European Journal of Agronomy 157 (2024) 127194

Contents lists available at ScienceDirect

European Journal of Agronomy

journal homepage: www.elsevier.com/locate/eja

# Shifting focus from external to in situ organic resources – The redesign of four tropical long-term experiments

Moritz Laub [a,*], Marc Corbeels [b,c,d], Samuel Mathu Ndungu [c], Monicah Wanjiku Mucheru-Muna [e], Daniel Mugendi [f], Rebecca Yegon [f], Wycliffe Waswa [c], Bernard Vanlauwe [c], Johan Six [a]

[a] Department of Environmental Systems Science, ETH Zurich (Swiss Federal Institute of Technology), Universitätstrasse 2, Zürich 8092, Switzerland
[b] CIRAD, UPR AIDA, Avenue d'Agropolis, Montpellier F-34398, France
[c] AIDA, CIRAD, Université Montpellier, Montpellier, France
[d] International Institute of Tropical Agriculture (IITA), c/o ICIPE Compound, P. O. Box 30772-00100, Nairobi, Kenya
[e] Department of Environmental Sciences and Education, Kenyatta University, P.O. Box 43844-00100, Nairobi, Kenya
[f] Department of Water and Agricultural Resource Management, University of Embu, P.O. Box 6-60100, Embu, Kenya

## ARTICLE INFO

*Keywords:*
Soil organic matter
Intercropping
Legumes
Maize
Regenerative agriculture
MBILI

## ABSTRACT

Long-term experiments (LTEs) are critical for evaluating strategies that can maintain or increase crop yields, soil fertility and soil organic carbon (SOC), and help adapt to climate change. Yet, scientific knowledge is advancing and research questions are evolving. Therefore, it is important to review the objectives of LTEs over time. A change in their design may be necessary to keep the experimental treatments scientifically interesting, innovative, and relevant in the context of evolving agricultural challenges. Here, we describe the process of redesigning four LTEs in Kenya. These LTEs are unique in that they represent four different pedoclimatic conditions but with identical experimental treatments across sites. Initially, they focused on investigating how to maintain or increase SOC and maize yields over time by applying a combination of mineral nitrogen (N) and external organic resources. Specifically, the experimental treatments consisted of maize monoculture with different rates (1.2 and 4 t C $ha^{-1}$ $yr^{-1}$) and qualities of organic resources, either with or without mineral N fertilizer input. After about 20 years, it became clear that SOC was lost in most treatments. Therefore, continuing with the current experimental design was not an option. Taking advantage of the fact that the different former treatments led to different levels of soil degradation, we redesigned the LTEs to study the effectiveness of regenerative cropping strategies in rebuilding SOC and increasing crop yields starting from the different levels of soil degradation. The focus shifted from external to in situ organic inputs by increasing the root biomass of the cultivated crops. The newly established cropping system treatments are maize-legume rotation, maize-legume intercropping (double row configuration) and relay intercropping of maize with forage grass. A key finding from the previous phase of the experiments, namely, that external organic inputs with low C:N ratios are most efficient in building SOC, has been incorporated into the redesign. The relative contribution of external versus in situ organic resources is tested by splitting the cropping system treatments into those receiving either farmyard manure or green manure in the form of *Tithonia diversifolia* prunings and those receiving no external inputs. Split-plot treatments with and without mineral N were retained. The overall objective of studying mechanisms of tropical soil fertility maintenance and, more specifically, SOC formation, remained unchanged. However, the redesign aligned the LTEs with the current state of knowledge and pressing research questions, specifically focusing on the relative effectiveness of in-situ versus external organic inputs in SOC formation.

## 1. Introduction

Initiatives such as “4 per 1000” - Soils for Food Security and Climate (Minasny et al., 2017; Rumpel et al., 2020), and attempts to compensate farmers for sequestering carbon (C) in their soils (Popkin, 2023), have recently brought attention to the prospect of increasing soil organic carbon (SOC) stocks for climate change mitigation and adaptation. Soil C sequestration, as a strategy to mitigate climate change, can be

---

* Corresponding author.
*E-mail address:* moritz.laub@usys.ethz.ch (M. Laub).

https://doi.org/10.1016/j.eja.2024.127194
Received 19 December 2023; Received in revised form 26 March 2024; Accepted 22 April 2024

achieved through various practices such as conservation agriculture, agroforestry (Corbeels et al., 2019), natural revegetation (Gvein et al., 2023), or afforestation (Beillouin et al., 2023). However, measuring soil C sequestration in the field is notoriously difficult, especially in the short term. The reasons for this are the natural variability of soil properties (Paul et al., 2023) and the typically gradual nature of changes in SOC, which often span decades. Long-term experiments (LTEs) can help to assess how much C can be sequestered by different crop management practices and, more importantly, compare the potential of these practices against each other. Despite the recent focus on increasing SOC for climate mitigation purposes, maintaining SOC and, more generally, soil fertility, has long been at the core of sustainable management of arable land (Bashir et al., 2021; Janzen et al., 2021; Lal, 2004). Long-term field experiments, such as the Broadbalk experiment at Rothamsted, established in 1843 in the UK (Watts et al., 2006), the Frame Trial in Ultuna, established in 1956 in Sweden (Kätterer et al., 2011), and the DOK experiment in Therwil, established in 1978 in Switzerland (Fließbach et al., 2007), continue to provide valuable insights into the gradual soil processes that affect crop yield. They further play a crucial role in providing datasets to evaluate and further develop agroecosystem models (e.g., Bruun and Jensen, 2002; Laub et al., 2020; Couëdel et al., 2024; Necpalova et al., 2018).

However, scientific understanding has advanced since the start of these experiments. For example, the theory that SOC mainly consists of humic substances, i.e., abiotic condensations of what remains after fresh plant litter input is decomposed (Frimmel and Christman, 1988; Woomer and Swift, 1994), has been replaced by a newer theory. This theory states that most SOC originates from microbial necromass, derived from several rounds of C and nutrient cycling in soil (Cotrufo et al., 2013; Denef et al., 2009; Kallenbach et al., 2016). Consequently, periodic review and adjustment of treatments in LTEs may be necessary to ensure their continued relevance to the contemporary scientific discourse. The process of redesigning and translating lessons from the previous to the new design is valuable knowledge for the LTE community and to LTE managers. However, because of the partially informal nature of such information, it may often remain internal knowledge, accessible only to those who are involved in the experiment. This article describes the process of reviewing and redesigning four unique LTEs with identical experimental treatments in contrasting pedoclimatic conditions in Kenya, in order to share this knowledge. The most critical points considered in the process are also highlighted.

## 2. The four long-term experiments in Kenya: setup and most important results

The four experiments were initially established as double-season maize monocropping experiments at four locations characterized by different pedoclimatic conditions. The experimental sites in western Kenya, Aludeka and Sidada, were established in 2005. They represent soils with low (13 %) and high (56 %) clay content in a humid climate with an annual rainfall of about 1700 mm and an average annual temperature of about 24 °C. In central Kenya, the experimental site at Embu is characterized by a drier climate with clayey soil (∼ 1200 mm, ∼ 20 °C, 60 % clay). Meanwhile, the experimental site at Machanga represents a less suitable climate for maize production with a sandy soil (∼ 800 mm, ∼ 24 °C, 13 % clay). Both experiments in central Kenya were established in 2002. The experiments aimed to study the effect of different quantities and qualities of organic inputs (C:N ratios, lignin and polyphenol contents (L&PP)), in combination with and without mineral nitrogen (N) fertilizer, on the evolution of SOC, soil fertility, and maize yields. The organic resources included the full range of qualities that are specified in the decision support tool of the organic resource database (Palm et al., 2001a, 2001b). The different sites were selected to study how climate and soil properties influenced the performance of organic resource and mineral fertilizer additions.

