



*Supplement of*

## **Limited effect of organic matter addition on stabilised organic carbon in four tropical arable soils**

**Marijn Van de Broek et al.**

*Correspondence to:* Marijn Van de Broek ([marijn.vandebroek@usys.ethz.ch](mailto:marijn.vandebroek@usys.ethz.ch))

The copyright of individual parts of the supplement might differ from the article licence.

# Supplementary information

## Contents

<b>S1 Supplementary Text</b>	<b>3</b>
S1.1 Statistics for yield and aboveground biomass . . . . .	3
S1.2 Statistics for bulk density between treatments at the same site . . . . .	3
S1.3 Statistics for the portion of POC and MAOC of total SOC . . . . .	3
<b>S2 Supplementary figures</b>	<b>4</b>

## **S1 Supplementary Text**

### **S1.1 Statistics for yield and aboveground biomass**

The analysis of significant differences in yield and total aboveground biomass between the treatments per site (Figures S4 and S5) was done after fitting a linear mixed effects model (using the *lme* function from the *nlme* package in R; Pinheiro et al., 2024) following the procedures outlined in Zuur et al. (2009), with site and treatment as fixed effect and nested site/block as random effect. A variance structure (*varIdent*) was added to allow for differences in variance for site and treatment (Pinheiro et al., 2024). Significant differences were determined using the *emmeans* function from the *emmeans* package (Lenth, 2024), and letters were assigned using the *cld* function from the *multcomp* package (Hothorn et al., 2008).

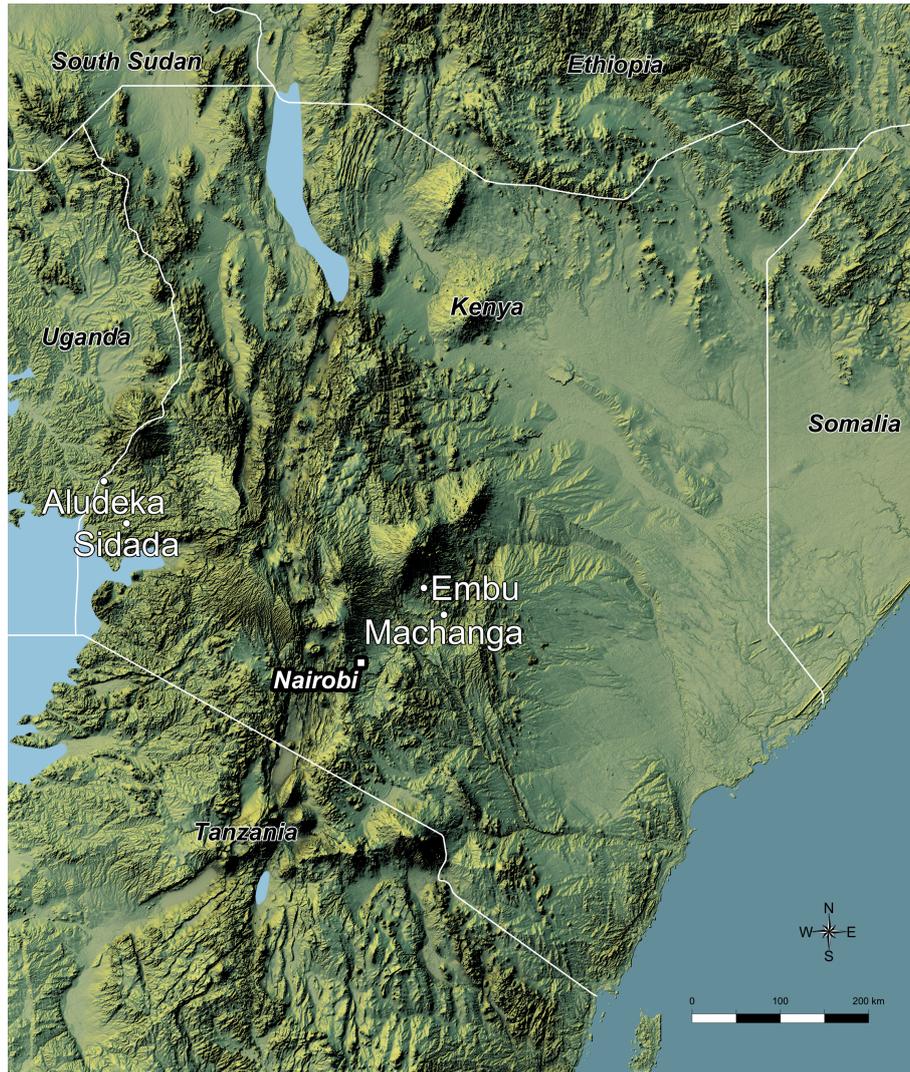
### **S1.2 Statistics for bulk density between treatments at the same site**

To determine if bulk density values were significantly different between treatments of the the same site and depth, a linear mixed effects model was applied in R (using the *lme* function from the *nlme* package in R; Pinheiro et al., 2024). The fixed effects included treatment, depth, and their interaction, while the random effect was the replicate. A variance structure for treatment, depth, and their interaction was included (Pinheiro et al., 2024). Significant differences were determined using the *emmeans* function from the *emmeans* package (Lenth, 2024), and letters were assigned using the *cld* function from the *multcomp* package (Hothorn et al., 2008).

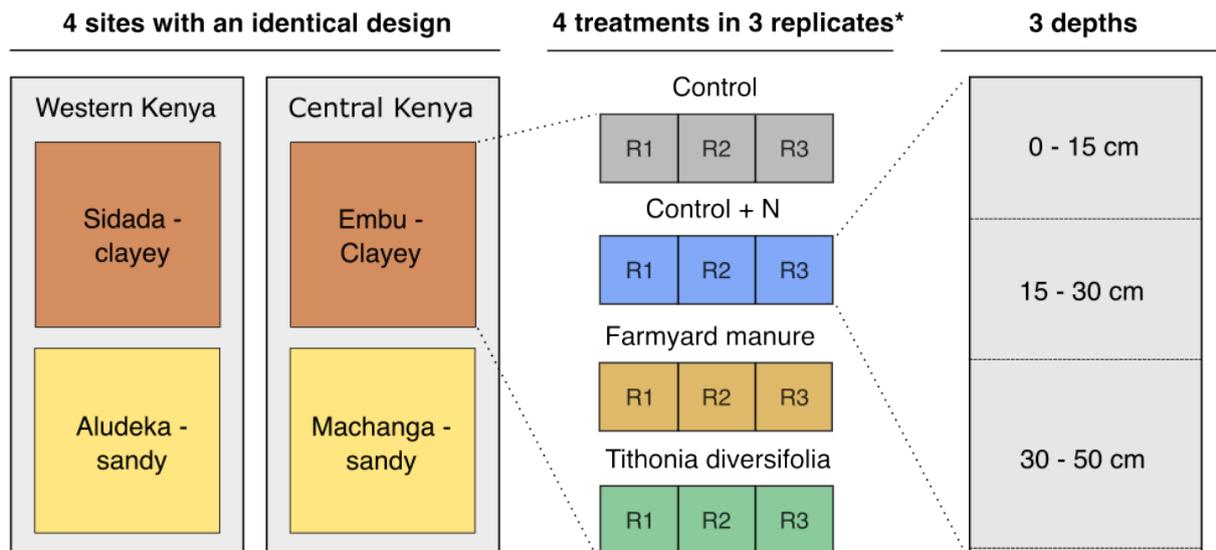
### **S1.3 Statistics for the portion of POC and MAOC of total SOC**

