1	Mulch application as the	overarching factor	explaining increase	e in soil organic carbon
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- 2 stocks under conservation agriculture in two 8-year-old experiments in Zimbabwe
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### 26 Abstract

Conservation agriculture (CA), combining reduced or no tillage, permanent soil cover and 27 improved rotations, is often promoted as a climate-smart practice. However, our understanding 28 29 about the impact of CA and its respective three principles on top and sub-soil organic carbon (SOC) stocks in low input cropping systems of sub-Saharan Africa is rather limited. The study 30 was conducted at two long-term experimental sites established in 2013 in Zimbabwe. The soil 31 types were abruptic Lixisols at Domboshava Training Centre (DTC) and xanthic Ferralsol at 32 the University of Zimbabwe farm (UZF). Six treatments, replicated four times were 33 investigated: conventional tillage (CT), conventional tillage with rotation (CTR), NT, no-34 tillage with mulch (NTM), no-tillage with rotation (NTR), no-tillage with mulch and rotation 35 (NTMR). Maize (Zea mays L.) was the main crop and treatments with rotation included cowpea 36 (Vigna unguiculata L. Walp.). SOC concentration and bulk density were determined for 37 38 samples taken from the 0-5, 5-10, 10-15, 15-20, 20-30, 30-40, 40-50, 50-75 and 75-100 cm depths. Cumulative organic inputs to the soil were also estimated in all treatments. SOC stocks 39 40 at equivalent soil mass were significantly (p < 0.05) higher under NTM, NTR and NTMR 41 compared to NT and CT in top 5 and 10 cm layers at UZF, while SOC stocks were only significantly higher under NTM and NTMR compared to NT and CT in top 5 cm at DTC. NT 42 alone had a slightly negative impact on top SOC stocks. Cumulative SOC stocks were not 43 significantly different between treatments when considering the whole 100 cm soil profile. Our 44 results showed the overarching role of crop residue mulching in CA cropping systems in 45 enhancing SOC stocks but that this effect was limited to the topsoil. The highest cumulative 46 organic carbon inputs to the soil were observed in NTM treatments at the two sites, and this 47 could probably explain the positive effect on SOC stocks. Our results also showed that the 48 combination of at least two CA principles including mulch is required to increase SOC stocks 49 in these low nitrogen input cropping systems. 50

51 Key words: organic inputs, climate change mitigation, climate-smart agriculture, deep soil
52 organic carbon, sustainable intensification

53

# 54 **1. Introduction**

Soil organic carbon (SOC) is an important determinant of soil fertility, productivity and sustainability, and is a useful indicator of soil quality in tropical agricultural systems where nutrient poor and highly weathered soils are managed with little external inputs (Lal, 1997; Feller and Beare, 1997; Chivenge et al., 2007). Therefore, rebuilding depleted SOC stocks in such soils holds potential to contribute to climate change mitigation (Bossio et al., 2020; Minasny et al., 2017; Swanepoel et al., 2016) through sustainable management of agricultural soils (Paustian et al., 2016; Dignac et al., 2017).

Conservation agriculture (CA), based on minimum soil disturbance, crop residue retention and 62 crop rotation, has been known to improve surface SOC, with beneficial effects on soil 63 functioning such as improved water infiltration (Thierfelder and Wall, 2012, 2009) and better 64 aggregate stability (Six et al., 1999; Thierfelder and Wall, 2012). The potential of CA to 65 increase SOC stocks and thereby mitigate climate change has, however, been much debated 66 67 (Corbeels et al., 2020a) but the general understanding is that, this potential is relatively low 68 (Du et al., 2017; Powlson et al., 2014, 2016; Cheesman et al., 2016; Corbeels et al., 2020a). In fact, soil C storage has often been over-estimated for CA due to shallow soil sampling. 69 Compared to conventional tillage systems, no-tillage redistributes SOC in the soil profile, with 70 higher concentrations in the topsoil but potentially lower concentrations below, which can 71 result in no differences in whole profile SOC stocks between no-tillage and conventional tillage 72 (Angers and Eriksen-Hamel, 2008). However, this lack of significant differences in many 73

studies assessing whole profile SOC stocks suffer from not enough statistical power to
accurately assess the potential significant SOC changes (Kravchenko and Robertson, 2011).

CA can potentially build SOC in deeper soil layers from e.g. the use of cover crops with more 76 extended root systems (Kell, 2011; Thorup-Kristensen et al., 2020; Yang et al., 2023). 77 However, proper soil sampling strategies, to account for both topsoil and subsoil (> 30 cm 78 depth) SOC stocks must, therefore, be prioritized. Soil sampling has often been limited to the 79 80 top soil plough layer (0-30 cm) in the past two decades (Dube et al., 2012; Powlson et al., 2016; Patra et al., 2019; Yost and Hartemink, 2020), where SOC concentrations, root densities 81 (Chikowo et al., 2003) and microbial activities (Mtambanengwe et al., 2004) are generally 82 83 largest (Rumpel et al., 2012) and which is the minimum default soil depth recommended by the Intergovernmental Panel for Climate Change (IPCC, 2019). In a meta-analysis of SOC 84 stocks in the top 1 m of soils, Balesdent et al. (2018) found that soils below 0.3 m contain on 85 86 average 47 % of total SOC stock in the 1 m soil depth and accounts for 19 % of the SOC that has been recently incorporated. Therefore, focusing on topsoil only, could underestimate the 87 potential of agricultural management practices to store SOC (Cardinael et al., 2015). In turn, 88 this can give wrong conclusions on the climate change mitigation potential of agricultural 89 90 management practices.

There have been many studies on the effects of CA on crop productivity and soil health benefits (Corbeels, Naudin, et al., 2020; Kimaro et al., 2016; Mhlanga et al., 2022a; Swanepoel et al., 2018; Thierfelder et al., 2015, 2017; Thierfelder & Mhlanga, 2022), and other studies have fuelled the debate on CA practicality and adoption in SSA (Giller et al., 2009, 2015; Kassam et al., 2019). However, the effects of CA on SOC dynamics has not been widely investigated in SSA. Thierfelder et al., (2017) have alluded to the fact that, data on climate change mitigation potential of CA in southern Africa is scanty hence the need for more research to better quantify the mitigation effects of CA as a climate-smart technology. It has also been observed that
depending on the socio-economic and biophysical conditions, farmers may find it easier to
adopt certain CA principles and/or their different combinations (Mbanyele et al., 2021;
Baudron et al., 2012), although this also opened up new debates (Thierfelder et al., 2018).
Therefore, in this study, the focus was on the individual versus combined effects of CA
principles (no-tillage, crop residue retention, crop rotation) on SOC stocks.

As changes in SOC stocks take time to be detected, long-term experiments are crucial, but are 104 rare, especially in Africa (Bationo et al., 2013; Cardinael et al., 2022; Powlson et al., 2016; 105 106 Thierfelder and Mhlanga, 2022). This study was conducted on two long-term experiments established in 2013 in Zimbabwe. We hypothesized that the full combination of CA 107 components would be associated with higher increases of in SOC stocks than adoption of only 108 109 one component., and that this This increase in SOC stocks would could mainly be due to 110 increased C inputs to the soil, especially under and minimum soil disturbance. However, C 111 inputs due to crop rotation might could be indirect and we therefore hypothesise that the through 112 increased crop productivity of the crops is enhanced due to reduction on biotic pressure (pests 113 and diseases), and therefore C inputs to the soil might be increased too. Secondly, cerealstoo. 114 Cereals, in a cereals-legumes rotations tend tomay benefit from added soil nitrogen through 115 biological nitrogen fixation by from the preceding legume crop hence enhancing more their 116 productivity. Crop diversification, on the other hand, can enhance soil biological processes by increasing the diversity and/or abundance of microfauna like mycorrhizae. This, in turn, 117 improves aggregate stability and offers physical protection for SOC. The third hypothesis is 118 119 that crop diversification enhances soil biological processes via different root systems, and enhanced microfauna diversity and/or abundance (e.g. mycorrhizae) that could improve 120 121 aggregate stability and therefore physical protection of soil carbon. Lastly, high quality

residues (from the legume crop) have been shown to be preferentially stabilized in the soil due to a higher carbon use efficiency of soil microbes (Cotrufo et al., 2013; Kopittke et al., 2018).

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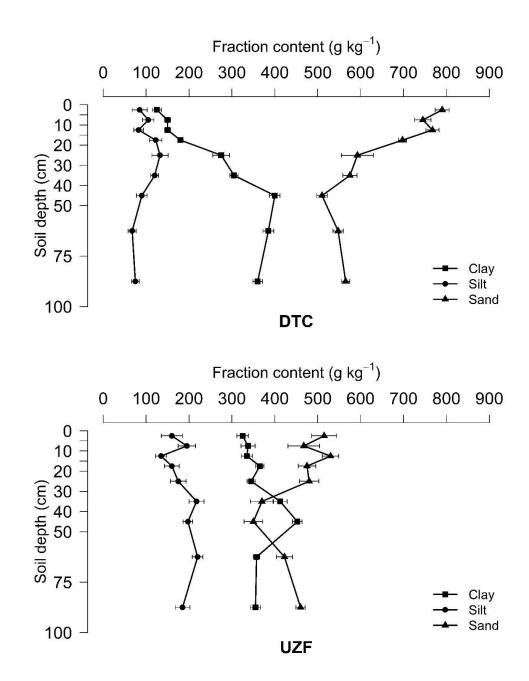
# 125 **2. Materials and methods**