Details on the setup of the field experiments can be found in two recent publications (Laub et al., 2023a, 2023b). In summary, the experiments consisted of 22 split plots (11 main treatments ±N) with three replicates. The main treatment involved the addition of five types of organic resources in quantities of either 1.2 or 4 t C ha$^{-1}$ yr$^{-1}$. The subplot treatment consisted of the application of 120 kg mineral N ha$^{-1}$ season$^{-1}$ (+ N) compared to a treatment without mineral N input (-N). The main plot size was either 12 × 5 m (Embu) or 12 × 6 m (other sites), with the split plot being half of that. Organic resources were applied once a year during the long rainy season, just before maize sowing. They were incorporated to a soil depth of about 15 cm using hand hoes. The organic resources were selected to include the four quality classes that had been defined based on differences in L&PP and C:N around the time of the start of the experiments (Palm et al., 2001a). Pruned leaves from *Tithonia diversifolia,* including stems less than 2 cm in thickness, were classified as a high-quality organic material with a rapid decomposition rate (low C:N, low in L&PP; class 1). Pruned leaves including small stems from *Calliandra calothyrsus* were classified as high- quality material with an intermediate decomposition rate (low C:N, medium L&PP; class 2). Maize stover was classified as low-quality material with a slow decomposition rate (high C:N, low in L&PP; class 3) and sawdust from *Grevillea robusta* trees as low-quality material with a very slow decomposition rate (high C:N, high in L&PP; class 4). Locally available farmyard manure (low C:N, medium L&PP) has an intermediate to high quality with an intermediate decomposition rate (Sileshi et al., 2019), but it does not have a defined class. The control treatment did not receive any organic resource additions. During harvest, all aboveground maize biomass was removed from the experimental plots. Therefore, the plots only received C inputs from the roots in addition to the external organic inputs from the experimental treatments. The decision to combine organic resource treatments with + N and -N treatments was based on the decision support tool. The tool suggested that pruning material from *Tithonia* with low C:N could be directly applied, while *Calliandra,* maize stover, and sawdust would have to be mixed with mineral N fertilizer (Palm et al., 2001a). The decision to use sawdust as a treatment, despite its uncommon usage among farmers, was based on the prevailing theory at the time of the experiments` establishment. This theory hypothesized that SOC is primarily composed of aromatic compounds that remain after the decomposition of added organic resources. Organic resources rich in lignin and polyphenols were thus thought to be the most efficient in forming SOC (Palm et al., 2001a; Woomer and Swift, 1994). Recent results, including those from our (Laub et al., 2023b) and similar (Puttaso et al., 2013) experiments, have since refuted this theory. Instead, they support the theory that most SOC comes from microbial turnover and is formed most efficiently from organic inputs with low C:N ratio, which microbes can process efficiently (Cotrufo et al., 2013; Denef et al., 2009). In fact, one of the main findings from our field experiments was that inputs of farmyard manure (and to a lesser extent of *Calliandra* and *Tithonia* prunings) were the most effective in maintaining SOC. This indicates that external organic resources with a low C:N ratio were converted into SOC most efficiently (Fig. 1). Furthermore, it was observed that treatments with organic resources of low C:N, such as *Tithonia* prunings and farmyard manure, were the most effective in improving long-term maize yields at the two sites where yields were sustained or increased (Sidada and Aludeka; Fig. 2).

Contrary to the initial research hypothesis of the experiment, the addition of external organic resources did not maintain SOC levels at three out of the four sites, even with the application of 4 t C ha$^{-1}$ yr$^{-1}$ of farmyard manure (Fig. 1). This was mainly due to the high turnover of organic materials and SOC, as well as the fact that the sites had been under cultivation for only a few decades (Laub et al., 2023b). Additionally, substantial soil erosion was observed in Machanga, although it was not quantified. Also, despite significantly higher maize yields and biomass productivity in the treatments that received mineral N fertilizer, there was no overall significant positive effect of mineral N fertilizer on SOC. In combination, these findings suggested that the contributions of maize root biomass to SOC were limited, and that external organic

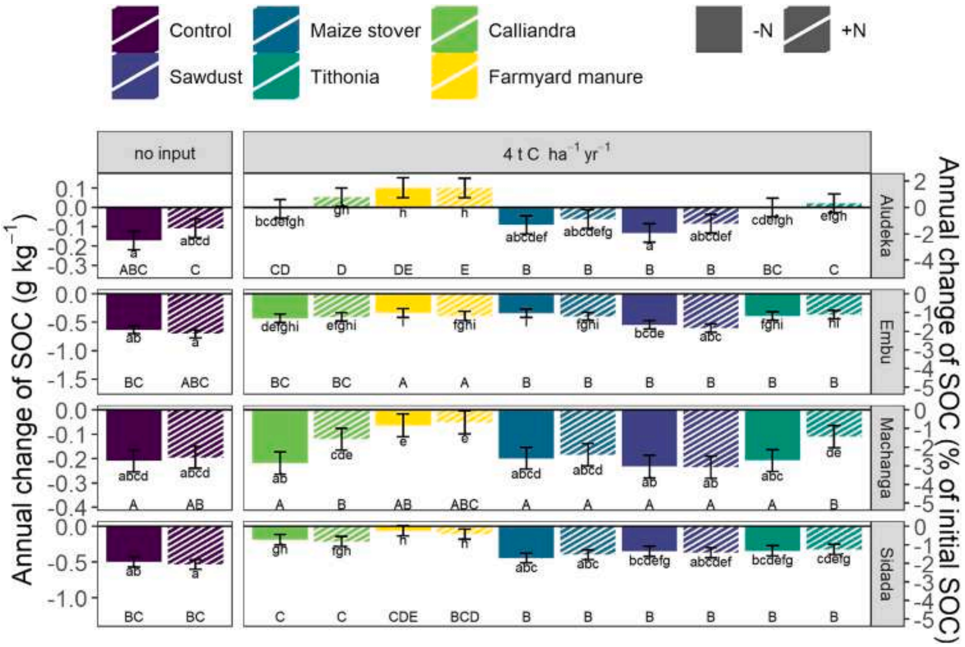

**Fig. 1.** Mean annual SOC changes in the four long-term experiments. At the Embu and Machanga sites (central Kenya), these were based on soil sampling campaigns conducted every two to three years; at the Sidada and Aludeka sites (western Kenya), sampling was only done in 2005, 2018, 2019, and 2021. The different experimental treatments led to significantly different levels of annual SOC depletion at the four sites. Over the duration of the experiments (20 years in central Kenya, 16 years in western Kenya), all sites lost SOC, except Aludeka. The farmyard manure treatment was the most effective in reducing SOC losses. Same lowercase letters indicate absence of a significant difference ($p < 0.05$) between treatments at the same site. Same uppercase letters indicate absence of a significant difference ($p < 0.05$) within the same organic resource treatment across sites. Error bars indicate the 95 % confidence intervals. Figure adapted from Laub et al. (2023b) under the Creative Common License 4 (https://creativecommons.org/licenses/by/4.0/).

resource inputs were mostly insufficient. In addition, the application of 4 t C ha$^{-1}$ yr$^{-1}$ of farmyard manure, which corresponds to about 16 t dry matter ha$^{-1}$ yr$^{-1}$, was deemed unrealistically high for local smallholder farmers.

Thus, while some experimental treatments increased maize yields, they were evidently not suitable crop management options for increasing SOC and promoting sustainable intensification. Recent studies have shown that plant roots can form SOC more efficiently than external organic inputs (Prescott et al., 2021; Sokol and Bradford, 2019). In line with our experimental results, we thus found that relying solely on external organic resources may not be the most effective method for increasing or maintaining SOC. As a result, we reevaluated and redesigned our experimental treatments.