The analysis of significant differences in the portion of POC and MAOC of total SOC in the topsoil (Fig. S15) and subsoil (Fig. S16) was done after fitting a linear mixed effects model (using the *lme* function from the *nlme* package in R; Pinheiro et al., 2024) following the procedures outlined in (Zuur et al., 2009), with site and treatment as fixed effect and nested site/block as random effect. A variance structure (*varIdent*) was added to allow for differences in variance for site in the topsoil, and for site and treatment in the subsoil (Pinheiro et al., 2024). Significant differences were determined using the *emmeans* function from the *emmeans* package (Lenth, 2024), and letters were assigned using the *cld* function from the *multcomp* package (Hothorn et al., 2008).

S2 Supplementary figures

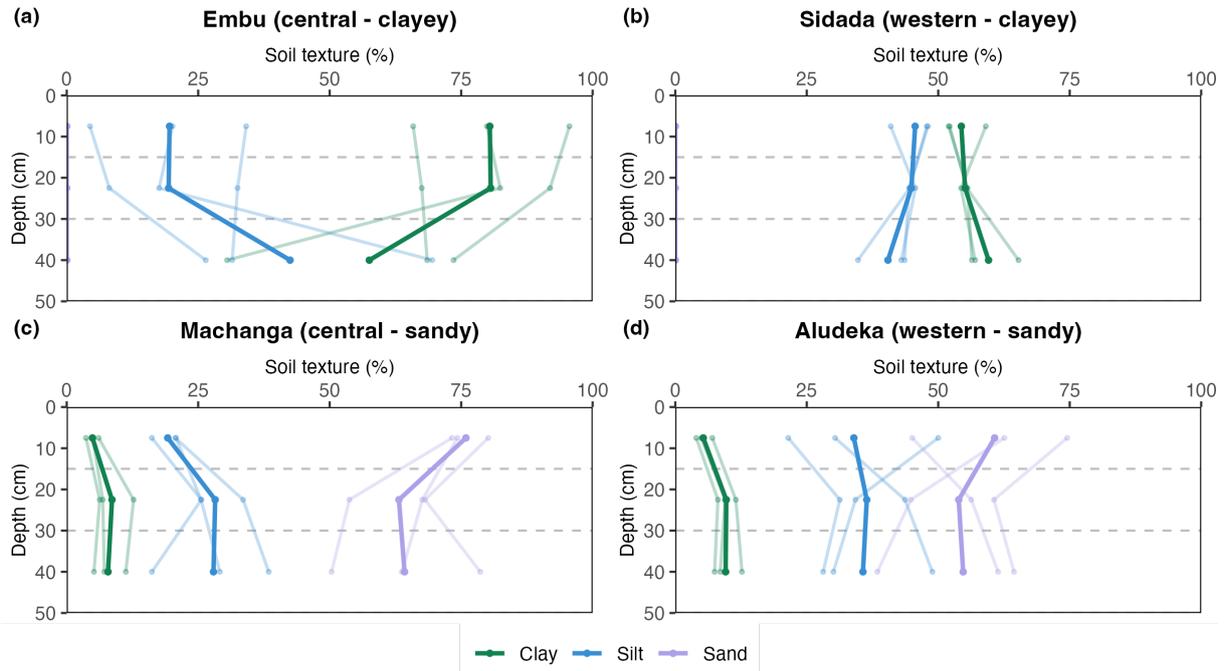


**Figure S1.** Map showing the locations of the fields trials in central (Embu and Machanga) and western (Sidada and Aludeka) Kenya. The map was produced in *R*, using the *elevatr* (Hollister et al., 2023) and *rayshader* (Morgan-Wall, 2024) packages.