### 126 **2.1 Study sites**

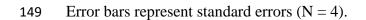
The study was conducted at two long-term experimental sites established in November 2013 127 128 by CIMMYT. The site at the University of Zimbabwe Farm (UZF) is located about 12 km north of Harare city centre (31° 00' 48" E; 17° 42' 24" S), while the site at the Domboshava Training 129 Centre (DTC) is located about 30 km north-east of Harare (31° 07' 33" E; 17° 35' 17" S). UZF 130 soils are dolerite-derived xanthic Ferralsols (FAO classification) and are medium-textured 131 sandy clay loams (34 % clay) in the top 20 cm with a subsoil (20-40 cm) of slightly higher clay 132 133 content (38 %). DTC soils are granite-derived abruptic Lixisols (FAO classification) and are light-textured sandy loams (15 % clay) in the 0-20 cm layer, overlying abruptly a heavier-134 textured subsoil (20-40 cm) of 30 % clay (Figure 1). 135

The two study sites have a sub-tropical climate with cool, dry winters and hot, wet summers 136 with mean annual minimum and maximum temperatures of  $12^{\circ}C$  and  $25^{\circ}C$ , respectively 137 138 (Mapanda et al., 2010). The rainy season starts in November and tails off in March with a mean annual rainfall of 826 and 814 mm at UZF and DTC, respectively (Mhlanga, et al., 2022b). 139 Cumulative seasonal rainfall in 2019/20 (474 mm) was almost half of rainfall received in the 140 141 2020/21 season (932 mm) at DTC (Shumba et al., 2023b). At UZF, cumulative seasonal rainfall was 551 mm and 637 mm in the 2019/20 and 2020/21 cropping seasons. On average, minimum 142 and maximum temperatures were 16.9 and 28.1 °C in 2019/20 and 15.5 and 27.2 °C in 2020/21 143

at DTC and 15.5 and 28.6 °C in 2019/20 and 15.3 and 27.5 °C in 2020/21 cropping season at
UZF.



**Figure 1**. Soil texture to 1 m soil depth at the DTC (top) and UZF (bottom) sites in Zimbabwe.



### 151 **2.2** Experimental treatments and crop management

Two identical experiments were set up at the study sites and treatments were maintained every season since November 2013. The experiments were set up in a randomised complete block design (RCBD) with eight treatments replicated in four blocks. However, in this study we investigated only six of these treatments. All crop residues were removed soon after harvesting in all treatments, stored and then applied prior to planting in treatments with mulch. The six treatments in our study were:

i. Conventional tillage (CT) – land preparation was done through digging with a hand hoe
and maize (*Zea mays* L.) was sown as a sole crop in rip lines that were created
afterwards using an animal-drawn Magoye ripper (a traditional plough with the
mouldboard replaced with a ripper tine) at DTC, and in planting basins (approximately
10 cm diameter and 10 cm depth) created using a hand hoe at UZF.

- ii. Conventional tillage with rotation (CTR) land preparation was done as in the CT
  treatment and maize was rotated with cowpea (*Vigna unguiculata* L.).
- 165 iii. No-tillage (NT) sole maize was sown in rip lines created using an animal-drawn
  166 Magoye ripper (no further soil disturbance was done) at DTC, and in planting basins
  167 (approximately 15 cm diameter and 15 cm depth) created using a hand hoe at UZF.
- iv. No-tillage with mulch (NTM) maize was sown as in the NT treatment and maize
   residues from the previous season were applied on the soil surface between maize
   rows at planting at a rate of 2.5 t DM ha<sup>-1</sup>.
- 171 v. No-tillage with rotation (NTR) maize was sown in rip lines and rotated with cowpea.
- vi. No-tillage with mulch and rotation (NTMR) maize was sown in rip lines and rotated
  with cowpea and maize residues were applied on the soil surface between maize rows
  at planting at a rate of 2.5 t DM ha<sup>-1</sup>.

175 Crop residues were removed every year after harvest and weighed in again to maintain the 176 exact 2.5 t ha<sup>-1</sup> residue weight year after year. There was a total of 24 plots at each site which 177 were 6 m wide and 12 m long (72 m<sup>2</sup>). Treatments with rotation (CTR, NTR, NTMR) were 178 split into 6 m wide and 6 m long (36 m<sup>2</sup>) subplots where maize and cowpea were grown 179 interchangeably every season (maize was sown on one side of the plot while cowpea on the 180 other side).

The inter-row spacing was 90 cm and 45 cm for maize and cowpea, respectively, and the in-181 row spacing was 25 cm for both crops which translated to 44,444 and 88,888 plants ha<sup>-1</sup>, 182 respectively. Three seeds were planted per planting station and thinned to one after emergence. 183 Basal dressing of nitrogen (N), phosphorus (P) and potassium (K) was applied within 5 cm of 184 the seeds in the form of compound fertilizer for both maize and cowpea at 11.6 kg N ha<sup>-1</sup>, 10.6 185 kg P ha<sup>-1</sup> and 9.6 kg K ha<sup>-1</sup>, respectively. Nitrogen top dressing to maize only, was applied at 186 4 and 8 weeks after emergence (WAE) in two equal splits of 23.1 kg N ha<sup>-1</sup> each, as ammonium 187 nitrate when soil moisture was adequate. However, in the 2019/20 cropping season at both sites 188 and in the 2020/21 cropping season at UZF, the first N top dressing was delayed by an average 189 of 4.5 and 2.0 weeks, respectively, due to mid-season dry spells. Ammonium nitrate was side 190 191 dressed within 5 cm of the maize stems. Glyphosate [N-(phosphono-methyl) glycine], a preemergent non-selective herbicide was applied at 1.025 L active ingredient ha<sup>-1</sup> soon after 192 193 sowing to control weeds. This was followed by manual hoe weeding whenever weeds reached a maximum of 10 cm height or 10 cm in diameter for stoloniferous weeds to achieve a weed 194 clean field. More details about the experiment can be found in Shumba et al., (2023b) and 195 Mhlanga et al., (2022a). 196

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# 198 **2.3** Soil sampling for bulk density determination and soil organic carbon analysis

Soil sampling was done in May 2021 at both sites. For each treatment and replicate, two 199 sampling points in the maize rows and two sampling points in the middle of the inter-rows 200 were randomly selected. The two samples from the rows were pooled into one sample per 201 depth, similarly to the two samples taken in the inter-rows. The following nine depth 202 increments were considered for both SOC and bulk density (BD) measurements: 0-5, 5-10, 10-203 15, 15-20, 20-30, 30-40, 40-50, 50-75 and 75-100 cm. Undisturbed soil samples using a metal 204 205 cylinder (5 cm diameter and 5 cm height) were taken from the following depth ranges 0-5, 5-10, 10-15, 15-20 and 20-30 cm for both SOC and BD measurements. A motorized, hand-held 206 207 soil corer, with an inside diameter of 10 cm, was used to take samples for the 30-40, 40-50, 50-75 and 75-100 cm depths for SOC analysis from the same positions where undisturbed samples 208 were taken. As no significant differences in BD were found below 20 cm between the different 209 210 treatments at the two sites (see results section) and to avoid too much destruction of the experimental plots, two soil pits were opened at the edges of the experimental plots (also 211 cropped with maize since 2013) at each site to take BD samples for the 30-40, 40-50, 50-75 212 and 75-100 cm depths. As a result, BD below 30 cm depth was assumed the same across the 213 214 treatments.

Soil samples were crumbled and fresh weight was determined using a field scale. Soil moisture 215 216 was determined on a sub-sample by drying it in an oven at 105°C for 48 hours. All samples were then air-dried and sieved through a 2 mm sieve to determine the mass proportion of coarse 217 soil (> 2 mm) as a fraction of the whole sample. Bulk density (BD) was determined by dividing 218 219 the dry mass of soil by the volume of the cylinder. Subsamples from the  $\leq 2$  mm soil fraction were grinded to  $< 200 \,\mu$ m for SOC analysis. SOC was analysed in the ISO9001:2015-certified 220 IRD LAMA's laboratory in Dakar by dry combustion on 100-mg aliquots of soil (ground to < 221 200 µm) using a CHN elemental analyser (Thermo Finnigan Flash EA1112, Milan, Italy). 222

### 224 **2.4 Soil organic carbon stocks calculation**

225 The mass proportion of the coarse fraction (> 2 mm) was removed to calculate SOC stocks. The equivalent soil mass (ESM) approach was used to determine SOC stocks to avoid 226 systematic bias in SOC calculation when using the fixed depth method (Ellert and Bettany, 227 1995; Wendt and Hauser, 2013; von Haden et al., 2020). We defined reference soil mass 228 profiles for each site, based on the lowest cumulative soil mass obtained for each replicate. For 229 230 these references, cumulative soil mass layers were 0-650, 650-1340, 1340-2060, 2060-4160, 4160-5590, 5590-7040, 7040-10550, 10550-13770 Mg soil ha<sup>-1</sup> at DTC and 0-460, 460-870, 231 870-1330, 1330-1840, 1840-2760, 2760-4030, 4030-5300, 5300-8190, 8190-11050 Mg soil 232 ha<sup>-1</sup> at UZF, which roughly corresponded to soil depth layers of 0-5, 5-10, 10-15, 15-20, 20-233 30, 30-40, 40-50, 50-75, 75-100 cm, respectively. SOC stocks were calculated on the basis of 234 the same soil mass as the reference profile but different soil depth layers which varied by < 1.5235 236 and < 0.6 cm at DTC and UZF, respectively. As mulch was only applied between maize rows, and fertilizer was only applied on maize rows, it was estimated that the row and interrow space 237 represented 22 and 78 % respectively, hence SOC calculations were weighted accordingly 238 (Shumba et al., 2023b). Change in cumulative SOC stock between treatments for a given soil 239 depth was determined using the CT treatment as the reference treatment: 240