## 3. Redesign process

After approximately two decades of conducting the experiments, it became evident that varying levels of degradation and loss of SOC occurred across the experimental treatments and sites (Fig. 1). This outcome provided a unique opportunity to shift our research focus towards studying whether new cropping strategies could regenerate SOC and improve crop yields, particularly when starting from different initial levels of soil degradation. The primary change was to decrease reliance on external organic resources and instead prioritize the generation of in-situ organic inputs by increasing root biomass through cultivated crops. However, important lessons from the previous experimental phase, such as the efficacy of the farmyard manure and *Tithonia diversifolia* treatments in maintaining SOC, as well as their synergistic effects with mineral N application for optimal maize yields, were used to determine which aspects to retain in the redesign. It was also considered essential to align the experimental treatments with the farming contexts of the local farmers. Therefore, we used a co-design process to redesign the experiments (Pohl et al., 2017). This process included obtaining extensive feedback from key stakeholders, such as local farmers and extension agents. The focus was on developing regenerative cropping strategies that are 'realistic', meaning they reflect cropping systems and crop management practices that are either already in use by farmers or that farmers find feasible to adopt. The goal was to ensure that the cropping strategies tested in the new experimental treatments are scalable and can be adopted by farmers with limited resources once proven valuable in the experiments. For the redesign, two workshops were held in January and September 2022, involving local and international collaborators from the International Institute of Tropical Agriculture (IITA), the "Centre de coopération Internationale en Recherche Agronomique pour le Développement" (CIRAD), Kenyatta University, the University of Embu, and ETH Zurich. In these workshops, we collectively evaluated the results from the previous experimental phase, identified the main critical issues, and then co-designed the new experimental treatments with the goal of regenerating SOC, increasing crop yield, and enhancing yield stability, particularly in the severely degraded treatments (see below).

The process of redesigning consisted of the following steps: i) grouping the previous experimental treatments to allow for the rerandomization of the new treatments; ii) co-designing new treatment options that are both suitable to test new state-of-the-art hypotheses on SOC formation and that are relevant for local smallholder farmers; iii) pre-testing the new treatments in plots adjacent to the main LTEs to verify their feasibility, and iv) implementing the new treatments.

In the first step, we grouped the previous treatments by their soil degradation level in 2021. This was done by ordering them based on the mean predicted SOC loss, which was calculated using the total time series of SOC measurements. We then assessed for significant differences in SOC losses between treatments (Laub et al., 2023b). Additionally, we examined pH and soil nutrient data, where available, to ensure maximal homogeneity in these parameters within each treatment group at each site. This grouping was crucial for the rerandomization of the new treatments among the previous ones. Rerandomization was necessary to statistically disentangle the effect of the old and the new treatments

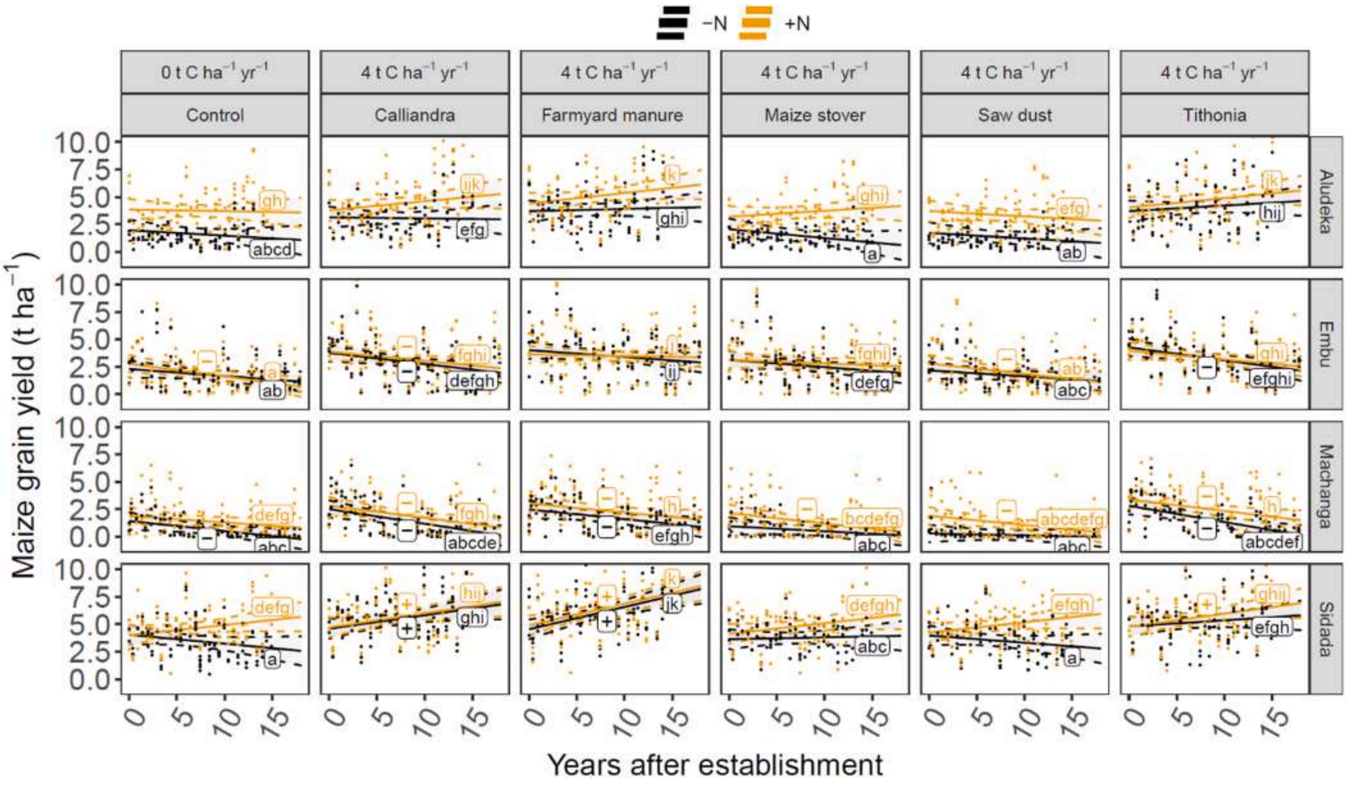

**Fig. 2.** Least square means of seasonal maize grain yields over time for different types of organic resources applied at 4 t C $\text{ha}^{-1}$ $\text{yr}^{-1}$ and with or without 120 kg N $\text{ha}^{-1}$ per season (+ N and -N) at four experimental sites in Kenya. Same lowercase letters within the same site indicate the absence of a significant difference between treatments at that site in year 16. Where either + or – are displayed at the regression center, the temporal trend is significantly larger or smaller than zero (all $p < 0.05$). The grey shaded areas constrained by the dashed lines indicate the 95 % confidence interval of the treatment mean for each site. Dots represent measured data (n = 3 per season and treatment). Figure adapted from Laub et al. (2023a) under the Creative Common License 4 (https://creativecommons.org/licenses/by/4.0/).

(Zhou et al., 2018) and to determine when a new treatment becomes the predominant factor affecting crop yield, biomass productivity, or SOC. This exercise identified four distinct levels of soil degradation (Table 1). The control, sawdust and maize stover treatments showed the most severe degradation, while the farmyard manure treatments exhibited the lowest soil degradation levels (Fig. 3). To ensure continued monitoring of soil degradation in the worst-case treatment with no inputs, the control treatments were maintained. Additionally, the C input rates (1.2 and 4 t C $\text{ha}^{-1}$ $\text{yr}^{1}$) from the previous farmyard manure treatments were kept as a form of positive control. The effects of the new cropping

**Table 1**
New experimental treatments for SOC regeneration, designed in the workshops, and the former treatments they are to be established in, grouped according to their soil degradation level.