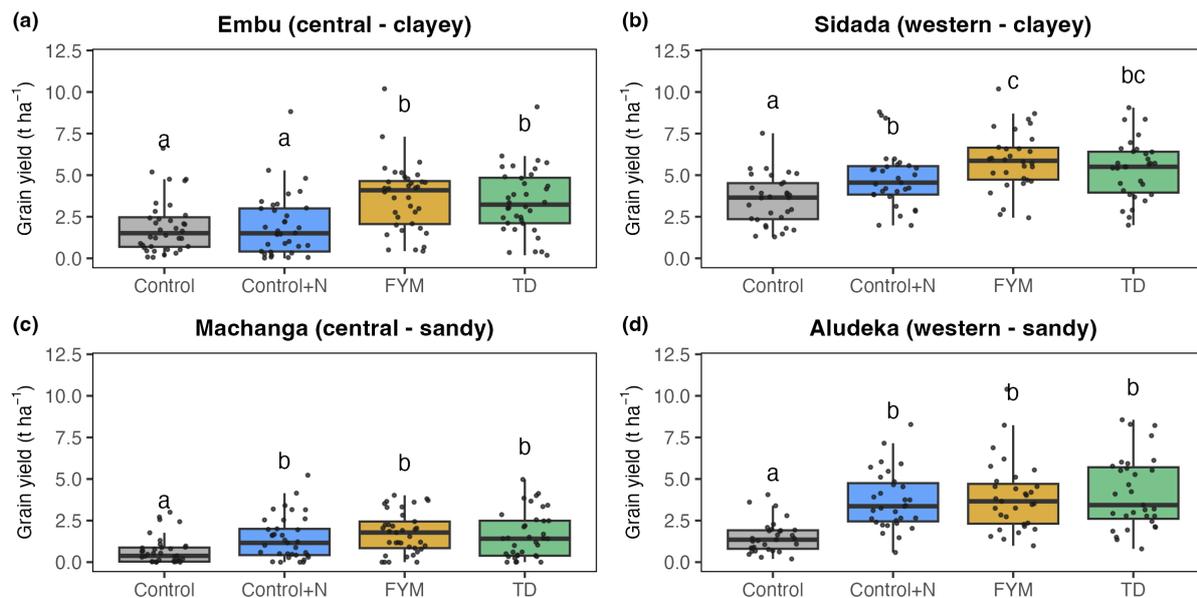


*\*In the field experiments, the replicates were organised in 3 blocks*

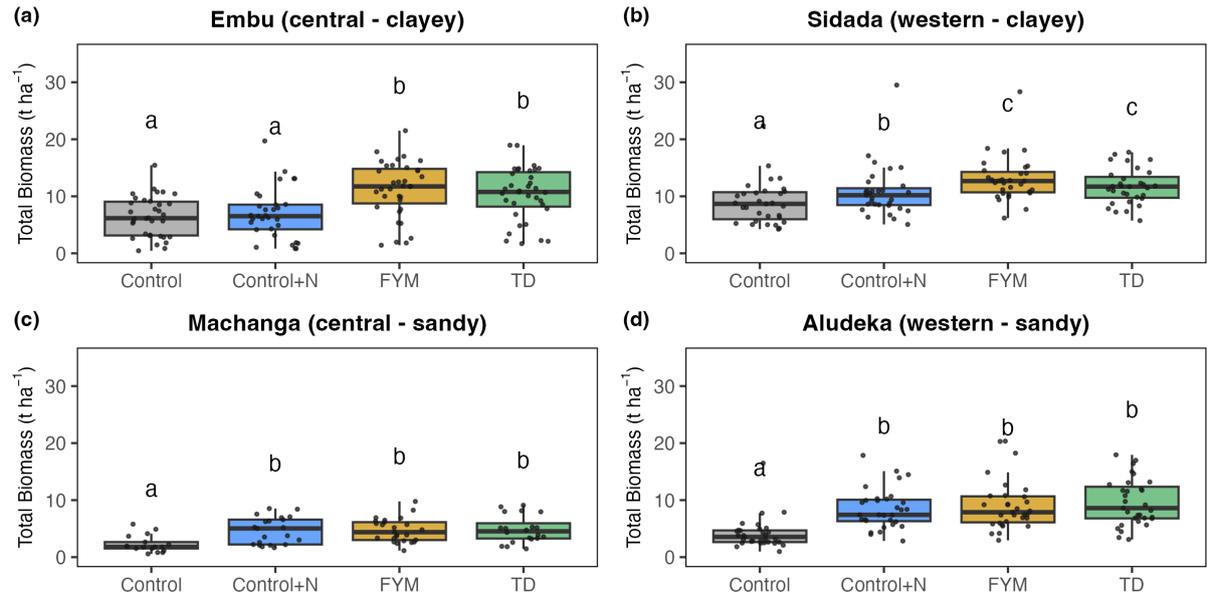
**Figure S2.** Overview of the design of the treatments and samples collected in the present study. A detailed description of the long-term experiments is provided in Laub et al. (2023a) and Laub et al. (2023b), including an overview of how the replicates were distributed over the three blocks.



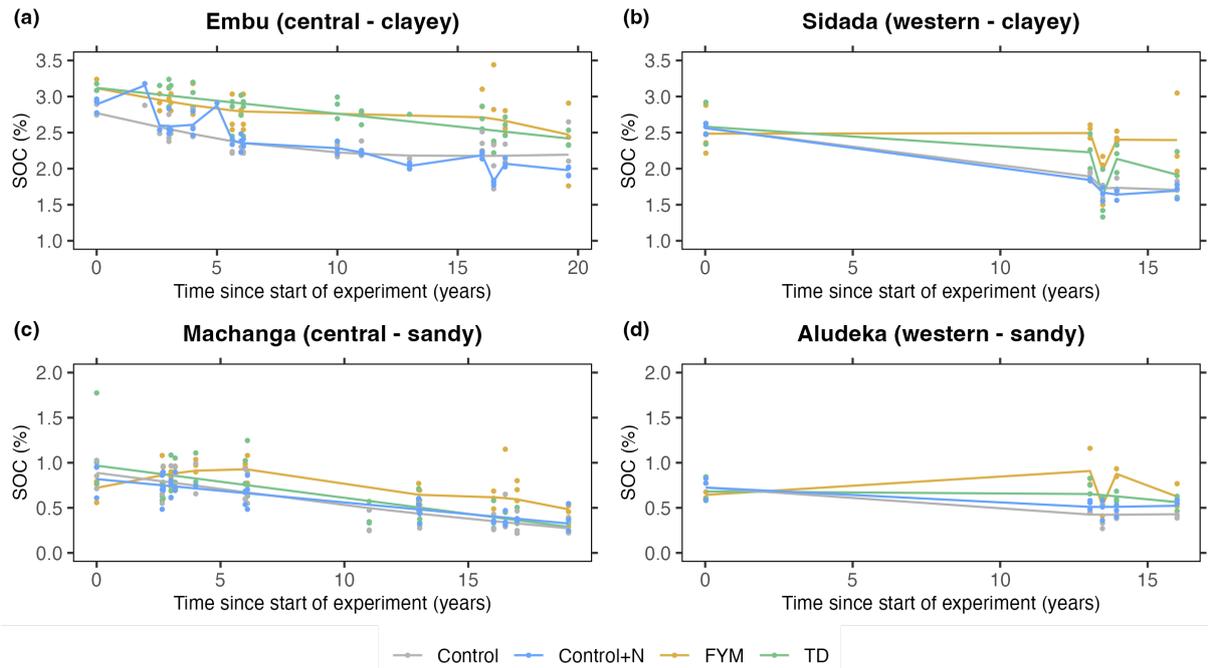
**Figure S3.** Depth profiles of soil texture at the study sites, determined for the control treatments. Transparent points and lines show the individual measurements, the thick lines show the average per depth layer. Vertical lines indicate the depth layers. Note that at the clayey sites (a, b) no sand-sized particles were present.