241  $\Delta SOC \ stock = SOC \ stock_{treatment(i)} - SOC \ stock_{CT(i)}$ , (Equation 1)

where SOC stock<sub>treatment</sub> is the cumulative SOC stock per treatment (CTR, NT, NTM, NRT, NTMR) at a given soil layer and (i) representing 0-5, 0-10, 0-15, 0-20, 0-30, 0-40, 0-50, 50-75, 75-100 cm. SOC accumulation or loss rates (kg C ha<sup>-1</sup> yr<sup>-1</sup>) were calculated by dividing the change in stocks by the number of years between the establishment of the experiment and the time of soil sampling (8 years):

248 SOC accumulation/loss rate = 
$$\frac{\Delta SOC \ stocks}{8} \times 1000$$
, (Equation 2)

249

# 250 **2.5 Estimation of organic carbon inputs to the soil**

Maize and cowpea yield and aboveground biomass were measured since the inception of the 251 experiment, except for cowpea during the 2013/14 season. This data gap was filled by using 252 the average cowpea yield and aboveground biomass values across seasons (from 2013/14 to 253 254 2020/21). We assumed that 5 % of the maize aboveground vegetative biomass remained in the field because maize stalk slashing at harvesting did not remove the whole stem. A root:shoot 255 ratio of 0.16 and 0.06 for maize and cowpea, respectively (Amos and Walters, 2006; Kahn and 256 257 Schroeder, 1999; Kimiti, 2011) was used to estimate the contribution of roots to organic C 258 inputs to the soils. Organic C input contribution from weeds was assumed insignificant since there was effective control of weeds through the use of pre-emergence herbicide (glyphosate) 259 and timely manual weeding throughout the cropping season. We also assumed that the relative 260 amounts of organic C transferred through rhizodeposition was the same for maize and cowpea 261 (i.e. 0.45 x root C biomass (Balesdent et al., 2011) and that the organic C content of all plant 262 parts was 430 g kg<sup>-1</sup> (Ma et al., 2018). Cumulative organic C inputs to the soil were then 263 estimated for each treatment (Cardinael et al., 2022). 264

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### 266 **2.6 Data analysis**

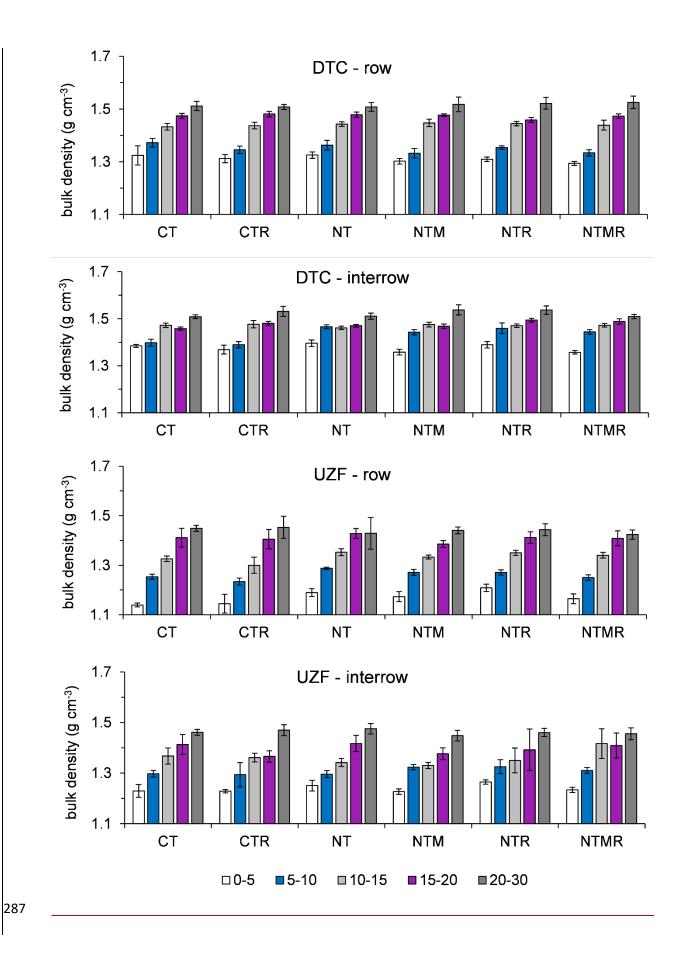
The full dataset is available in the CIRAD repository (Shumba et al., 2023a). Statistical analyses were performed using R software, version 4.0.0 (R Core Team 2020). Normality was tested by the Kolmogorov-Smirnov test. After confirming that data were normally distributed, analyses of variance (ANOVA) was carried out to establish any significant treatment effects on BD, SOC concentration, and SOC stock. Separation of means was done using the post hoc Tukey test at 5 % significance level using the *emmeans* function from the *emmeans* package (Bolker et al., 2009).

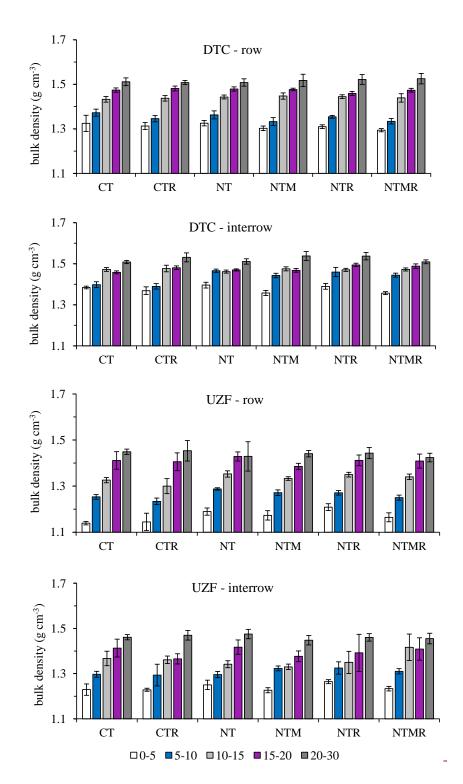
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### 275 **3. Results**

### 276 **3.1 Soil bulk density**

The different cropping systems (CT, CTR, NT, NTM, NTR, NTMR) had no significant (p > 277 0.05) effect on BD across all soil depths except in the 5-10 cm depth in the inter-row at DTC 278 (Figure 2) where BD was on average 5 % lower in the conventional tillage treatments (CT, CTR) 279 than in the no-tillage treatments (NT, NTR). However, soil depth and location (row or inter-row), 280 281 and the soil depth x location interaction had significant (p < 0.001) effects on BD. In the tillage layer (0-15 cm), BD was at least 2 % higher in the inter-rows than in the rows at both sites. In 282 the deeper soil layer (15 - 30 cm), there were no significant (p > 0.05) differences. BD for depths 283 below 30 cm were the same across treatments since it was determined from pits outside the 284 experiment. It ranged between 1.47 - 1.51 and 1.47 - 1.49 g cm<sup>-3</sup> (Table S1) in the subsoil (30 -285 100 cm layers) at DTC and UZF, respectively. 286





**Figure 2**. Top soil bulk density (0-30 cm) at the Domboshava Training Centre (DTC) and University of Zimbabwe Farm (UZF) experimental sites in Zimbabwe. CT: conventional tillage, CTR: conventional tillage with rotation, NT: no-tillage, NTM: no-tillage with mulch, NTR: notillage with rotation, NTMR: no-tillage with mulch and rotation. Error bars represent standard errors (N = 4).

# **3.2 SOC concentration**

299

SOC concentration decreased significantly (p < 0.001) with soil depth (Figure 3, Table S2) and

- was highest in the surface soil (0-5 cm) for all treatments (Table S2). There were significant
- treatment effects in the 0-5 cm (p = 0.001) and 5-10 cm (p = 0.005) soil layers at DTC and in
- the 0-5 cm layer (p < 0.001) only, at UZF. NTM had significantly (p < 0.05) higher SOC
- soil layer at both sites (Figure 3); the increase in SOC concentration ranged between 31 to 46

concentration compared to conventional tillage treatments (CT, CTR) and NT in the 0-5 cm

- 301 % and 14 to 22 % at DTC and UZF, respectively. However, SOC concentration in NTM was
- equal (p > 0.05) to NTR and NTMR treatments at both sites.
- 303 In the 5-10 cm soil layer of DTC, SOC concentrations in NTM and NTR were at least 19 %
- higher (p = 0.005) than in NT and CT (Table S2). There were no significant (p > 0.05) treatment
- effects on SOC concentration below 10 cm soil depth at DTC and below 5 cm depth at UZF.

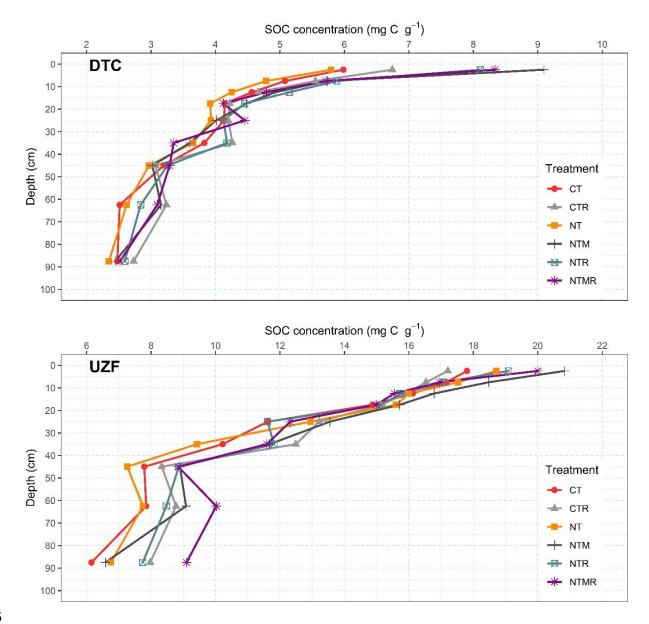




Figure 3. Soil depth distribution of organic carbon concentration for the different experimental
treatments at the Domboshava Training Centre (DTC) and University of Zimbabwe Farm
(UZF) experimental sites in Zimbabwe. Error bars represent standard errors (N = 4). CT:
conventional tillage, CTR: conventional tillage with rotation, NT: no-tillage, NTM: no-tillage
with mulch, NTR: no-tillage with rotation, NTMR: no-tillage with mulch and rotation.