| Previous SOC treatments | New treatments (from 2023 onwards) | TRT No | Degradation (site mean) | Mean SOC loss after 16–20 years (% of initial SOC) | (95 % confidence interval) |
|---|---|---|---|---|---|
| Control (± N) | Maize/Maize Controls (± N) | 1 | severe | -42 % | (-50 % to – 35 %) |
| | Maize/Grass relay + 3 C $\text{ha}^{-1}$ farmyard manure (± N) | 2a | | | |
| | Maize/Grass relay – farmyard manure (± N) | 2b | | | |
| 1.2 & 4 t C $\text{ha}^{-1}$ of saw dust/maize stover (± N) | Maize/Legume rotation + 3 C $\text{ha}^{-1}$ farmyard manure (± N) | 2c | severe to strong | -37 % | (-44 % to – 29 % |
| | Maize/Legume rotation – farmyard manure (± N) | 2d | | | |
| 1.2 t C $\text{ha}^{-1}$ of *Tithonia* and *Calliandra* (± N) | Maize/Legume rotation + 3 t C $\text{ha}^{-1}$ *Tithonia* (± N) | 3a | strong | -36 % | (-44 % to – 28 %) |
| | Maize/Legume MBILI + 3 t C $\text{ha}^{-1}$ *Tithonia* (± N) | 3b | | | |
| 4 t C $\text{ha}^{-1}$ of *Tithonia* and *Calliandra* (± N) | Maize/Legume rotation + 3 t C $\text{ha}^{-1}$ *Tithonia* (± N) | 4a | medium | -25 % | (-32 % to – 17 %) |
| | Maize/Legume MBILI + 3 t C $\text{ha}^{-1}$ *Tithonia* (± N) | 4b | | | |
| 1.2 t C $\text{ha}^{-1}$ of farmyard manure (± N) | Maize/Legume MBILI + 1.2 t C $\text{ha}^{-1}$ farmyard manure (± N) | 5 | medium | -27 % | (-35 % to – 20 %) |
| 4 t C $\text{ha}^{-1}$ of farmyard manure (± N) | Maize/Legume MBILI + 4 t C $\text{ha}^{-1}$ farmyard manure (± N) | 6 | low | -13 % | (-21 % to – 5 %) |

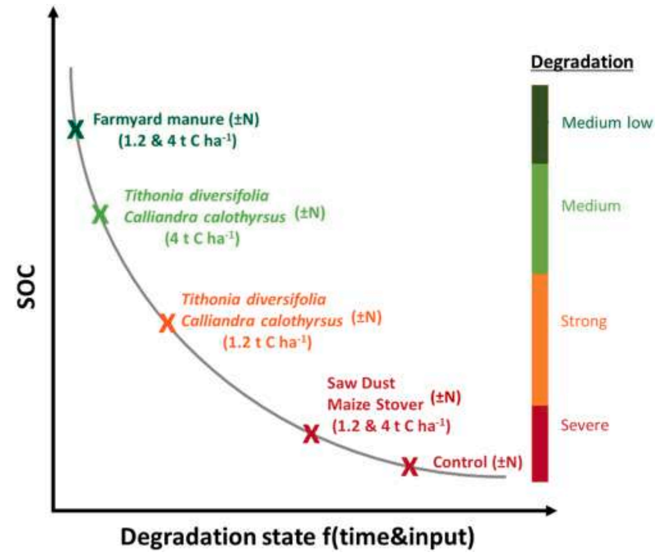

**Fig. 3.** Schematic illustration of the experimental treatments grouped together according to similar SOC decline and soil degradation. The degradation state is a function of time and the type and quantity of resource inputs. The conceptual degradation framework was a basis for the redesign of the new experimental treatments outlined in Table 1. The ± N stands for the addition or absence of mineral N fertilizer.

systems aimed at soil regeneration, were studied using treatments that had resulted in medium to severe soil degradation levels. To achieve higher SOC regeneration potential, these treatments were replaced with new ones. It was acknowledged that as the severity of SOC degradation increased, increasingly dense root systems would be necessary. The modifications aim to study the effectiveness of increasing SOC through inputs of root litter and rhizodeposition compared to inputs of external organic resources. The perceived limitation of the experiments, having different SOC contents after about 20 years of treatment implementation, was thereby turned into a strength. The different SOC contents allowed testing a range of treatments for their potential to regenerate SOC, starting from different levels of degradation, all while maintaining the same types of soil texture and mineralogy across four different experimental sites with a consistent design. Locating such a gradient on new sites with unclear past management and ambiguous soil degradation levels would be challenging. Additionally, precise knowledge of past management and weather is crucial for modeling studies. Finally, erosion barriers were installed around individual plots at the Machanga site in response to observed topsoil erosion in the previous design.

We redesigned the experiment based on the finding that external organic resource inputs alone were insufficient to increase SOC. Our hypothesis was that inputs from the cropping system itself, such as roots and their exudates, play a dominant role in regenerating SOC (Prescott et al., 2021). The reasons for this postulation are that rhizosphere microbes are more efficient in stabilizing SOC, mainly due to their proximity to soil minerals. In contrast, surface-deposited organic materials undergo more decomposition and recycling phases before they are stabilized as SOC (Sokol et al., 2019; Sokol and Bradford, 2019). Therefore, our focus shifted towards new cropping systems that can increase the quantity and/or quality of root inputs while simultaneously increasing crop yields. To improve the quality of root inputs, we included legume species that are commonly planted by farmers to diversify their cropping system (Oduor et al., 2022). These species have shown potential to regenerate soils (Nezomba et al., 2015; Boddey et al., 2010; Yang et al., 2023). We recognized the importance of comparing crop rotation and intercropping of maize and legumes, to disentangle the possible benefits of intercropping on SOC formation and maize yield through facilitation mechanisms such as nutrient acquisition and pest control. The “Managing Beneficial Interactions in Legume intercrops” (MBILI) system (Woomer et al., 2004; Oduor et al., 2022) was chosen, as a promising intercropping method that alternates double rows of maize with double rows of legumes. It has been shown to achieve higher crop yields compared to single row intercropping (Mucheru-Muna et al., 2010; Ngetich et al., 2014). The hypothesis is that the MBILI system also results in greater root biomass inputs with a lower C:N ratio than the maize monocrop. Another cropping system that is believed to increase the quantity of root inputs involves using forage grasses with a permanent root system, planted in relay with maize (February et al., 2020). Relay intercropping of maize with fodder grasses has been shown to produce significantly more root biomass without strongly reducing maize grain yield (Canisares et al., 2021; Souza et al., 2024) and to enhance SOC formation (da Silva Rodrigues Pinto et al., 2022). Given the prevalence of mixed crop-livestock systems in Kenya, the implementation of a grass is a suitable treatment option – it produces animal fodder for the commonly practiced zero-grazing system of smallholder dairy production, which is also a source of farmyard manure. The experiment tests the significance of external versus in-situ C inputs in regenerating SOC in severely degraded plots, with treatments including and excluding farmyard manure addition.

Due to the density of the root system of fodder grasses, it is hypothesized that this treatment will be the most effective in regenerating SOC in the plots with severe degradation. Conversely, the treatments with medium levels of soil degradation are hypothesized to require lower rates of root inputs for recovery, allowing for a more intensive cropping system. These treatments were thus used to test the effect of maize-legume MBILI intercropping compared to maize-legume rotation, with identical legume species in both systems. The previous farmyard manure treatments were transitioned to the MBILI intercropping system instead of maize monocropping to investigate whether a denser root system can increase SOC beyond current levels. Other potential options, such as installing a permanent grass vegetation in the plots for several years or replacing maize with millet and sorghum, were considered but ultimately discarded due to their low suitability for farmers. In central and western Kenya, most farmers are smallholders who rely on farming for their food security and livelihoods. They have small field plots, averaging 0.5 ha (Muyanga and Jayne, 2019) and maize is their main staple and cash crop (Marenya et al., 2022). Therefore, farmers are hesitant to adopt cropping systems that do not involve maize or make the land unproductive for extended periods.

While these new treatments/cropping systems add more complexity to the experiments, they are appropriate to test the benefits of in situ versus external organic resources (on the severely degraded treatments) and of intercropping versus crop rotation (on the strongly to medium degraded plots). In addition, they provide an opportunity to assess the potential for increasing SOC beyond current levels through intercropping intensification (on the plots with medium to low degradation, i.e., the former farmyard manure treatments). The split-plot mineral N fertilizer treatments (i.e., with versus without N fertilizer) are maintained to continue testing the effects of N limitation on crop yield, root-derived C inputs, and SOC. In addition, to increase N use efficiency, the N fertilizer rate is reduced to 90 kg N ha$^{-1}$ season$^{-1}$. The baseline phosphorus (P) fertilizer rate is also reduced, based on analyzes of available P in 2022: 10 kg P ha$^{-1}$ season$^{-1}$ in Aludeka and Machanga, and 30 and 20 kg P ha$^{-1}$ in Embu and Sidada, respectively. Finally, for the most efficient use of organic resources, they are now applied in two parts, i.e., half before planting in the long rainy season and the other half before planting in the short rainy season.