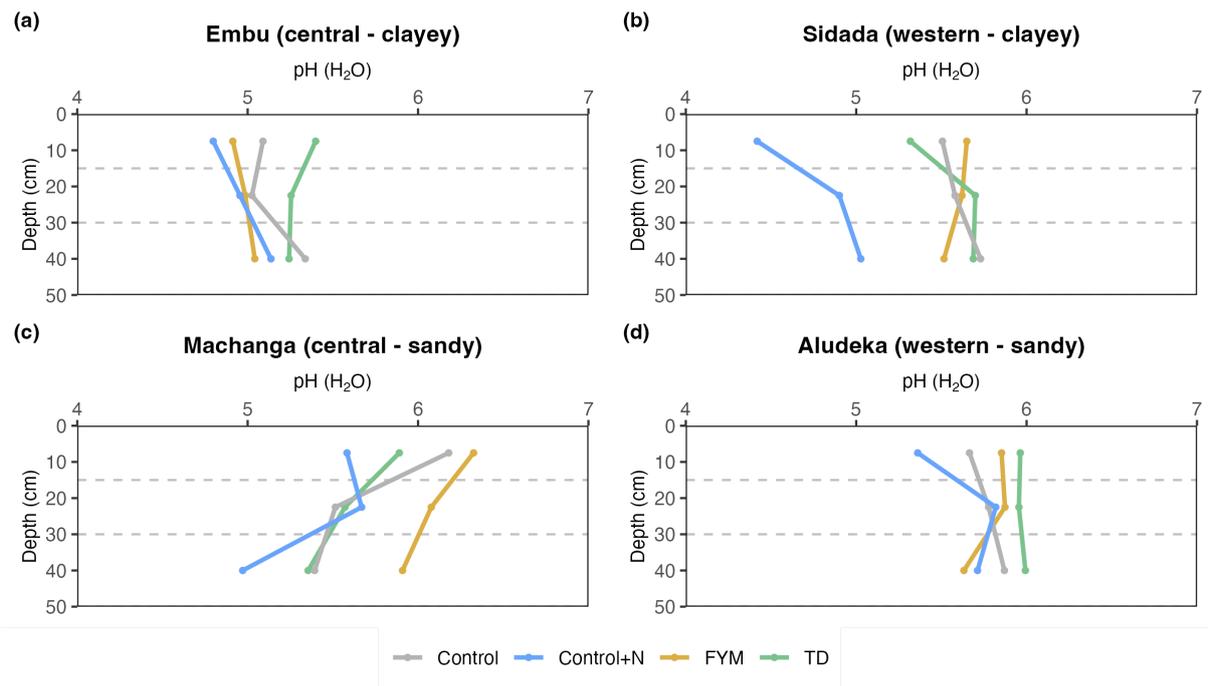
**Figure S4.** Maize grain yield at the different study sites expressed as ton grain ha<sup>-1</sup> per growing season, for the studied treatments (Control is the control treatment, Control+N received 120 t ha<sup>-1</sup> yr<sup>-1</sup> mineral N fertilizer, FYM received 4 t C ha<sup>-1</sup> yr<sup>-1</sup> in the form of farmyard manure and TD received 4 t C ha<sup>-1</sup> yr<sup>-1</sup> in the form of *Tithonia diversifolia*). Treatments within the same site that do not share any letter are significantly different ( $p < 0.05$ ). Total maize biomass production is shown in Figure S5. Source of the data: Laub et al. (2023b).



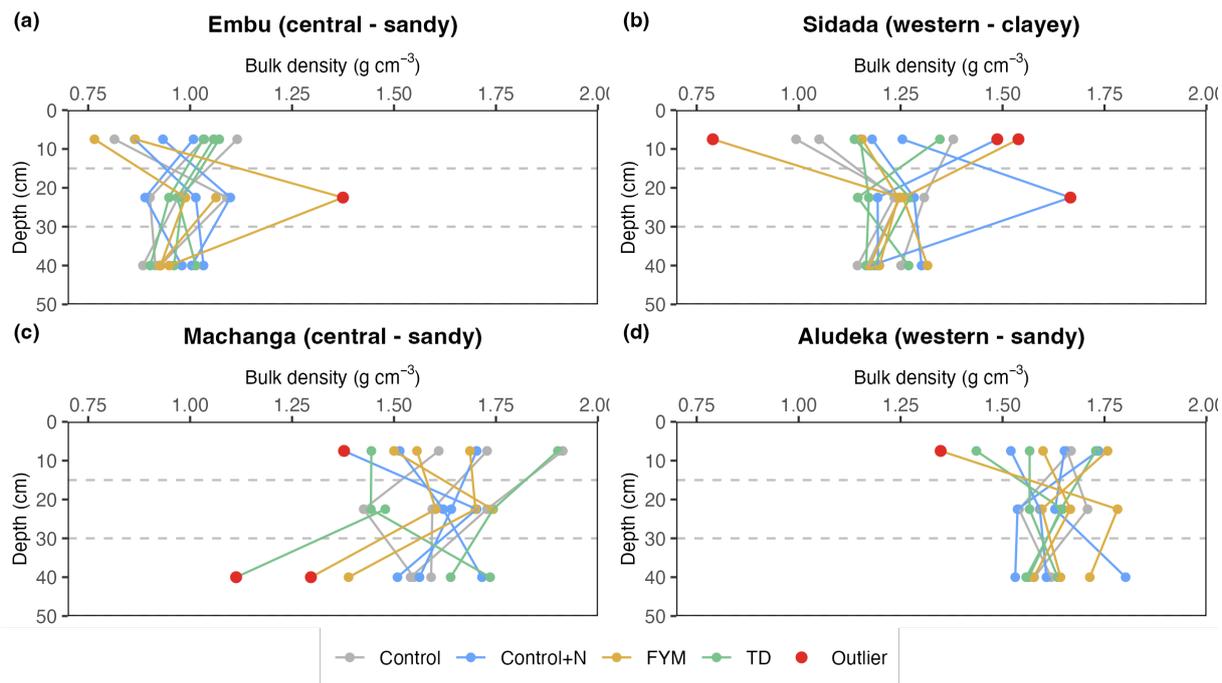
**Figure S5.** Total aboveground biomass production at the different study sites expressed as ton biomass ha<sup>-1</sup> per growing season, for the studied treatments (Control is the control treatment, Control+N received 120 t ha<sup>-1</sup> yr<sup>-1</sup> mineral N fertilizer, FYM received 4 t C ha<sup>-1</sup> yr<sup>-1</sup> in the form of farmyard manure and TD received 4 t C ha<sup>-1</sup> yr<sup>-1</sup> in the form of *Tithonia diversifolia*). Treatments within the same site that do not share any letter are significantly different ( $p < 0.05$ ). Maize yield is shown in Figure S4. Source of the data: Laub et al. (2023b).