### 314 **3.3 SOC stock**

315 There were significant (p < 0.05) treatment effects on SOC stocks per soil layer in the top 5 at UZF and 10 cm at DTC (Table S3). Compared to CT, CTR and NT, NTM had at least 1.1 and 316 317 1.3 times more SOC stocks in the top 5 and 10 cm layers at UZF and DTC, respectively. In terms of cumulative SOC stocks, significant (p < 0.05) treatment effects were limited to the top 318 30 cm at DTC and the 20 cm at UZF, where no tillage with mulching (NTM) increased SOC 319 320 stocks (Table 1). There were no significant (p > 0.05) tillage effects on SOC stocks (CT vs NT) for both sites. The rotation component had no significant (p > 0.05) effects on SOC stocks 321 when comparing CTR and NTR at DTC. However, the maize-cowpea rotation under NT (NTR) 322 323 had at least 16 % higher SOC stocks in the top 30 cm compared to NT. In contrast, NTR had at least 7 % more SOC stocks than CTR in the top 10 cm at UZF. Compared to NT and CT, 324 the mulching component significantly (p < 0.05) increased SOC stocks by at least 8 % at UZF 325 326 and 13 % at DTC in the top 20 and 30 cm soil layers, respectively. SOC stocks in the full CA treatment (NTMR) were not significantly (p > 0.05) different with the other combinations of 327 CA principles (NTM, NTR) and CTR at DTC. At UZF, the full CA treatment had similar SOC 328 stocks as all the other NT treatments (NT, NTM, NTR). 329

SOC stocks for the whole soil profile for this study (0-100 cm) were at least 8.1, 3.5 and 2.1
times higher at DTC and 11.6, 4.4 and 2.4 times higher at UZF than the SOC stocks in the 0-5
cm (surface soil), 0-15 cm (tillage depth) and 0-30 cm (IPCC standard depth) layers. SOC
stocks for the subsoil (30-100 cm) ranged from 18.4 to 51.4 Mg C ha<sup>-1</sup> at DTC and 41.9 to
124.9 Mg C ha<sup>-1</sup> at UZF. Therefore, subsoil represented more than half (at least 53 % at DTC
and 58 % at UZF) of SOC stocks for the whole 100 cm soil profile.