Following the January 2022 workshop, pre-trials (Fig. 4) were conducted on plots adjacent to the main LTEs to test different legume species (soybean (*Glycine max*), groundnut (*Arachis hypogaea*), pigeonpea (*Cajanus cajan*)) and forage species (*Panicum maximum* and *Brachiaria brizantha*) in sole cropping and MBILI intercropping, prior to their possible establishment in the experiments. These pre-trials were also an opportunity to discuss the feasibility of the new cropping systems with

**Fig. 4.** Preliminary tests of groundnut and soybean, both as sole legume crops and intercropped (MBILI) legumes have been conducted on plots adjacent to the main experiment. From left to right: sole soybean, sole groundnut, soybean intercropped with maize, and groundnut intercropped with maize (pictures from June 2022 at the Sidada experimental site in western Kenya).

local farmers. In the September 2022 workshop, we evaluated the results of the pre-trials and identified the final experimental crops for the redesigned experiments (Table 1): namely, soybean was chosen as the legume and *Brachiaria brizantha* as the forage grass for the experiments in western Kenya, and groundnut and *Panicum maximum* were selected for central Kenya. These selections were based on discussions with local farmers and on the yield potential of each species, including their land equivalent ratios in the intercropping systems. The redesign of the experiments was implemented in early 2023.

Finally, in July 2023, two workshops were held with the participation of local farmers, extension officers and government bodies – one in western and one in central Kenya. During these workshops, we collectively visited the redesigned field experiments, engaged in discussions about both the old and new designs, and evaluated the feasibility of the new cropping systems. The objectives were to strengthen the relationship with local stakeholders for the upcoming experimental phase, establish the experimental sites as local innovation hubs, and to ensure the feasibility of the new treatments by incorporating insights from local stakeholders. The workshops emphasized the importance of integrating the experiments with local extension activities, such as using them as field demonstration sites for farmers. Local stakeholders were also impressed by the differences in standing biomass between the treatments, which were mostly attributable to the old treatments (e.g., sawdust versus farmyard manure) at that time. During the field visits with farmers and other stakeholders, the MBILI system, while not yet widely known, was perceived as very interesting. Although local stakeholders expressed skepticism about the forage grass relay system, they indicated their willingness to learn more about it through the experiments.

## 4. Key lessons learned

Several key lessons were learned from the previous design and applied to the redesign. Firstly, only organic resources that were low in C:N ratio, such as farmyard manure and *Tithonia*, were effective in forming SOC. Secondly, external organic resources, even at unrealistically high rates compared to farmers` practices, were mostly insufficient to build enough new SOC to transition a site from a loss to a gain trajectory. Thirdly, although mineral N addition significantly increased maize yields, particularly when combined with organic resources, its impact on SOC was minimal, at least in maize monocultures. Fourthly, the treatments that were the most effective in preserving SOC (namely, farmyard manure and to some extent *Tithonia* and *Calliandra*) were also the most effective in maintaining maize yields over time. Therefore, the subsequent phase of the experiments required a focus on enhancing SOC formation from within the system itself, while maintaining the most effective organic resource treatments at more practical application rates.

Furthermore, practical lessons were learned regarding the frequency and design of soil sampling. For instance, a statistical analysis was conducted to determine the minimum number of years of data needed to detect significant differences in SOC. Subsets of data with varying numbers of years included were analyzed using a linear mixed-effects model with random intercepts per replicate and a slope effect on experimental time. The post-hoc analysis showed that it would take a minimum of three to five years of SOC measurements to detect significant differences in SOC contents between the treatments at the same site (Fig. 5). This finding is consistent with earlier studies on plot-scale variability of soils (Poeplau et al., 2022). Another lesson was that the soil samples collected in 2021, i.e., from depths of the 50–70 cm layer, exhibited a higher variability than previous samples taken from the top 15 cm. Due to the difficulty in sampling deep soil layers, the 2021 samples were obtained from a single insertion, unlike previous years when samples were composite samples of five insertions. Therefore, all future soil sampling will utilize composite sampling methods. In cases where labor is a constraint, the number of sampled treatments will be reduced. Finally, it is important to avoid gaps in SOC data in the current redesigned LTE. For instance, there was a gap in soil sampling between 2005 and 2018 in Aludeka and Sidada due to the transfer of the management responsibility from the International Center for Tropical Agriculture (CIAT) to IITA, and due to budget limitations during several years.

## 5. The way forward

The four SOC experiments in Kenya were redesigned to include experimental treatments that are feasible for farmers and have the potential to increase SOC. These treatments are also well-suited to contribute data to the ongoing discourse on the most efficient mechanisms for SOC formation (Angst et al., 2021; Cotrufo et al., 2013; Kleber et al., 2021; Prescott et al., 2021). The redesign was guided by important research hypotheses, which are summarized in Box 1.

With the redesign, we moved away from outdated concepts that emphasized the importance of "recalcitrant" plant material and the concept that C use efficiency is an exclusive property of organic resources (Manzoni et al., 2012). Instead, a more holistic perspective has been adopted, considering SOC storage and stabilization efficiency at the agroecosystem level (Manzoni et al., 2018), while also taking into account potential trade-offs with crop productivity and yield. In the redesigned experiments, organic inputs will be measured via root biomass and quality (Palm et al., 2001a), and SOC formation will be measured using different operational fractions of SOC. Sensitive indicators, such as SOC in the free silt and clay particle size fraction versus water stable aggregates and particulate organic C (Cotrufo et al., 2022; Six et al., 1999), as well as microbial biomass C and N (Amato and Ladd, 1988; Brookes et al., 1985; Vance et al., 1987), will serve as early indicators of SOC formation. These measurements will be conducted for the first time two to three years after the redesign of the experiment. By utilizing these measurements to adjust the parameters of the most recent SOC models (Abramoff et al., 2022; Della Chiesa et al., 2022; Laub et al., 2023c), the revised experiments will enhance our fundamental comprehension of how SOC is formed in situ and its relationship with attainable crop yields. It is planned to continue assessing seasonal yields and measuring bulk SOC every three to four years. Soil samples taken just before the experiment redesign were archived for future comparisons of SOC and SOC fractions. By rerandomizing the new treatments within groups of soil degradation levels, it should be possible to

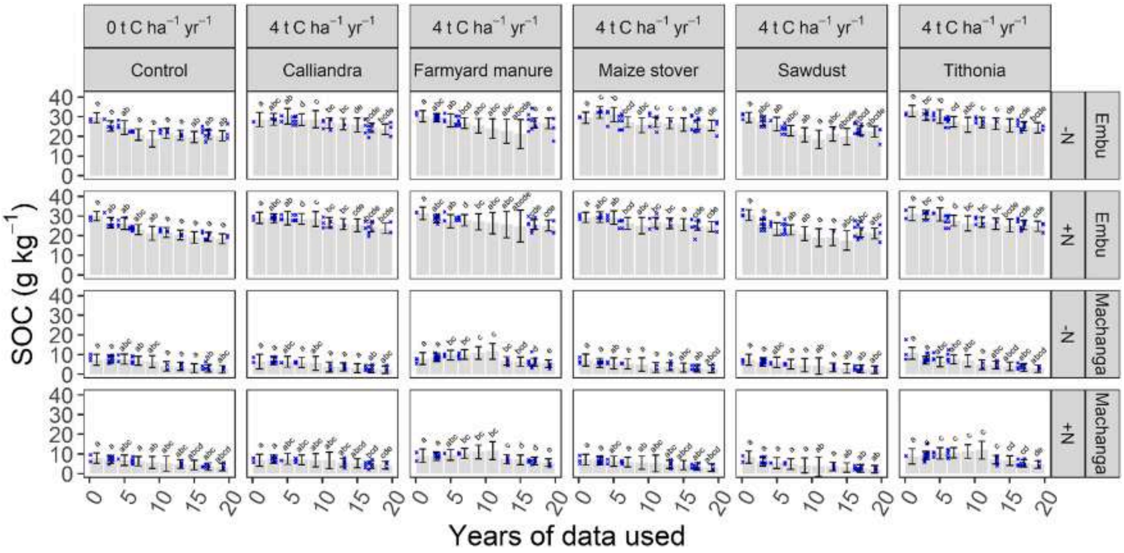

**Fig. 5.** Statistical analysis of SOC content data, showing the effect of the number of years of available SOC data on predicted SOC contents. The mixed statistical model employed a treatment × time interaction per site with a random intercept per experimental replicate. The displayed post hoc predictions (bars and error bars) for each year only made use of the SOC data that was available until that year. Same lowercase letters indicate the absence of significantly different SOC contents between treatments at the same site and time point ($p < 0.05$). The error bars areas indicate the 95 % confidence intervals for the true mean of SOC post hoc predictions. Blue crosses represent measured data.

