasts with results from a recent multi-site investigation of cover crop residue decomposition in agroecosystems, where researchers found that finer textured soils increased k (Thapa et al., 2022). The authors attributed this effect to finer textured soils being wetter and accelerating cover crop residue decomposition in these potentially water-limited siteyears. Our substrates were decomposed while buried in heavier textured soil (mean clay content 28.8% vs. 16.5%), instead of litter bags placed on the soil surface, and decomposing in a less waterlimited year for Iowa. Supporting these differences, an incubation experiment tested this question of buried vs. incorporated residues, and showed diverging trends in decomposition between surface and incorporated residues (Scott et al., 1996).

This intriguing finding of smaller particle sizes slowing the rate, but increasing the extent, of rooibos tea decomposition may be indicative of the widely accepted trend of finer textured soils having greater SOM (Burke et al., 1989; Plante et al., 2006; Grandy et al., 2009). Soils with smaller particles (i.e., more reactive surfaces) will have greater contact with low-quality residue, providing more reactive surfaces for efficient decomposition and greater SOM stabilization capacity (Kleber et al., 2007; Poeplau et al., 2015). It has also been shown that soil texture relates to SOM quality, and that finer textured soils have a greater relative abundance of N-bearing compounds (Grandy et al., 2009) and "biochemically protected" C (Plante et al., 2006). Our results demonstrate the value in using a large, local decomposition study—combined with RFR—to connect decomposition (early stages of SOM formation) to important soil processes like long-term C accrual.

Soil health-promoting practices regulate decomposition in both direct and indirect ways. The second best predictor of green Y_0 , second only to soil temperature, was the SHPS derived from the NRCS principles of soil health (USDA-NRCS, 2023; McDaniel and Middleton, 2024). The greater adoption of principles like reducing disturbance (e.g., no-till), increasing cover (e.g., residue), diversifying (e.g., manure input and extended rotations), and extending the length of plant from annual crops (e.g., perennial crops or prairie) increased green Y₀. This also highlights and connects our findings with applications for citizen science in agroecosystems. If, for example, growers wanted to track soil improvements from adopting soil health-promoting practices, then they may want to monitor the final mass of green tea. Due to Y_0 and S_{TBI} being so strongly correlated, as well as the treatment effect measured empirically and with S_{TBI} (Figure 2), it may be sufficient to use TBI to monitor changes in soil health. Thus, the TBI appears to be an inexpensive approach to monitor changes in soil health (Figure 3), further inform growers of changes in their soil, and expand the soil health movement.

5 Conclusion

We used multiple long-term experiments within relatively narrow environmental conditions to tease apart more local regulators driving decomposition of common substrates. Our use of "pre-packaged," common substrates (i.e., manufactured nylon

tea bags) made monitoring decomposition more accessible in these highly managed agroecosystems, and we were able to confirm the scientific robustness of the Tea Bag Index approach for monitoring changes in management practices. This is good news for farmers who want to measure changes in soil biological activity due to a management intervention.

From a basic science perspective, we found that management practices do affect decomposition and not always in explainable or predictable ways. While the decomposition paradigm suggests that climate and residue quality are the ultimate regulators of decomposition, we show that more local factors, such as soil particle size, microbial biomass nitrogen, crop yield, bioavailable nutrients, and management practices, can also predict decomposition. These findings expand our knowledge of the regulators of decomposition, especially in agroecosystems, and are critical to improving our basic understanding of decomposition. Measuring and modeling decomposition can help inform ways to manage agroecosystems to be more environmentally sustainable and climate-smart.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation. The data are also available at Iowa State University DataShare or at https://doi.org/10.25380/iastate.30104761.

Author contributions

MM: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Visualization, Writing – original draft, Writing – review & editing. PM: Conceptualization, Methodology, Software, Validation, Writing – original draft, Writing – review & editing. GH: Conceptualization, Investigation, Methodology, Project administration, Supervision, Writing – review & editing. TM: Data curation, Formal analysis, Investigation, Methodology, Resources, Validation, Writing – original draft, Writing – review & editing.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsufs.2025. 1665233/full#supplementary-material

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