LEMM (Mg ha <sup>-1</sup> )cml deptn (cm)CTCTRNTNTM(Mg ha <sup>-1</sup> )(cm) $C = 3.9 \pm 0.8c$ $4.4 \pm 0.4bc$ $3.8 \pm 0.7c$ $5.9 \pm 1.2a$ $530$ $0-5$ $3.9 \pm 0.8c$ $4.4 \pm 0.4bc$ $3.8 \pm 0.7c$ $5.9 \pm 1.8a$ $1340$ $0-10$ $7.4 \pm 1.3c$ $8.2 \pm 0.6bc$ $7.1 \pm 1.1c$ $9.9 \pm 1.8a$ $2760$ $0-15$ $10.7 \pm 1.6c$ $11.6 \pm 0.8bc$ $10.1 \pm 1.3c$ $13.5 \pm 2.0a$ $27760$ $0-20$ $13.6 \pm 1.7b$ $14.6 \pm 1.0ab$ $12.9 \pm 1.4b$ $16.7 \pm 2.1a$ $27590$ $0-20$ $19.4 \pm 1.9ab$ $20.5 \pm 1.2ab$ $18.4 \pm 1.6b$ $27.5 \pm 2.3a$ $7040$ $0-50$ $24.9 \pm 2.0a$ $26.5 \pm 1.3a$ $23.7 \pm 1.7a$ $27.5 \pm 2.4a$ $10550$ $0-75$ $29.6 \pm 1.9ab$ $31.2 \pm 1.3a$ $28.0 \pm 1.8a$ $2.0 \pm 2.4a$ $10750$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $28.0 \pm 1.8b$ $7.5 \pm 2.4a$ $460$ $0-50$ $29.5 \pm 1.9a$ $37.3 \pm 2.0a$ $37.5 \pm 2.4a$ $450$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $28.6 \pm 0.6bc$ $7.5 \pm 2.4a$ $870$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $27.5 \pm 2.4a$ $13770$ $0-100$ $46.5 \pm 2.0a$ $31.3 \pm 2.0ab$ $33.3 \pm 2.4a$ $13330$ $0-15$ $22.9 \pm 1.9a$ $23.4 \pm 1.8b$ $27.5 \pm 2.4a$ $13330$ $0-15$ $29.2 \pm 1.5b$ $21.4 \pm 1.2b$ $21.4 \pm 1.2b$ $13330$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $15.9 \pm 1.7a$ $13330$ <		Cumulative	Cumulative Approximate		Cumu	Cumulative SOC stocks (Mg C ha <sup>-1</sup> )	ocks (Mg C h	a <sup>-1</sup> )			
650 $0-5$ $3.9 \pm 0.8c$ $4.4 \pm 0.4bc$ $3.8 \pm 0.7c$ $5.9 \pm 1.2a$ 1340 $0-10$ $7.4 \pm 1.3c$ $8.2 \pm 0.6bc$ $7.1 \pm 1.1c$ $9.9 \pm 1.8a$ 2060 $0-15$ $10.7 \pm 1.6c$ $11.6 \pm 0.8bc$ $10.1 \pm 1.3c$ $13.5 \pm 2.0a$ 2760 $0-20$ $13.6 \pm 1.7b$ $14.6 \pm 1.0ab$ $12.9 \pm 1.4b$ $16.7 \pm 2.1a$ 2760 $0-20$ $13.6 \pm 1.7b$ $14.6 \pm 1.0ab$ $12.9 \pm 1.4b$ $25.3 \pm 2.2a$ 5590 $0-40$ $24.9 \pm 2.0a$ $26.6 \pm 1.3a$ $23.7 \pm 1.7a$ $27.5 \pm 2.3a$ 7040 $0-50$ $24.9 \pm 2.0a$ $26.5 \pm 1.3a$ $23.7 \pm 1.7a$ $27.5 \pm 2.4a$ 10550 $0-75$ $38.5 \pm 2.0a$ $41.8 \pm 1.6b$ $22.5 \pm 2.4a$ 10570 $0-70$ $38.5 \pm 2.0a$ $41.8 \pm 2.0a$ $47.5 \pm 2.4a$ 10570 $0-100$ $46.5 \pm 2.0a$ $41.8 \pm 1.0c$ $9.5 \pm 1.7a$ 13770 $0-100$ $46.5 \pm 2.0a$ $7.1 \pm 1.3a$ $28.0 \pm 1.8a$ 373 $20.7 \pm 1.3a$ $37.3 \pm 2.0a$ $37.5 \pm 2.4a$ 13370 $0-10$ $46.5 \pm 2.0a$ $7.9 \pm 0.5d$ $47.8 \pm 2.0a$ 870 $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $17.3 \pm 1.7a$ 13330 $0-15$ $22.9 \pm 1.9b$ $29.9 \pm 1.3b$ $17.3 \pm 1.7a$ 13330 $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $17.3 \pm 1.7a$ 13330 $0-10$ $15.4 \pm 1.5bc$ $29.9 \pm 2.1b$ $31.3 \pm 2.2a$ 13330 $0-10$ $15.4 \pm 1.5c$ $29.9 \pm 2.2b$ $31.3 \pm 2.2a$ 1330 $0-20$ $30.$	SILE	ESM (Mg ha <sup>-1</sup> )	soil depth – (cm)	CT	CTR	NT	NTM	NTR	NTMR	חגח	Dignificance
1340 $0-10$ $7.4 \pm 1.3c$ $8.2 \pm 0.6bc$ $7.1 \pm 1.1c$ $9.9 \pm 1.8a$ 2060 $0-15$ $10.7 \pm 1.6c$ $11.6 \pm 0.8bc$ $10.1 \pm 1.3c$ $13.5 \pm 2.0a$ 2760 $0-20$ $13.6 \pm 1.7b$ $14.6 \pm 1.0ab$ $12.9 \pm 1.4b$ $16.7 \pm 2.1a$ 2760 $0-30$ $13.6 \pm 1.7b$ $14.6 \pm 1.0ab$ $12.9 \pm 1.4b$ $16.7 \pm 2.1a$ 5590 $0-40$ $24.9 \pm 2.0a$ $20.5 \pm 1.2ab$ $18.4 \pm 1.6b$ $2.7.5 \pm 2.3a$ 5590 $0-40$ $24.9 \pm 2.0a$ $21.2 \pm 1.3a$ $23.7 \pm 1.7a$ $27.5 \pm 2.3a$ 10550 $0-75$ $29.6 \pm 1.9a$ $31.2 \pm 1.3a$ $23.7 \pm 1.7a$ $27.5 \pm 2.4a$ 13770 $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $37.3 \pm 2.0a$ $39.5 \pm 2.4a$ 460 $0-5$ $38.5 \pm 2.0a$ $7.9 \pm 0.5d$ $8.6 \pm 0.6bc$ $9.6 \pm 1.0a$ 870 $0-100$ $46.5 \pm 2.0a$ $7.9 \pm 0.5d$ $8.6 \pm 0.6bc$ $9.6 \pm 1.0a$ 870 $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $15.9 \pm 1.3b$ $17.3 \pm 1.7a$ 1330 $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $15.9 \pm 1.3b$ $25.1 \pm 2.1a$ 870 $0-10$ $15.4 \pm 1.5bc$ $12.8 \pm 1.0a$ $31.3 \pm 2.0ab$ $33.3 \pm 2.4a$ 1330 $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $23.4 \pm 1.8b$ $25.1 \pm 2.1a$ 870 $0-10$ $12.8 \pm 2.2a$ $44.8 \pm 2.0a$ $47.8 \pm 2.2a$ $47.8 \pm 2.2a$ 1330 $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $23.4 \pm 1.8b$ $25.1 \pm 2.1a$ 870 $0-10$ $2$		650	0-5	$3.9\pm0.8c$	$4.4 \pm 0.4 \mathrm{bc}$	$3.8\pm0.7c$	$5.9 \pm 1.2a$	$5.3 \pm 1.1ab$	$5.4 \pm 0.7 ab$	0.9	p < 0.001
2060 $0-15$ $10.7\pm1.6c$ $11.6\pm0.8bc$ $10.1\pm1.3c$ $13.5\pm2.0a$ $2760$ $0-20$ $13.6\pm1.7b$ $14.6\pm1.0ab$ $12.9\pm1.4b$ $16.7\pm2.1a$ $4160$ $0-30$ $19.4\pm1.9ab$ $20.5\pm1.2ab$ $18.4\pm1.6b$ $22.3\pm2.2a$ $5590$ $0-40$ $24.9\pm2.0a$ $26.6\pm1.3a$ $23.7\pm1.7a$ $27.5\pm2.3a$ $7040$ $0-50$ $29.6\pm1.9a$ $31.2\pm1.3a$ $28.0\pm1.8a$ $32.0\pm2.4a$ $10550$ $0-75$ $38.5\pm2.0a$ $42.6\pm1.3a$ $37.3\pm2.0a$ $39.5\pm2.4a$ $13770$ $0-100$ $46.5\pm2.0a$ $51.4\pm1.3a$ $44.8\pm2.0a$ $47.5\pm2.4a$ $460$ $0-5$ $8.2\pm0.9cd$ $7.9\pm0.5d$ $8.6\pm0.6bc$ $9.6\pm1.0a$ $870$ $0-100$ $15.4\pm1.5bc$ $14.8\pm1.0c$ $17.3\pm1.7a$ $870$ $0-10$ $15.4\pm1.5bc$ $14.8\pm1.0c$ $15.9\pm1.3b$ $17.3\pm1.7a$ $1330$ $0-10$ $15.4\pm1.5bc$ $22.1\pm1.6b$ $23.4\pm1.8b$ $25.1\pm2.1a$ $870$ $0-10$ $15.4\pm1.5bc$ $14.8\pm1.0c$ $15.9\pm1.3b$ $17.3\pm1.7a$ $870$ $0-10$ $0-20$ $30.8\pm2.2b$ $29.9\pm2.1b$ $21.4\pm1.8b$ $25.1\pm2.1a$ $1330$ $0-10$ $55.2\pm2.2a$ $44.1\pm2.18$ $46.4\pm2.8a$ $53.1\pm2.2a$ $4030$ <td></td> <td>1340</td> <td>0-10</td> <td><math>7.4 \pm 1.3c</math></td> <td><math>8.2\pm0.6bc</math></td> <td><math>7.1 \pm 1.1c</math></td> <td><math>9.9 \pm 1.8a</math></td> <td><math>9.4 \pm 1.5ab</math></td> <td><math>9.4 \pm 1.1ab</math></td> <td>1.2</td> <td>p &lt; 0.001</td>		1340	0-10	$7.4 \pm 1.3c$	$8.2\pm0.6bc$	$7.1 \pm 1.1c$	$9.9 \pm 1.8a$	$9.4 \pm 1.5ab$	$9.4 \pm 1.1ab$	1.2	p < 0.001
$2760$ $0-20$ $13.6 \pm 1.7b$ $14.6 \pm 1.0ab$ $4160$ $0-30$ $19.4 \pm 1.9ab$ $20.5 \pm 1.2ab$ $5590$ $0-40$ $24.9 \pm 2.0a$ $26.6 \pm 1.3a$ $7040$ $0-50$ $29.6 \pm 1.9a$ $31.2 \pm 1.3a$ $7040$ $0-50$ $29.6 \pm 1.9a$ $31.2 \pm 1.3a$ $10550$ $0-75$ $38.5 \pm 2.0a$ $42.6 \pm 1.3a$ $13770$ $0-100$ $46.5 \pm 2.0a$ $42.6 \pm 1.3a$ $13770$ $0-100$ $46.5 \pm 2.0a$ $7.9 \pm 0.5d$ $870$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $870$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $55.2 \pm 2.0a$ $29.9 \pm 2.1b$ $2760$ $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ $2760$ $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ $2760$ $0-20$ $89.3 \pm 2.4a$ $42.8 \pm 2.2a$ $4030$ $0-76$ $89.3 \pm 2.7a$ $70.4 \pm 3.0a$ $8190$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $8190$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		2060	0-15		$11.6 \pm 0.8 bc$	$10.1 \pm 1.3c$	$13.5 \pm 2.0a$	13.1 ± 1.7ab 12.9 ± 1.2ab	$12.9 \pm 1.2ab$	1.7	p < 0.05
$4160$ $0-30$ $19.4 \pm 1.9ab$ $20.5 \pm 1.2ab$ $5590$ $0-40$ $24.9 \pm 2.0a$ $26.6 \pm 1.3a$ $7040$ $0-50$ $29.6 \pm 1.9a$ $31.2 \pm 1.3a$ $7040$ $0-75$ $38.5 \pm 2.0a$ $42.6 \pm 1.3a$ $10550$ $0-75$ $38.5 \pm 2.0a$ $42.6 \pm 1.3a$ $13770$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $460$ $0-5$ $8.2 \pm 0.9cd$ $7.9 \pm 0.5d$ $870$ $0-100$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-10$ $15.4 \pm 1.5bc$ $29.9 \pm 2.1b$ $1330$ $0-10$ $52.2 \pm 2.6a$ $29.9 \pm 2.1b$ $2760$ $0-30$ $42.3 \pm 2.2a$ $42.8 \pm 2.2a$ $4030$ $0-930$ $42.3 \pm 2.7a$ $70.4 \pm 3.0a$ $5300$ $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ $8190$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $11050$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		2760	0-20		$14.6 \pm 1.0ab$	$12.9 \pm 1.4b$	$16.7 \pm 2.1a$	$16.2 \pm 1.9a$	$15.8 \pm 1.4a$	2.1	p < 0.05
5590 $0-40$ $24.9 \pm 2.0a$ $26.6 \pm 1.3a$ 7040 $0-50$ $29.6 \pm 1.9a$ $31.2 \pm 1.3a$ 10550 $0-75$ $38.5 \pm 2.0a$ $42.6 \pm 1.3a$ 13770 $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ 460 $0-5$ $8.2 \pm 0.9cd$ $7.9 \pm 0.5d$ 870 $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ 1330 $0-10$ $5.22.9 \pm 1.9b$ $22.1 \pm 1.6b$ 1330 $0-10$ $55.2 \pm 2.2b$ $29.9 \pm 2.1b$ 2760 $0-30$ $42.3 \pm 2.24a$ $42.8 \pm 2.2a$ 4030 $0-30$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ 8190 $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ 8190 $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ 11050 $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$	DTC	4160	0-30	$19.4 \pm 1.9ab$	$20.5 \pm 1.2ab$	$18.4 \pm 1.6b$	$22.3 \pm 2.2a$	$22.0 \pm 1.9a$	$22.0 \pm 1.5a$	2.7	p < 0.05
70400-50 $29.6 \pm 1.9a$ $31.2 \pm 1.3a$ $10550$ $0-75$ $38.5 \pm 2.0a$ $42.6 \pm 1.3a$ $13770$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $460$ $0-5$ $8.2 \pm 0.9cd$ $7.9 \pm 0.5d$ $870$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $1330$ $0-15$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ $1840$ $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ $2760$ $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ $4030$ $0-40$ $55.2 \pm 2.6a$ $58.1 \pm 2.6a$ $8190$ $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ $8190$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $11050$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		5590	0-40	+1	$26.6 \pm 1.3a$	$23.7 \pm 1.7a$	$27.5 \pm 2.3a$	$27.9 \pm 2.0a$	$26.9 \pm 1.6a$	3.1	ns
10550 $0.75$ $38.5 \pm 2.0a$ $42.6 \pm 1.3a$ $13770$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $460$ $0.5$ $8.2 \pm 0.9cd$ $7.9 \pm 0.5d$ $870$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $870$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $1330$ $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $1840$ $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ $2760$ $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ $4030$ $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ $8190$ $0-76$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ $8190$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $11050$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		7040	0-50	$29.6\pm1.9a$	$31.2 \pm 1.3a$	$28.0\pm1.8a$	$32.0 \pm 2.4a$	$32.7 \pm 2.1a$	31.7 ± 1.7a	3.4	ns
$13770$ $0-100$ $46.5 \pm 2.0a$ $51.4 \pm 1.3a$ $460$ $0-5$ $8.2 \pm 0.9cd$ $7.9 \pm 0.5d$ $870$ $0-10$ $15.4 \pm 1.5bc$ $14.8 \pm 1.0c$ $1330$ $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $1330$ $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ $1840$ $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ $2760$ $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ $4030$ $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ $5300$ $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ $8190$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $11050$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		10550	0-75		$42.6\pm1.3a$	$37.3 \pm 2.0a$	$39.5 \pm 2.4a$	$42.7 \pm 2.1a$	$42.6 \pm 1.9a$	5.2	ns
460 $0-5$ $8.2 \pm 0.9 \text{cd}$ $7.9 \pm 0.5 \text{d}$ $870$ $0-10$ $15.4 \pm 1.5 \text{bc}$ $14.8 \pm 1.0 \text{c}$ $1330$ $0-15$ $22.9 \pm 1.9 \text{b}$ $22.1 \pm 1.6 \text{b}$ $1840$ $0-20$ $30.8 \pm 2.2 \text{b}$ $29.9 \pm 2.1 \text{b}$ $2760$ $0-30$ $42.3 \pm 2.4 \text{a}$ $42.8 \pm 2.2 \text{a}$ $4030$ $0-30$ $42.3 \pm 2.4 \text{a}$ $42.8 \pm 2.2 \text{a}$ $4030$ $0-40$ $55.2 \pm 2.6 \text{a}$ $58.1 \pm 2.6 \text{a}$ $5300$ $0-50$ $66.3 \pm 2.7 \text{a}$ $70.4 \pm 3.0 \text{a}$ $8190$ $0-75$ $89.3 \pm 3.1 \text{a}$ $95.9 \pm 3.3 \text{a}$ $11050$ $0-100$ $107.8 \pm 3.5 \text{a}$ $119.1 \pm 3.7 \text{a}$		13770	0-100		$51.4 \pm 1.3a$	$44.8\pm2.0a$	$47.5 \pm 2.4a$	$51.1 \pm 2.2a$	$50.7 \pm 2.0a$	6.3	ns
870 $0-10$ $15.4 \pm 1.5$ bc $14.8 \pm 1.0$ c $1330$ $0-15$ $22.9 \pm 1.9$ b $22.1 \pm 1.6$ b $1840$ $0-20$ $30.8 \pm 2.2$ b $29.9 \pm 2.1$ b $2760$ $0-30$ $42.3 \pm 2.4$ a $42.8 \pm 2.2$ a $4030$ $0-40$ $55.2 \pm 2.6$ a $58.1 \pm 2.6$ a $5300$ $0-60$ $66.3 \pm 2.7$ a $70.4 \pm 3.0$ a $8190$ $0-75$ $89.3 \pm 3.1$ a $95.9 \pm 3.3$ a $11050$ $0-100$ $107.8 \pm 3.5$ a $119.1 \pm 3.7$ a		460	0-5	$8.2 \pm 0.9$ cd	$7.9 \pm 0.5d$	$8.6 \pm 0.6 bc$	$9.6 \pm 1.0a$	$8.8 \pm 0.9 bc$	$9.2 \pm 0.9ab$	0.7	p < 0.001
1330 $0-15$ $22.9 \pm 1.9b$ $22.1 \pm 1.6b$ 1840 $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ 2760 $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ 4030 $0-40$ $55.2 \pm 2.6a$ $58.1 \pm 2.6a$ 5300 $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ 8190 $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ 11050 $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		870	0-10		$14.8\pm1.0\mathrm{c}$	$15.9 \pm 1.3b$	$17.3 \pm 1.7a$	$15.9 \pm 1.6b$	$16.3 \pm 1.4ab$	1.1	p < 0.05
1840 $0-20$ $30.8 \pm 2.2b$ $29.9 \pm 2.1b$ 2760 $0-30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ 4030 $0-40$ $55.2 \pm 2.6a$ $58.1 \pm 2.6a$ 5300 $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ 8190 $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ 11050 $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		1330	0-15		$22.1 \pm 1.6b$	$23.4\pm1.8b$	$25.1 \pm 2.1a$	$23.2\pm1.9b$	$23.6\pm1.7ab$	1.7	p < 0.05
2760 $0.30$ $42.3 \pm 2.4a$ $42.8 \pm 2.2a$ 4030 $0.40$ $55.2 \pm 2.6a$ $58.1 \pm 2.6a$ 5300 $0.50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ 8190 $0.75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ 11050 $0.100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		1840	0-20	+1	$29.9 \pm 2.1b$	$31.3 \pm 2.0ab$	$33.3 \pm 2.4a$	$30.9 \pm 2.2b$	$31.0 \pm 2.1b$	7	p < 0.05
$0-40$ $55.2 \pm 2.6a$ $58.1 \pm 2.6a$ $0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$	UZF	2760	0-30	+1	$42.8\pm2.2a$	$44.1 \pm 2.1a$	$46.4 \pm 2.8a$	$41.9 \pm 2.7a$	43.3 ± 2.7a	3.3	ns
$0-50$ $66.3 \pm 2.7a$ $70.4 \pm 3.0a$ $0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		4030	0-40	+1	$58.1 \pm 2.6a$	$57.2 \pm 2.2a$	$61.0 \pm 3.3a$	$56.7 \pm 3.0a$	$57.5 \pm 3.2a$	4.8	ns
$0-75$ $89.3 \pm 3.1a$ $95.9 \pm 3.3a$ $0-100$ $107.8 \pm 3.5a$ $119.1 \pm 3.7a$		5300	0-50	+1	$70.4 \pm 3.0a$	$67.5 \pm 2.3a$	73.1 ± 3.9a	$68.8 \pm 3.1a$	$69.7 \pm 3.3a$	6.6	ns
$0-100 \qquad 107.8 \pm 3.5a \qquad 119.1 \pm 3.7a$		8190	0-75	+1	$95.9 \pm 3.3a$	$90.0 \pm 2.7a$	$89.9 \pm 4.6a$	$93.7 \pm 3.9a$	$98.4 \pm 4.3a$	17	su
		11050	0-100	$107.8\pm3.5a$	$119.1 \pm 3.7a$	$109.8\pm3.3a$	$110.9\pm5.2a$	116.1 ± 4.9a	$124.9 \pm 5.6a$	19	Su