**Box 1**

Main research hypotheses underlying the new experimental design:

1) Root inputs are more efficient in forming SOC than external inputs.
2) Moderate N limitations result in more root C input into the soil compared to situations with no N limitations.
3) The efficiency of mineral-attached SOC stabilization is determined by the C:N ratio and aromatic compounds of root (and external organic) inputs.
4) With increasing soil degradation, external organic resources become increasingly important for the formation of SOC and root inputs.
5) SOC and crop yields are more difficult to regenerate in soils with more weathered clay types, such as kaolinite, or with lower amounts of clay.

distinguish between the legacy effects of the old treatments and the effects of the new treatments. The new treatments were designed with the farmers' perspectives in mind, including valuable crops for smallholders. Planned analyses of yield, land equivalent ratios and value-to-cost ratios (Njoroge et al., 2017) will provide further insights into the agronomic feasibility of the new cropping systems for smallholders. Finally, analyzing the relationship between crop yield and SOC formation can provide new insights into potential trade-offs between yield and SOC increase. This topic remains scientifically significant and motivates us to engage in future collaborative efforts to conduct research on a broader scale. These efforts would involve the integration of data from both existing and upcoming long-term experiments in modeling studies. Ideally, they should be conducted across various regions, both within the African continent and globally. The ultimate objective is to generate novel research inquiries on soil health restoration strategies for sustainable intensification and climate change adaptation.

## CRediT authorship contribution statement

**Monicah Wanjiku Mucheru-Muna:** Writing – review & editing, Resources, Project administration, Funding acquisition, Data curation. **Samuel Mathu Ndungu:** Writing – review & editing, Resources, Project administration, Data curation. **Marc Corbeels:** Writing – review & editing, Supervision, Funding acquisition, Conceptualization. **Moritz Laub:** Writing – review & editing, Writing – original draft, Visualization, Investigation, Data curation. **Bernard Vanlauwe:** Writing – review & editing, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization. **Wycliffe Waswa:** Resources, Project administration, Methodology, Data curation. **Rebecca Yegon:** Writing – review & editing, Resources, Project administration, Conceptualization. **Daniel Mugendi:** Supervision, Resources, Funding acquisition, Conceptualization. **Johan Six:** Writing – review & editing, Writing – original draft, Resources, Project administration, Investigation, Funding acquisition, Conceptualization.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

Stored soil samples, and subsamples of maize harvest for some years, as well as the raw data from the long-term experiments are available to share with interested parties (For SOC: https://doi.org/10.25502/wdh5-6c13/d. For yields and biomass: https://doi.org/10.25502/be9y-xh75/d.).

## Acknowledgements

The work in the four LTEs in Kenya has been supported by the Schweizerischer Nationalfonds zur Förderung der Wissenschaftlichen Forschung (Grant no. 172940), the Agropolis Fondation (Grant no. ANR-10-LABX-0001-01), the TOTAL Foundation (patronage agreement), the European Union's Horizon 2020 framework (LANDMARC; Grant no. 869367), DSCATT Project "Agricultural Intensification and Dynamics of Soil Carbon Sequestration in Tropical and Temperate Farming Systems" (Grant nos. AF 1802-001 and FT C002181) and the CGIAR Excellence in Agronomy (EiA) Initiative 11.

## References

Abramoff, R.Z., Guenet, B., Zhang, H., Georgiou, K., Xu, X., Viscarra Rossel, R.A., Yuan, W., Ciais, P., 2022. Improved global-scale predictions of soil carbon stocks with Millennial Version 2. Soil Biol. Biochem. 108466 https://doi.org/10.1016/j.soilbio.2021.108466.

Amato, M., Ladd, J.N., 1988. Assay for microbial biomass based on ninhydrin-reactive nitrogen in extracts of fumigated soils. Soil Biol. Biochem. https://doi.org/10.1016/0038-0717(88)90134-4.

Angst, G., Mueller, K.E., Nierop, K.G.J., Simpson, M.J., 2021. Plant- or microbial-derived? A review on the molecular composition of stabilized soil organic matter. Soil Biol. Biochem. 156, 108189 https://doi.org/10.1016/j.soilbio.2021.108189.

Bashir, O., Ali, T., Baba, Z.A., Rather, G.H., Bangroo, S.A., Mukhtar, S.D., Naik, N., Mohiuddin, R., Bharati, V., Bhat, R.A., 2021 soil organic matter and its impact on soil properties and nutrient status. In: Dar, G.H., Bhat, R.A., Mehmood, M.A., Hakeem, K.R. (eds.), Microbiota and Biofertilizers, Vol 2: Ecofriendly Tools for Reclamation of Degraded Soil Environs. Springer International Publishing, Cham, pp. 129–59. 〈https://doi.org/10.1007/978-3-030-61010-4_7〉.

Beillouin, D., Corbeels, M., Demenois, J., Berre, D., Boyer, A., Fallot, A., Feder, F., Cardinael, R., 2023. A global meta-analysis of soil organic carbon in the Anthropocene. Nat. Commun. 14, 3700. https://doi.org/10.1038/s41467-023-39338-z.

Boddey, R.M., Jantalia, C.P., Conceição, P.C., Zanatta, J.A., Bayer, C., Mielniczuk, J., Dieckow, J., Dos Santos, H.P., Denardin, J.E., Aita, C., Giacomini, S.J., Alves, B.J., 2010. r., and Urquiaga, S.: carbon accumulation at depth in Ferralsols under zero-till subtropical agriculture. Glob. Change Biol. 16, 784–795. https://doi.org/10.1111/j.1365-2486.2009.02020.x.

Brookes, P.C., Landman, A., Pruden, G., Jenkinson, D.S., 1985. Chloroform fumigation and the release of soil nitrogen: a rapid direct extraction method to measure microbial biomass nitrogen in soil. Soil Biol. Biochem. 17, 837–842. https://doi.org/10.1016/0038-0717(85)90144-0.

Bruun, S., Jensen, L.S., 2002. Initialisation of the soil organic matter pools of the Daisy model. Ecol. Model. 153, 291–295. https://doi.org/10.17665/1676-4285.20155108.

Canisares, L.P., Rosolem, C.A., Momesso, L., Crusciol, C.A.C., Villegas, D.M., Arango, J., Ritz, K., Cantarella, H., 2021. Maize-Brachiaria intercropping: a strategy to supply recycled N to maize and reduce soil N2O emissions? Agric. Ecosyst. Environ. 319, 107491 https://doi.org/10.1016/j.agee.2021.107491.

Corbeels, M., Cardinael, R., Naudin, K., Guibert, H., Torquebiau, E., 2019. The 4 per 1000 goal and soil carbon storage under agroforestry and conservation agriculture systems in sub-Saharan Africa. Soil Tillage Res. 188, 16–26. https://doi.org/10.1016/j.still.2018.02.015.

Cotrufo, M.F., Haddix, M.L., Kroeger, M.E., Stewart, C.E., 2022. The role of plant input physical-chemical properties, and microbial and soil chemical diversity on the formation of particulate and mineral-associated organic matter. Soil Biol. Biochem., 108648 https://doi.org/10.1016/j.soilbio.2022.108648.