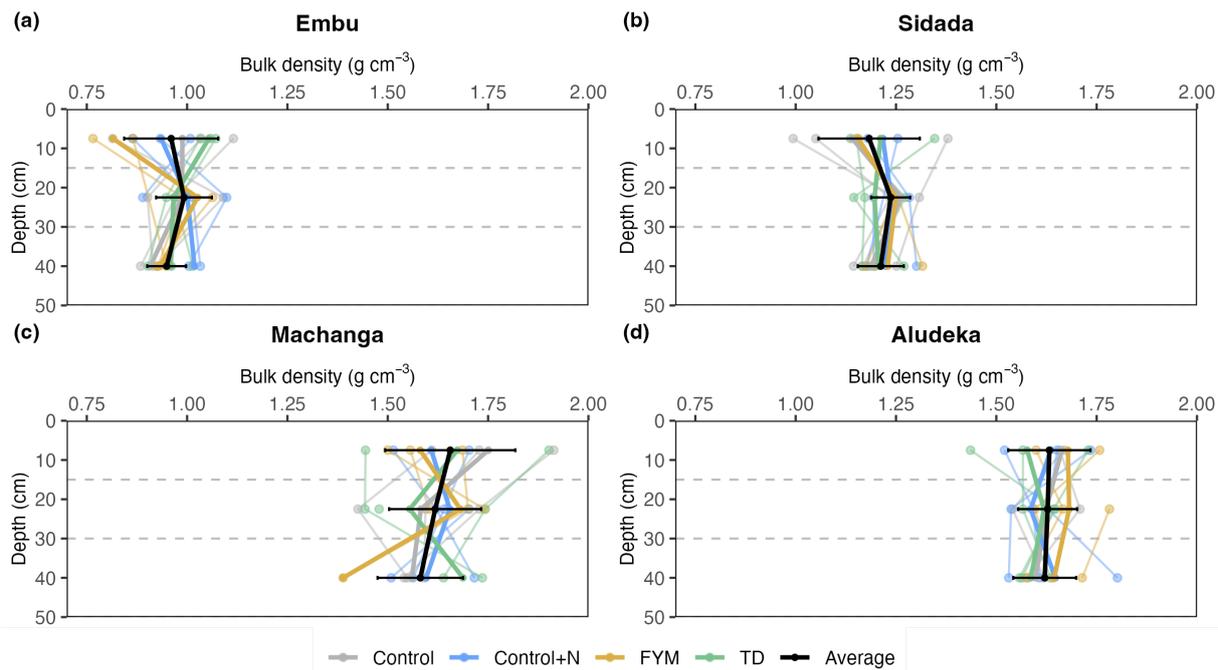
**Figure S6.** Evolution in topsoil OC (%) (0–15 cm) per treatment at the four long-term field trials. Dots represent measurements, while lines denote splines to illustrate the temporal evolution per treatment. Note that in Aludeka and Sidada, regular sampling of the topsoil only occurred 13 years after the initiation of these experiments. A detailed analysis of the evolution of the topsoil OC content of all treatments is described in Laub et al. (2023a).



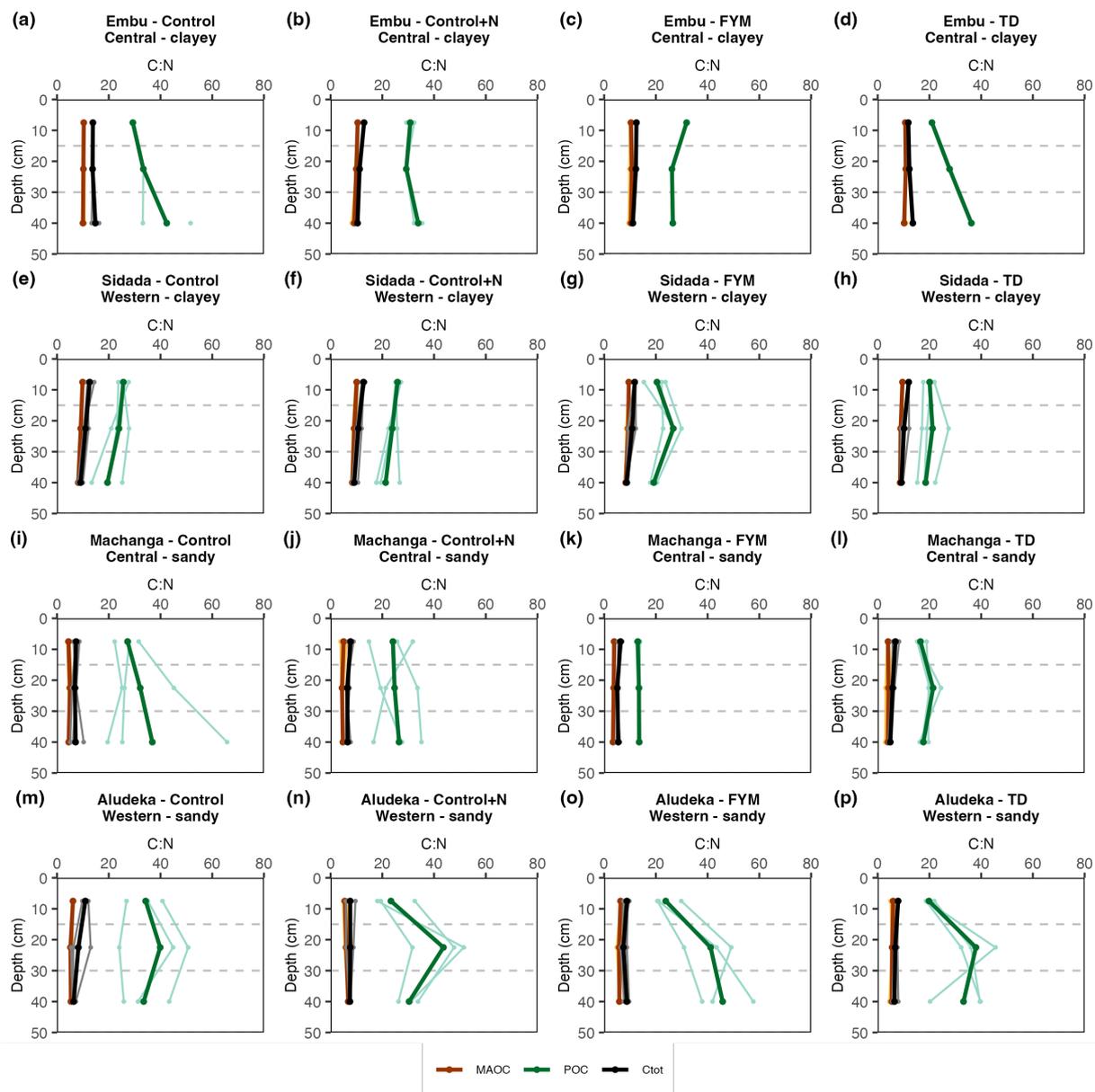
**Figure S7.** Depth profiles of soil  $\text{pH}_{\text{H}_2\text{O}}$  (i.e., the pH measured in water) for the studied treatments.