Table 1. Cumulative SOC stocks at the Domboshava Training Centre (DTC) and University
of Zimbabwe Farm (UZF) after 8 years of different tillage, residue and crop management
systems. Means in the same row followed by different superscript letters are significantly

different and associated errors are standard errors (N = 4). CT: conventional tillage, CTR: conventional tillage with rotation, NT: no-tillage, NTM: no-tillage with mulch, NTR: no-tillage with rotation, NTMR: no-tillage with mulch and rotation.

343

# 344 **3.4 SOC accumulation and loss rates**

SOC accumulation rates at UZF differed significantly (p < 0.05) with soil depth where top soil layers (0-5, 0-10, 0-15, 0-20 and 0-30 cm) had SOC accumulation rates that were at least 6.9 times less than when considering the 0-100 cm soil profile (Table 2). In contrast, there were no significant (p > 0.05) differences, at DTC in SOC accumulation rates with depth. On average, SOC accumulation rates ranged between 0.13 and 0.08 Mg C ha<sup>-1</sup> yr<sup>-1</sup> in the top soil (0-5 cm) to 0.33 and 1.16 Mg C ha<sup>-1</sup> yr<sup>-1</sup> for the whole 1 m soil profile at DTC and UZF, respectively. The depth and treatment interaction had no significant (p > 0.05) effects at both sites.

On the other hand, the different treatments in this study had significant (p < 0.05) effects in 352 SOC accumulation / loss rates in the top 20 cm soil layer at both sites (Table 2). At DTC, NT 353 354 had significant (p < 0.05) net loss of SOC in the 0-20 cm layer, ranging between -0.09 and -0.02 Mg C ha<sup>-1</sup> yr<sup>-1</sup>, whereas NT treatments (NTM, NTR, NTMR) had SOC accumulation rates 355 ranging from 0.17 to 0.38 Mg C ha<sup>-1</sup> yr<sup>-1</sup>. However, maize stover mulching (NTM) had 356 significantly (p < 0.05) higher SOC accumulation rates than CTR (2.9 - 4.2 times) and NT (5.2357 -13.5 times) in the top 15 cm and 20 cm layers, respectively. The different combinations of 358 mulching and rotation under NT (NTM, NTR and NTMR) had no significant (p > 0.05)359 differences in SOC accumulation rates. Similarly, rotation treatments (CTR, NTR, NTMR) 360 showed no significant (p > 0.05) differences in SOC accumulation rates. Thus, the full CA 361 treatment had similar SOC accumulations rates to treatments with at least 2 combinations of 362 CA principles (NTM and NTR) and to CTR. 363

364	In contrast, at UZF, CTR had significant ( $p < 0.05$ ) net loss of SOC in the top 20 cm (Table 2).
365	The no-tillage treatments (NT, NTM, NTR, NTMR) showed significantly ( $p < 0.05$ ) higher
366	SOC accumulation rates $(0.05 - 0.25 \text{ Mg C ha}^{-1} \text{ yr}^{-1})$ than CTR which ranged between -0.07 to
367	-0.03 Mg C ha <sup>-1</sup> yr <sup>-1</sup> in top 10 cm soil layer. NTM had the highest SOC accumulations rates
368	(0.28 to 0.32 Mg C ha <sup>-1</sup> yr <sup>-1</sup> ) when considering the 0-15 and 0-20 cm soil layers. SOC
369	accumulation rates in NTM were at least 3.8, 3.5 and 2.4 times higher than CTR, NT and NTR
370	in the top 20 cm. The full CA treatment (NTMR) had significantly ( $p < 0.05$ ) higher SOC
371	accumulation rates compared to CTR ( $2.5 - 5.3$ times) in the top 10 cm and lower SOC
372	accumulation rate to NTM in the 0-10 cm (2.3 times) and 0-20 cm (10.6 times) soil layer.
373	However, there were no significant ( $p > 0.05$ ) differences in SOC accumulation rates between
374	treatments beyond 20 cm soil layer at both sites.