Cotrufo, M.F., Wallenstein, M.D., Boot, C.M., Denef, K., Paul, E., 2013. The Microbial Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: do labile plant inputs form stable soil organic matter? Glob. Change Biol. 19, 988–995. https://doi.org/10.1111/gcb.12113.

Couëdel, A., Falconnier, G.N., Adam, M., Cardinael, R., Boote, K., Justes, E., Smith, W.N., Whitbread, A.M., Affholder, F., Balkovic, J., Basso, B., Bhatia, A., Chakrabarti, B., Chikowo, R., Christina, M., Faye, B., Ferchaud, F., Folberth, C., Akinseye, F.M., Gaiser, T., Galdos, M.V., Gayler, S., Gorooei, A., Grant, B., Guibert, H., Hoogenboom, G., Kamali, B., Laub, M., Maureira, F., Mequanint, F., Nendel, C., Porter, C.H., Ripoche, D., Ruane, A.C., Rusinamhodzi, L., Sharma, S., Singh, U., Six, J., Srivastava, A., Vanlauwe, B., Versini, A., Vianna, M., Webber, H., Weber, T.K. D., Zhang, C., Corbeels, M., 2024. Long-term soil organic carbon and crop yield feedbacks differ between 16 soil-crop models in sub-Saharan Africa. Eur. J. Agron. 155, 127109 https://doi.org/10.1016/j.eja.2024.127109.

da Silva Rodrigues Pinto, L.A., de Sousa Morais, I., Ozório, J.M.B., de Melo, T.R., Rosset, J.S., Pereira, M.G., 2022. Soil aggregation and associated organic matter under management systems in sandy-textured soils, subtropical region of Brazil. Environ. Monit. Assess. 195, 253. https://doi.org/10.1007/s10661-022-10892-1.

Della Chiesa, T., Del Grosso, S.J., Hartman, M.D., Parton, W.J., Echarte, L., Yahdjian, L., Piñeiro, G., 2022. A novel mechanism to simulate intercropping and relay cropping using the DayCent model. Ecol. Model. 465, 109869 https://doi.org/10.1016/j.ecolmodel.2021.109869.

Denef, K., Plante, A.F., Six, J., 2009. Characterization of soil organic matter. In: Heinemeyer, A., Bahn, M., Kutsch, W.L. (eds.), Soil Carbon Dynamics: An Integrated Methodology. Cambridge University Press, Cambridge, pp. 91–126. 〈https://doi.org/10.1017/CBO9780511711794.007〉.

February, E., Pausch, J., Higgins, S.I., 2020. Major contribution of grass roots to soil carbon pools and CO2 fluxes in a mesic savanna. Plant Soil 454, 207–215. https://doi.org/10.1007/s11104-020-04649-3.

Fließbach, A., Oberholzer, H.-R., Gunst, L., Mäder, P., 2007. Soil organic matter and biological soil quality indicators after 21 years of organic and conventional farming. Agric. Ecosyst. Environ. 118, 273–284. https://doi.org/10.1016/j.agee.2006.05.022.

Frimmel, F.H., Christman, R.F., 1988. Humic Substances and Their Role in the Environment. John Wiley and Sons Ltd.

Gvein, M.H., Hu, X., Næss, J.S., Watanabe, M.D.B., Cavalett, O., Malbranque, M., Kindermann, G., Cherubini, F., 2023. Potential of land-based climate change mitigation strategies on abandoned cropland. Commun. Earth Environ. 4, 1–16. https://doi.org/10.1038/s43247-023-00696-7.

Janzen, H.H., Janzen, D.W., Gregorich, E.G., 2021. The 'soil health' metaphor: illuminating or illusory? Soil Biol. Biochem. 159, 108167 https://doi.org/10.1016/j.soilbio.2021.108167.

Kallenbach, C.M., Frey, S.D., Grandy, A.S., 2016. Direct evidence for microbial-derived soil organic matter formation and its ecophysiological controls. Nat. Commun. 7, 1–10. https://doi.org/10.1038/ncomms13630.

Kätterer, T., Bolinder, M.A., Andrén, O., Kirchmann, H., Menichetti, L., 2011. Roots contribute more to refractory soil organic matter than above-ground crop residues, as revealed by a long-term field experiment. Agric. Ecosyst. Environ. 141, 184–192. https://doi.org/10.1016/j.agee.2011.02.029.

Kleber, M., Bourg, I.C., Coward, E.K., Hansel, C.M., Myneni, S.C.B., Nunan, N., 2021. Dynamic interactions at the mineral–organic matter interface. Nat. Rev. Earth Environ. 2, 402–421. https://doi.org/10.1038/s43017-021-00162-y.

Lal, R., 2004. Soil carbon sequestration impacts on global climate change and food security. Science 304, 1623–1627. https://doi.org/10.1126/science.1097396.

Laub, M., Blagodatsky, S., Van De Broek, M., Schlichenmaier, S., Kunlanit, B., Six, J., Vityakon, P., Cadisch, G., 2023c. SAMM version 1.0: a numerical model for microbial mediated soil aggregate formation. EGUsphere 1–38. https://doi.org/10.5194/egusphere-2023-1414.

Laub, M., Corbeels, M., Couëdel, A., Ndungu, S.M., Mucheru-Muna, M.W., Mugendi, D., Necpalova, M., Waswa, W., Van de Broek, M., Vanlauwe, B., Six, J., 2023b. Managing soil organic carbon in tropical agroecosystems: evidence from four long-term experiments in Kenya. Soil 9, 301–323. https://doi.org/10.5194/soil-9-301-2023.

Laub, M., Corbeels, M., Mathu Ndungu, S., Mucheru-Muna, M.W., Mugendi, D., Necpalova, M., Van de Broek, M., Waswa, W., Vanlauwe, B., Six, J., 2023a. Combining manure with mineral N fertilizer maintains maize yields: evidence from four long-term experiments in Kenya. Field Crops Res. 291, 108788 https://doi.org/10.1016/j.fcr.2022.108788.

Laub, M., Demyan, M.S., Nkwain, Y.F., Blagodatsky, S., Kätterer, T., Piepho, H., Cadisch, G., 2020. DRIFTS band areas as measured pool size proxy to reduce parameter uncertainty in soil organic matter models. Biogeosciences 17, 1393–1413. https://doi.org/10.5194/bg-17-1393-2020.

Manzoni, S., Čapek, P., Porada, P., Thurner, M., Winterdahl, M., Beer, C., Brüchert, V., Frouz, J., Herrmann, A.M., Lindahl, B.D., Lyon, S.W., Šantrůčková, H., Vico, G., Way, D., 2018. Reviews and syntheses: carbon use efficiency from organisms to ecosystems – definitions, theories, and empirical evidence. Biogeosciences 15, 5929–5949. https://doi.org/10.5194/bg-15-5929-2018.

Manzoni, S., Taylor, P., Richter, A., Porporato, A., Ågren, G.I., 2012. Environmental and stoichiometric controls on microbial carbon-use efficiency in soils. New Phytol. 196, 79–91. https://doi.org/10.1111/j.1469-8137.2012.04225.x.

Marenya, P.P., Wanyama, R., Alemu, S., Woyengo, V., 2022. Building resilient maize production systems with stress-adapted varieties: farmers' priorities in Western Kenya. Front. Sustain. Food Syst. 6.

Minasny, B., Malone, B.P., McBratney, A.B., Angers, D.A., Arrouays, D., Chambers, A., Chaplot, V., Chen, Z.-S., Cheng, K., Das, B.S., Field, D.J., Gimona, A., Hedley, C.B., Hong, S.Y., Mandal, B., Marchant, B.P., Martin, M., McConkey, B.G., Mulder, V.L., O'Rourke, S., Richer-de-Forges, A.C., Odeh, I., Padarian, J., Paustian, K., Pan, G., Poggio, L., Savin, I., Stolbovoy, V., Stockmann, U., Sulaeman, Y., Tsui, C.-C., Vågen, T.-G., van Wesemael, B., Winowiecki, L., 2017. Soil carbon 4 per mille. Geoderma 292, 59–86. https://doi.org/10.1016/j.geoderma.2017.01.002.