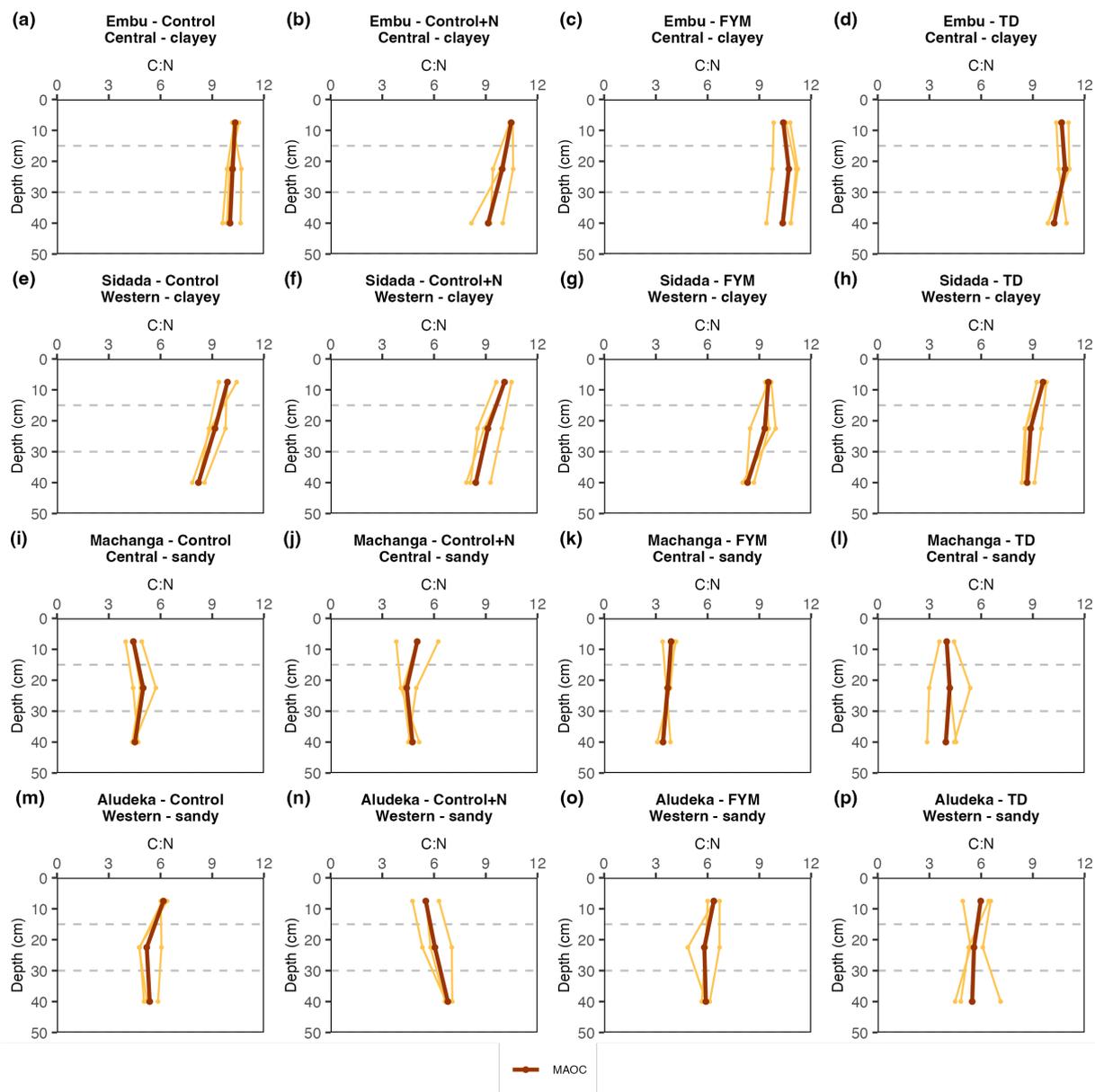
**Figure S8.** Depth profiles of soil bulk density for the individual collected soil samples. Data points identified as outliers are highlighted in red (see Section 2.2 in the manuscript for details).



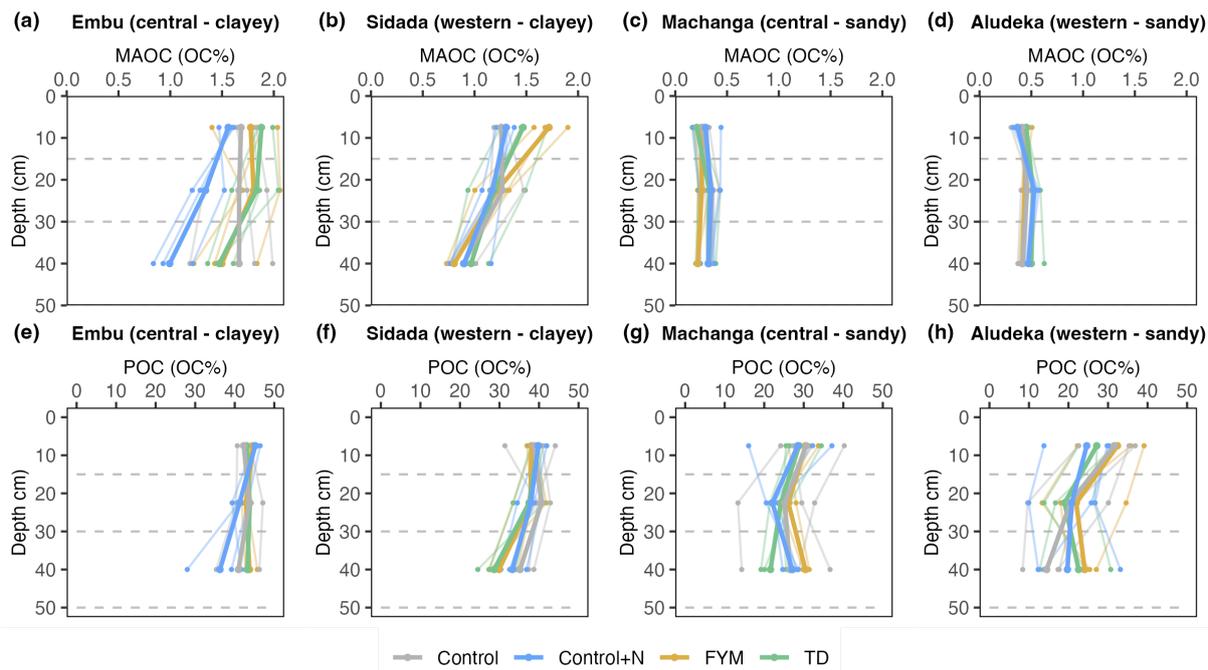
**Figure S9.** Depth profiles of soil bulk density, without outliers (see Figure S8). The thinnest lines show the individual depth profiles, thick colored lines show the averages per treatment, and black lines the average per site. Error bars show the standard deviation.