Site	Approximate soil depth SOC accumulation or loss rate (Mg C ha <sup>-1</sup> yr <sup>-1</sup> )						LSD	Sig
Site	(cm)	CTR	NT	NTM	NTR	NTMR	_	
	0-5	$0.06\pm0.05bc$	$-0.02 \pm 0.02c$	$0.25\pm0.05a$	$0.17 \pm 0.02ab$	$0.19\pm0.04ab$	0.13	***
	0-10	$0.10\pm0.09 bc$	$\textbf{-0.04} \pm 0.04c$	$0.31\pm0.09a$	$0.24 \pm 0.01 ab$	$0.25\pm0.08ab$	0.16	***
	0-15	$0.12\pm0.13 bc$	$\textbf{-0.07} \pm 0.05c$	$0.35\pm0.13a$	$0.30 \pm 0.01 ab$	$0.27 \pm 0.12 ab$	0.23	**
DTC	0-20	$0.12\pm0.17 ab$	$\textbf{-0.09} \pm 0.05b$	$0.38\pm0.17a$	$0.32\pm0.01a$	$0.27 \pm 0.16a$	0.29	**
	0-30	$0.13 \pm 0.25a$	$-0.13 \pm 0.07a$	$0.36\pm0.25a$	$0.32\pm0.08a$	$0.33\pm0.20a$	0.35	ns
	0-40	$0.22 \pm 0.25a$	$-0.15 \pm 0.07a$	$0.33\pm0.25a$	$0.38\pm0.07a$	$0.25\pm0.23a$	0.41	ns
	0-50	$0.20 \pm 0.27a$	$\textbf{-0.20} \pm 0.14a$	$0.30\pm0.27a$	$0.40\pm0.09a$	$0.26 \pm 0.22a$	0.46	ns
	0-75	$0.51 \pm 0.28a$	$-0.15 \pm 0.28a$	$0.13\pm0.28a$	$0.53 \pm 0.13a$	$0.51\pm0.20a$	0.73	ns
	0-100	$0.62 \pm 0.32a$	$\textbf{-0.20} \pm \textbf{0.37a}$	$0.13\pm0.32a$	$0.58\pm0.29a$	$0.53\pm0.20a$	0.86	ns
	0-5	$-0.03\pm0.03c$	$0.05\pm0.04b$	$0.17\pm0.05a$	$0.07\pm0.04b$	$0.13 \pm 0.06ab$	0.08	***
	0-10	$\textbf{-0.07} \pm 0.04c$	$0.07\pm0.08b$	$0.25\pm0.09a$	$0.07\pm0.07b$	$0.11\pm0.08b$	0.13	**
	0-15	$\textbf{-0.10} \pm 0.03 b$	$0.06\pm0.11b$	$0.28\pm0.13a$	$0.04\pm0.07b$	$0.09 \pm 0.11$ ab	0.22	**
UZF	0-20	$\textbf{-0.11} \pm 0.07 b$	$0.06\pm0.14b$	$0.32\pm0.17a$	$0.02\pm0.11b$	$0.03\pm0.12b$	0.25	**
	0-30	$0.06 \pm 0.15a$	$0.22\pm0.25a$	$0.51\pm0.25a$	$\textbf{-0.05} \pm \textbf{0.18a}$	$0.12 \pm 0.16a$	0.44	ns
	0-40	$0.37 \pm 0.11a$	$0.25\pm0.27a$	$0.72\pm0.25a$	$0.19 \pm 0.28a$	$0.29\pm0.14a$	0.65	ns
	0-50	$0.51 \pm 0.20a$	$0.15\pm0.34a$	$0.85\pm0.27a$	$0.31 \pm 0.41a$	$0.43\pm0.10a$	0.88	ns
	0-75	$0.83\pm0.56a$	$0.08\pm0.55a$	$0.08\pm0.28a$	$0.55\pm0.76a$	$1.14\pm0.44a$	1.17	ns
	0-100	$1.41 \pm 0.86a$	$0.25\pm0.75a$	$0.98 \pm 0.32a$	$1.03 \pm 1.26a$	$2.14\pm0.99a$	2.31	ns

**Table 2**. SOC change rates ( $\pm$  standard error, N = 4) of the different treatments compared to

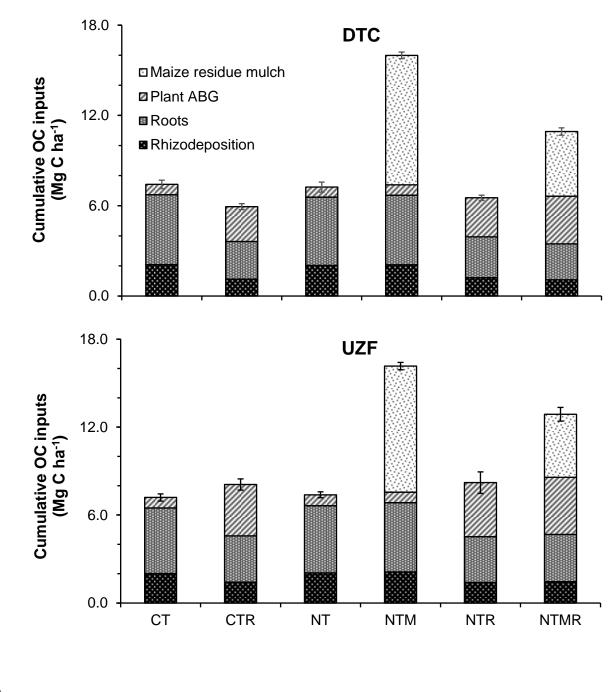
376 CT (conventional tillage) at Domboshava Training Centre (DTC) and University of Zimbabwe

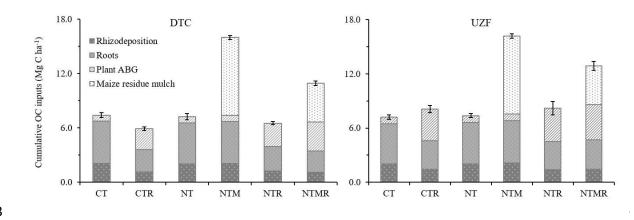
farm (UZF). Means in the same row followed by different superscripts are significantly different. CTR: conventional tillage with rotation, NT: no-tillage, NTM: no-tillage with mulch, NTR: no-tillage with rotation, NTMR: no-tillage with mulch and rotation, LSD = least significance difference, ns = not significant, Sig = significance, \*\* = p < 0.05, \*\*\* = p < 0.001.

381

# 382 3.5 Organic carbon inputs via crops residues, root mortality and rhizodeposition to the 383 soil

There were significant (p < 0.001) differences in cumulative OC inputs between treatments 384 (Figure 4). Cumulative OC inputs were at least 1.5 times higher in mulch treatments (NTM, 385 386 NTMR) than in treatments without mulch. However, the mulch plus rotation treatment (NTMR) had significantly (p < 0.001) lower cumulative OC inputs than continuous mulching 387 (NTM). Cumulative OC input after 8 seasons was as high as 16.0 and 10.5 Mg C ha<sup>-1</sup> at DTC 388 and 16.2 and 12.4 Mg C ha<sup>-1</sup> at UZF in NTM and NTMR, respectively (Figure 4), resulting in 389 mean annual OC input rates of about 1.3 to 1.6 Mg C ha<sup>-1</sup> yr<sup>-1</sup> for NTMR and 2.0 Mg C ha<sup>-1</sup> yr<sup>-1</sup> 390 <sup>1</sup> for NTM. The other treatments had mean annual OC input rates  $\leq 1.0 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ . 391





393

Figure 4. Estimated cumulative organic carbon (OC) inputs to the soil from the 2013/14 to the 394 2020/21 cropping season for the different treatments at the Domboshava Training Centre 395 (DTC) and the University of Zimbabwe Farm (UZF) experimental sites. Error bars represent 396 standard errors (n = 4) for the cumulative OC. CT: conventional tillage, CTR: conventional 397 tillage with rotation, NT: no-tillage, NTM: no-tillage with mulch, NTR: no-tillage with 398 399 rotation, NTMR: no-tillage with mulch and rotation, ABG: aboveground biomass (corresponding to an estimated 5 % of the maize aboveground vegetative biomass remained in 400 401 the field because maize stalk slashing at harvesting did not remove the whole stem).

# 403 **4. Discussion**

# 404 4.1 Role of soil texture in SOC accumulation

Soil texture is widely recognized to influence SOC stocks (Sun et al., 2020) through physical
and chemical protection of SOC against microbially mediated decomposition (Chivenge et al.,
2007; Mtambanengwe et al., 2004). In our study, the main difference between the two study
sites is soil texture in the top soil (0-30 cm), where DTC had light textured (sandy loams) soils
and UZF had medium to heavy textured (sandy-clay-loams) soils (Figure 1). These soil textural
differences explain why there were no differences in SOC stocks, changes and accumulation

411 rates between NTM, NTR and NTMR at DTC regardless of higher cumulative OC inputs in NTM and NTMR (Figure 4). The direct SOC inputs in the top soil at DTC, where SOC was 412 more concentrated (Table S2, Figure 3) was subject to mineralization because of low elay 413 414 content and thus low protection by soil micro-aggregates (Chivenge et al., 2007; Mtambanengwe et al., 2004; Sun et al., 2020), such that the differences in OC inputs had little 415 effect. Light textured soils have large pores which cannot protect SOC against microbial 416 417 decomposition (Mtambanengwe et al., 2004; Christensen, 1987; Sun et al., 2020; Kravehenko and Guber, 2017). Additionally, the low clay content meant less surface area for SOC 418 419 adsorption (Han et al., 2016; Churchman et al., 2020) which is another mechanism for SOC protection from mineralization. In contrast, there were differences between NTM and NTR at 420 UZF in the top soil layers and intermediate between NTM and NTMR. Cumulative OC inputs 421 422 in NTMR (12.4 Mg C ha<sup>-1</sup>) were about 75 % of cumulative OC inputs in NTM (16.2 Mg C ha<sup>-1</sup>) 423 <sup>+</sup>) (Figure 4) after 8 seasons. The added C, especially from maize stover mulch, most likely was protected by clay particles as well as formation of organo-mineral complexes (Malepfane 424 425 et al., 2022; Chivenge et al., 2007; Jephita et al., 2023) which protects SOC from mineralization (Dunjana et al., 2012; Shumba et al., 2020; Button et al., 2022; Rumpel et al., 2012; Sanaullah 426 et al., 2016). 427