Mucheru-Muna, M., Pypers, P., Mugendi, D., Kung'u, J., Mugwe, J., Merckx, R., Vanlauwe, B., 2010. A staggered maize–legume intercrop arrangement robustly increases crop yields and economic returns in the highlands of Central Kenya. Field Crops Res. 115, 132–139. https://doi.org/10.1016/j.fcr.2009.10.013.

Muyanga, M., Jayne, T.S., 2019. Revisiting the farm size-productivity relationship based on a relatively wide range of farm sizes: evidence from Kenya. Am. J. Agric. Econ. 101, 1140–1163. https://doi.org/10.1093/ajae/aaz003.

Necpalova, M., Lee, J., Skinner, C., Büchi, L., Wittwer, R., Gattinger, A., van der Heijden, M., Mäder, P., Charles, R., Berner, A., Mayer, J., Six, J., 2018. Potentials to mitigate greenhouse gas emissions from Swiss agriculture. Agric. Ecosyst. Environ. 265, 84–102. https://doi.org/10.1016/j.agee.2018.05.013.

Nezomba, H., Mtambanengwe, F., Tittonell, P., Mapfumo, P., 2015. Point of no return? Rehabilitating degraded soils for increased crop productivity on smallholder farms in eastern Zimbabwe. Geoderma 239–240, 143–155. https://doi.org/10.1016/j.geoderma.2014.10.006.

Ngetich, K.F., Diels, J., Shisanya, C.A., Mugwe, J.N., Mucheru-Muna, M., Mugendi, D.N., 2014. Effects of selected soil and water conservation techniques on runoff, sediment yield and maize productivity under sub-humid and semi-arid conditions in Kenya. Catena 121, 288–296. https://doi.org/10.1016/j.catena.2014.05.026.

Njoroge, R., Otinga, A.N., Okalebo, J.R., Pepela, M., Merckx, R., 2017. Occurrence of poorly responsive soils in western Kenya and associated nutrient imbalances in maize (Zea mays L.). Field Crops Res. 210, 162–174. https://doi.org/10.1016/j.fcr.2017.05.015.

Oduor, N., Mucheru-Muna, M., Mugwe, J., Sijali, I., Nyabuga, G., Mugendi, D.N., 2022. Nutrient and organic carbon losses by erosion, and their economic and environmental implications in the drylands of Kenya. Environ. Chall. 7, 100519 https://doi.org/10.1016/j.envc.2022.100519.

Palm, C., Gachengo, C.N., Delve, R.J., Cadisch, G., Giller, K.E., 2001b. Organic inputs for soil fertility management in tropical agroecosystems: application of an organic resource database. Agric. Ecosyst. Environ. 83, 27–42. https://doi.org/10.1016/S0167-8809(00)00267-X.

Palm, C., Giller, K.E., Mafongoya, P.L., Swift, M.J., 2001a. Management of organic matter in the tropics: translating theory into practice. Nutr. Cycl. Agroecosyst. 61, 63–75. https://doi.org/10.1023/A:1013318210809.

Paul, C., Bartkowski, B., Dönmez, C., Don, A., Mayer, S., Steffens, M., Weigl, S., Wiesmeier, M., Wolf, A., Helming, K., 2023. Carbon farming: are soil carbon certificates a suitable tool for climate change mitigation? J. Environ. Manag. 330, 117142 https://doi.org/10.1016/j.jenvman.2022.117142.

Poeplau, C., Prietz, R., Don, A., 2022. Plot-scale variability of organic carbon in temperate agricultural soils—implications for soil monitoring. J. Plant Nutr. Soil Sci. 185, 403–416. https://doi.org/10.1002/jpln.202100393.

Pohl, C., Krütli, P., Stauffacher, M., 2017. Ten reflective steps for rendering research societally relevant. GAIA - Ecol. Perspect. Sci. Soc. 26, 43–51. https://doi.org/10.14512/gaia.26.1.10.

Popkin, G., 2023. Shaky ground. Science 381, 369–373. https://doi.org/10.1126/science.adj9318.

Prescott, C.E., Rui, Y., Cotrufo, M.F., Grayston, S.J., 2021. Managing plant surplus carbon to generate soil organic matter in regenerative agriculture. J. Soil Water Conserv. 76, 99A–104A. https://doi.org/10.2489/jswc.2021.0920A.

Puttaso, A., Vityakon, P., Rasche, F., Saenjan, P., Treloges, V., Cadisch, G., 2013. Does organic residue quality influence carbon retention in a tropical sandy soil? Soil Sci. Soc. Am. J. 77, 1001–10011. https://doi.org/10.2136/sssaj2012.0209.

Rumpel, C., Amiraslani, F., Chenu, C., Garcia Cardenas, M., Kaonga, M., Koutika, L.-S., Ladha, J., Madari, B., Shirato, Y., Smith, P., Soudi, B., Soussana, J.-F., Whitehead, D., Wollenberg, E., 2020. The 4p1000 initiative: opportunities, limitations and challenges for implementing soil organic carbon sequestration as a sustainable development strategy. Ambio 49, 350–360. https://doi.org/10.1007/s13280-019-01165-2.

Sileshi, G.W., Jama, B., Vanlauwe, B., Negassa, W., Harawa, R., Kiwia, A., Kimani, D., 2019. Nutrient use efficiency and crop yield response to the combined application of cattle manure and inorganic fertilizer in sub-Saharan Africa. Nutr. Cycl. Agroecosyst. 113, 181–199. https://doi.org/10.1007/s10705-019-09974-3.

Six, J., Elliott, E.T., Paustian, K., 1999. Aggregate and soil organic matter dynamics under conventional and no-tillage systems. Soil Sci. Soc. Am. J. 63, 1350–1358. https://doi.org/10.2136/sssaj1999.6351350x.

Sokol, N.W., Bradford, M.A., 2019. Microbial formation of stable soil carbon is more efficient from belowground than aboveground input. Nat. Geosci. 12, 46–53. https://doi.org/10.1038/s41561-018-0258-6.

Sokol, N.W., Sanderman, J., Bradford, M.A., 2019. Pathways of mineral-associated soil organic matter formation: integrating the role of plant carbon source, chemistry, and point of entry. Glob. Change Biol. 25, 12–24. https://doi.org/10.1111/gcb.14482.

Souza, L.F.N., Ciampitti, I.A., Fernandez, J.A., Favarin, J.L., de Oliveira, S.M., 2024. Maize-Brachiaria grass intercropping: a meta-analysis of major productivity drivers in Brazil. Field Crops Res. 306, 109205 https://doi.org/10.1016/j.fcr.2023.109205.

Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. An extraction method for measuring soil microbial biomass C. Soil Biol. Biochem. 19, 703–707. https://doi.org/10.1016/0038-0717(87)90052-6.

Watts, C.W., Clark, L.J., Poulton, P.R., Powlson, D.S., Whitmore, A.P., 2006. The role of clay, organic carbon and long-term management on mouldboard plough draught measured on the Broadbalk wheat experiment at Rothamsted. Soil Use Manag. 22, 334–341. https://doi.org/10.1111/j.1475-2743.2006.00054.x.

Woomer, P.L., Lan'gat, M., Tungani, J.O., 2004. Innovative maize-legume intercropping results in above- and below-ground competitive advantages for understorey legumes. West Afr. J. Appl. Ecol. 6 https://doi.org/10.4314/wajae.v6i1.45609.

Woomer, P.L., Swift, M.J., 1994. The Biological Management of Tropical Soil Fertility. John Wiley, Chichester, UK, p. 243.

Yang, L., Luo, Y., Lu, B., Zhou, G., Chang, D., Gao, S., Zhang, J., Che, Z., Cao, W., 2023. Long-term maize and pea intercropping improved subsoil carbon storage while reduced greenhouse gas emissions. Agric. Ecosyst. Environ. 349, 108444 https://doi.org/10.1016/j.agee.2023.108444.

Zhou, Q., Ernst, P.A., Morgan, K.L., Rubin, D.B., Zhang, A., 2018. Sequential rerandomization. Biometrika 105, 745–752. https://doi.org/10.1093/biomet/asy031.