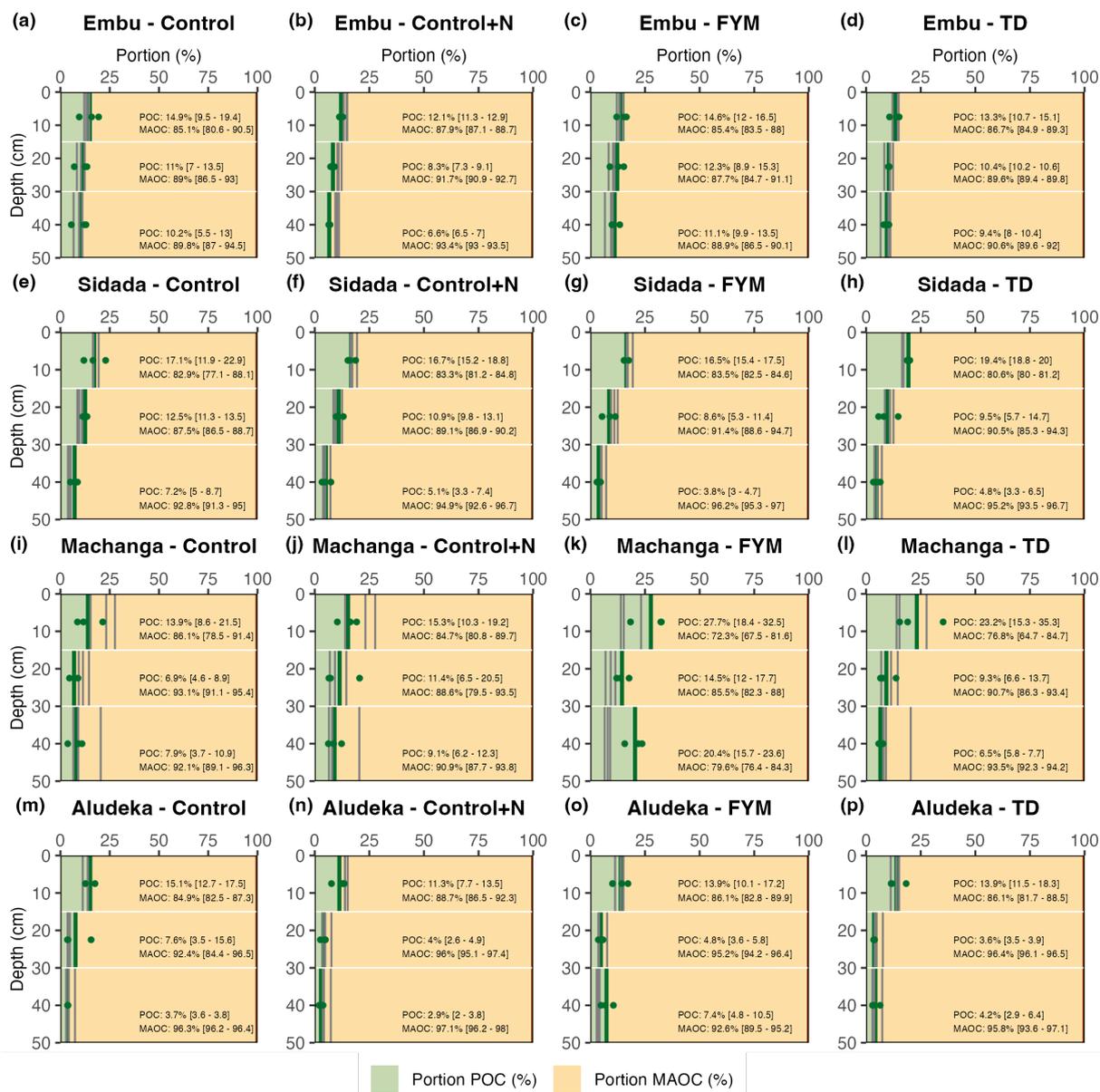
**Figure S10.** Depth profiles of the C:N ratio of mineral-associated OC (MAOC), particulate OC (POC) and total SOC (Ctot). Light lines show the individual depth profiles, dark lines the averages per site.



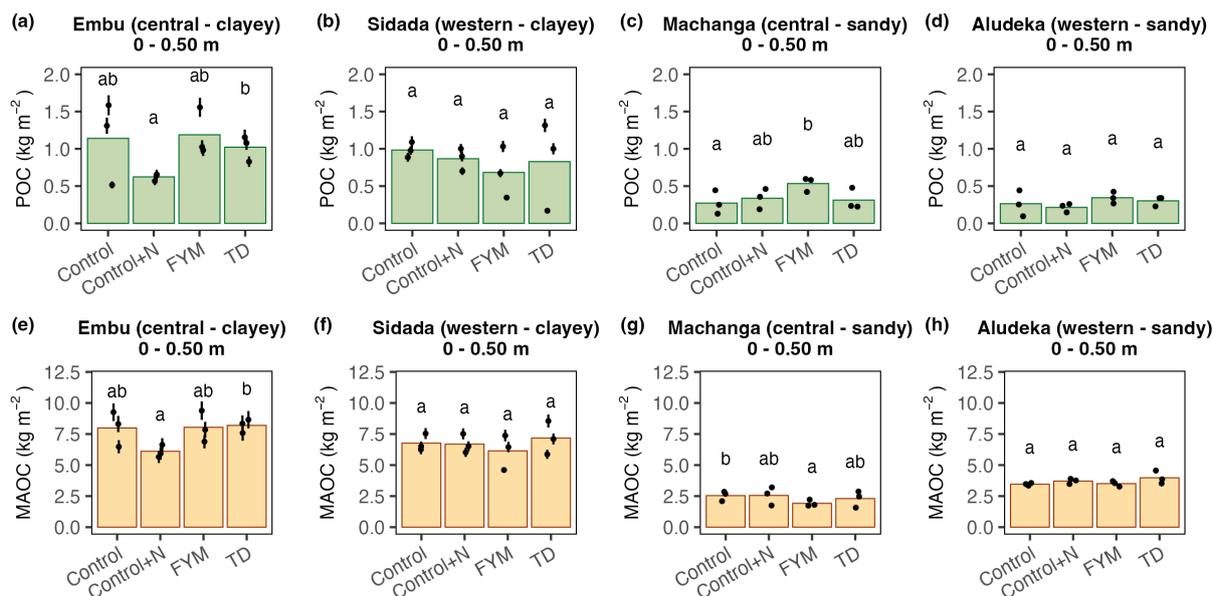
**Figure S11.** Depth profiles of the C:N ratio of mineral-associated OC (MAOC). Light lines show the individual depth profiles, dark lines the averages per site. The data is the same as in Fig. S10, but zoomed in on MAOC.



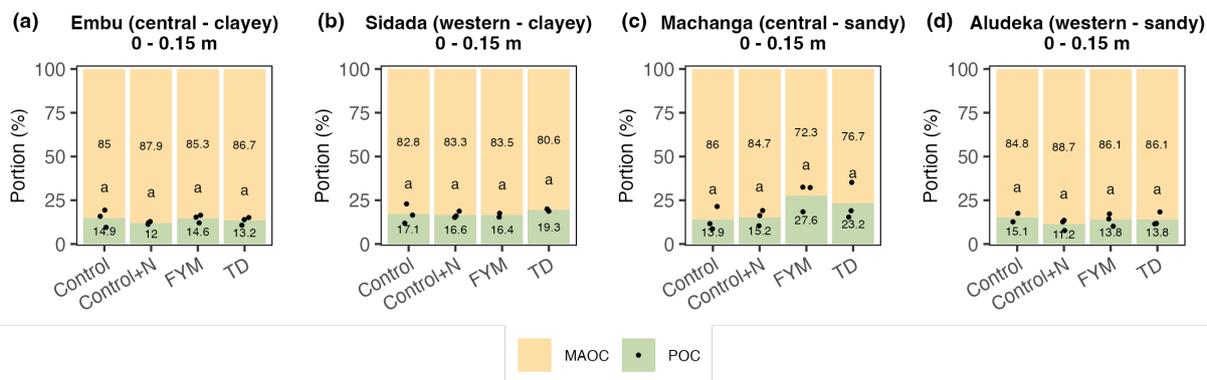
**Figure S12.** The OC content (%) of the MAOC (a - d) and POC (e - h) fractions. Light lines show the individual depth profiles, dark lines the averages per site.



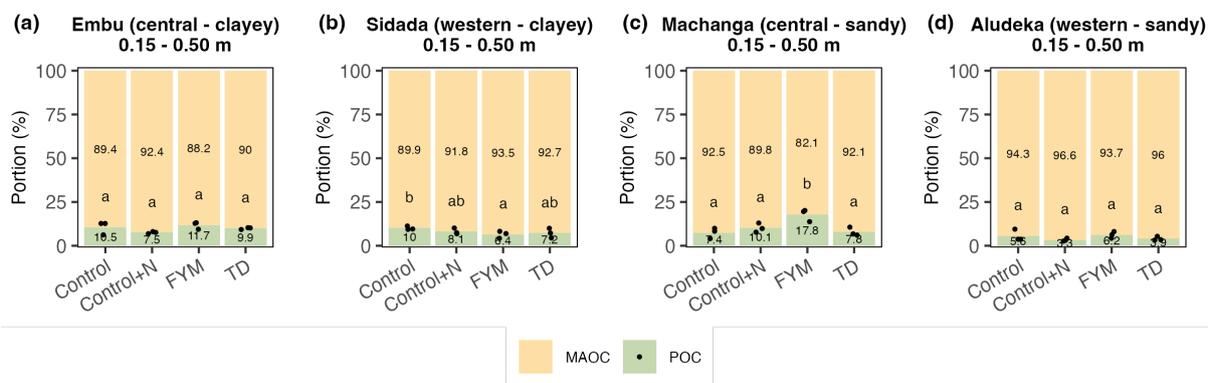
**Figure S13.** Depth profiles of the portion (%) of POC (green) and MAOC (orange) for the different measured depth layers. Dots show the individual measurements. Dark lines show the values for the plot, while the grey lines show the portions for the other plots of the same site and the same depth. The two top rows show results for the clayey sites, the bottom two rows show results for the sandy sites. The numbers in the different depth layers denote the average portion of POC and MAOC in total SOC, with the numbers between brackets being the lowest and highest portion of the replicates.



**Figure S14.** Stocks of POC (top row, a-d) and MAOC (bottom row, e-h) for the soil down to 50 cm depth (expressed as kg OC m<sup>-2</sup> down to 50 cm). Bars show the average stocks, while dots show the individual measurements. Treatments within the site plot that do not share any letter are significantly different (*p* < 0.05). The vertical lines show the uncertainty of individual data points due to variation in bulk density between the three replicates (*n* = 12). No error for the averages (the bars) are shown, due to the low number of replicates (*n*=3).



**Figure S15.** The relative portion POC and MAOC in the plough layer (0-15 cm depth) for the four treatments. Bars show the averages, which are also displayed in numbers, while the black dots show the individual measurements. Treatments within the same plot that do not share any letter are significantly different (*p* < 0.05).



**Figure S16.** The relative portion POC and MAOC below the plough layer (15-50 cm depth) for the four treatments. Bars show the averages, which are also displayed in numbers, while the black dots show the individual measurements. Treatments within the same plot that do not share any letter are significantly different ( $p < 0.05$ ).

## References

- Hollister, J., Shah, T., Nowosad, J., Robitaille, A. L., Beck, M. W., and Johnson, M.: elevatr: Access Elevation Data from Various APIs, <https://doi.org/10.5281/zenodo.8335450>, r package version 0.99.0, 2023.
- Hothorn, T., Bretz, F., and Westfall, P.: Simultaneous Inference in General Parametric Models, *Biometrical Journal*, 50, 346–363, 2008.
- 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., and Six, J.: 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>, publisher: Copernicus GmbH, 2023a.
- Laub, M., Corbeels, M., Mathu Ndungu, S., Mucheru-Muna, M. W., Mugendi, D., Necpalova, M., Van de Broek, M., Waswa, W., Vanlauwe, B., and Six, J.: Combining manure with mineral N fertilizer maintains maize yields: Evidence from four long-term experiments in Kenya, *Field Crops Research*, 291, 108 788, <https://doi.org/10.1016/j.fcr.2022.108788>, 2023b.
- Lenth, R. V.: emmeans: Estimated Marginal Means, aka Least-Squares Means, <https://CRAN.R-project.org/package=emmeans>, r package version 1.10.3, 2024.
- Morgan-Wall, T.: rayshader: Create Maps and Visualize Data in 2D and 3D, <https://www.rayshader.com>, r package version 0.38.1, <https://github.com/tylormorganwall/rayshader>, 2024.
- Pinheiro, J., Bates, D., and R Core Team: nlme: Linear and Nonlinear Mixed Effects Models, <https://CRAN.R-project.org/package=nlme>, r package version 3.1-165, 2024.
- Zuur, A. F., Ieno, E. N., Walker, N., Saveliev, A. A., and Smith, G. M.: Mixed effects models and extensions in ecology with R, *Statistics for Biology and Health*, Springer, New York, NY, ISBN 978-0-387-87457-9 978-0-387-87458-6, <https://doi.org/10.1007/978-0-387-87458-6>, 2009.