428

# 429 **4.2-1** SOC distribution across soil depth

Cumulative SOC stocks for the whole soil profile (0-100 cm) for this study were at least 8.0,
4.0 and 2.0 times higher than the 0-5 (surface soil), 0-15 (tillage depth for the study) and 0-30
(IPCC standard depth for SOC studies), for the two sites, respectively. This means that, over
half of SOC stocks for this study were in the sub-soil (30-100 cm) which reflects on the
importance of sub-soil SOC stocks. Significant SOC stocks in the sub-soil have also been

reported by other authors (Yost and Hartemink, 2020; Balesdent et al., 2018; Cardinael et al., 435 2015; Harrison et al., 2011; Lal, 2018). Significant effects of mulch and/or rotation under NT 436 were restricted to the top 30 cm in our study as well as other studies in SSA (Dube et al., 2012; 437 Powlson et al., 2016) and the world at large (Balesdent et al., 2018; Yost and Hartemink, 2020), 438 which is most likely why the default soil depth for IPCC for SOC studies is 0-30 cm (IPCC, 439 2019). However, this underestimates whole soil profile C storage (Harrison et al., 2011; Singh 440 441 et al., 2018; Lorenz and Lal, 2005). Therefore, it is crucial to consider whole soil profile sampling when monitoring SOC storage in agricultural ecosystems to determine their C 442 443 sequestration potential in the pursuit of climate change mitigation (Malepfane et al., 2022). SOC mineralization is relatively low in the sub-soil due to lack of oxygen and physical 444 protection of SOC (aggregate protected C) (Rumpel et al., 2012; Sanaullah et al., 2016; Shumba 445 et al., 2020; Button et al., 2022). Therefore, in the pursuit to improve subsoil (> 30 cm) SOC 446 stocks through root mortality and exudates, crop varieties with higher root-length densities 447 (Chikowo et al., 2003) in the subsoil are recommended. 448

449 In this study, there is a conspicuous rotation effect at UZF in the subsoil (60-100 cm) on SOC concentration which is however, not significant. However, there is a block effect at UZF on 450 SOC concentration in the subsoil as shown in Figure S2 (see Figure S1 for DTC where no 451 block effect was observed) where there seems to be "outliers" in blocks 3 and 4. We decided 452 453 not to exclude the "outliers" since we thought it was a block effect rather than a treatment 454 effect. As a result, there are no significant differences in SOC concentration between treatments in deep soils. If we exclude the "outliers" in deep soils the graph for SOC concentration is as 455 456 shown in Figure S3 where the rotation effects tend to diminish. Nonetheless, all raw data of this paper can be freely accessed on the CIRAD data repository and linked to this paper 457 458 (Shumba et al., 2023a).

460

# 4.3-2 Cumulative SOC stocks and accumulation rates

Overall, the insignificant differences in cumulative SOC stocks and SOC accumulation / loss 461 rates between the different cropping systems in this and other studies (Laura et al., 2022; Leal 462 et al., 2020) when considering whole soil profile (0-100 cm) is alluded to the dilution effect 463 due to low OC inputs beyond 30 cm. Organic C inputs in the subsoil (> 30 cm) through crop 464 root mortality and root exudates are highly reduced due to low root biomass (Button et al., 465 466 2022; Chikowo et al., 2003). Several authors have also reported similar cumulative SOC stocks for soil profile depth > 30 cm between different tillage and residue management practices 467 (Angers et al., 1997; Doran et al., 1998; Lal, 2015; Powlson et al., 2014). This can be alluded 468 469 to an accumulation of uncertainty when cumulating SOC stocks in several soil layers with their respective error of measurement. This weakens the power of detecting statistically significant 470 differences even where such differences exist (Kravchenko and Robertson, 2011). Kravchenko 471 and Robertson, (2011) bemoaned the lack of enough replication when sampling deep soil 472 horizons to reduce variability and the importance of post hoc power analysis to reduce Type II 473 474 error. This study was limited to four replicates which might not have enough statistical power 475 to detect significant differences between treatments when considering the whole soil profile.

# 476 **4.<u>32</u>.1 Mulching**

The overarching role of mulching in cumulative SOC stocks and accumulation / loss rates at both sites (Tables 1 and 2), albeit, in the top soil (< 30 cm) has been shown in this study. Cumulative SOC stocks (Table 1) and SOC accumulation / loss rates (Table 2) did not differ with residue management under NT systems (NTM, NTMR) in the top soil at DTC regardless of high external OC inputs through maize residue application in mulch treatments (Figure 4, Table S4). This was attributed to low clay content (< 15 % clay) in the top 20 cm hence low

physical SOC protection (Chivenge et al., 2007; Mtambanengwe et al., 2004; Sun et al., 2020), 483 such that the differences in OC inputs had little effect. Alternatively, SOC can be protected 484 from mineralization through adsorption to clay particles (Han et al., 2016; Churchman et al., 485 2020). However, there was low surface area for SOC adsorption due to low clay content in the 486 top soil at DTC. Conversely, maize residue mulching effects were significant at UZF though 487 NTMR was indifferent when compared to the rest of the NT treatments. Cumulative OC inputs 488 in NTMR (12.4 Mg C ha<sup>-1</sup>) were about 77 % of cumulative OC inputs in NTM (16.2 Mg C ha<sup>-1</sup>) 489 <sup>1</sup>) but at least 57 % higher OC inputs than NT and NTR after 8 seasons (Figure 4). This was 490 491 alluded to SOC adsorption and physical protection due to higher clay content at UZF. Nonetheless, several studies have shown that aboveground biomass is less effective in 492 sustaining SOC stocks compared to belowground biomass (Hirte et al., 2018, 2021; Jones et 493 al., 2009; Villarino et al., 2021) and we attribute that to the insignificant cumulative SOC stocks 494 and accumulation rates between the NT treatments other than NTM regardless of higher 495 aboveground biomass in NTR and NMTR than NT (Figure 4). 496

# 497 4.<u>32</u>.2 Tillage

498 Significant changes in SOC stocks and accumulation rates were restricted to the top 20 cm at both sites below which there were no differences between treatments (Table 2). The 499 consistently low SOC stocks under NT, CT and CTR and SOC accumulation rates (CTR and 500 NT) at both sites was attributed to low OC inputs (Table S4, Figure 4). NT and CT had 501 generally low yields in terms of grain and vegetative aboveground biomass (Mhlanga et al., 502 2021; Shumba et al., 2023b) since the establishment of the experiment in the 2013/14 season 503 504 and hence low OC inputs through stubble, root mortality and root exudates. Our results dovetails with studies done elsewhere (Du et al., 2017; Koga and Tsuji, 2009) and meta-505 analyses and reviews (Corbeels et al., 2020a; Lal, 2018, 2015) where the authors found that 506

NT alone does not significantly improve SOC. However, higher SOC stocks were observed 507 when NT was combined with at least two CA principles (mulching and rotation) at DTC in the 508 top 20 cm (Table 1). It has been reported that NT cropping systems enhance SOC accumulation 509 through increasing C inputs in the top layers and reducing erosion through minimum soil 510 disturbance (Six et al., 2000; Lal, 2015, 2018; Bai et al., 2019; Cai et al., 2022). Minimum soil 511 disturbance through NT also physically protects SOC in microaggregates from exposure to 512 513 oxidative losses (Shumba et al., 2020; Six et al., 2002; Dolan et al., 2006; Liang et al., 2020). However, NT without mulch is a nonentity compared to other combinations of CA principles 514 515 for long-term sustainability in cropping systems (Nyamangara et al., 2013; Kodzwa et al., 2020; Mhlanga et al., 2021; Li et al., 2020; Bohoussou et al., 2022) and NT is only effective in 516 increasing SOC stocks when it is associated with other CA principles, especially mulch. On 517 the other hand, our study suggest that NT can achieve the same results of SOC storage as NTR 518 and NTMR since they had similar SOC stocks at UZF. This can be explained by the low 519 aboveground OC inputs in rotation treatments during the season when cowpeas were grown. 520

521

# 4.32.3 Maize-cowpea rotation

522 Legume rotations have been found to improve SOC accumulation rates and subsequent soil structural improvement (aggregation) induced by the addition of organic residues with 523 favourable C/N ratio (Virk et al., 2022; Laub et al., 2023; Jephita et al., 2023). However, in our 524 525 study, cowpea rotation benefits on SOC accumulation rates were not significant at DTC. Maize-cowpea rotation had no significant effects on maize yield (Shumba et al., 2023b; 526 Mhlanga et al., 2021) which corresponded to low belowground biomass as well. Instead, maize 527 528 stover mulching improved maize yields at DTC. Nevertheless, benefits from cowpea rotation under NT cropping systems (NTR, NTMR) compared to CT cropping systems (CTR) were 529 significant, albeit only in the top 10 cm, at UZF; CTR had a net loss of SOC (-0.07  $\pm$  0.04 to 530

 $0.03 \pm 0.03$  Mg C ha<sup>-1</sup> yr<sup>-1</sup>). Significantly higher maize yields in rotation treatments were 531 observed at UZF (Shumba et al., 2023b; Mhlanga et al., 2021) and were attributed to more soil 532 mineral N due to biological nitrogen fixation from the preceding cowpeas. Higher aboveground 533 biomass is positively related to below ground biomass resulting in significant belowground OC 534 inputs, of higher quality in the rotation treatments in the season when maize is grown. However, 535 the net SOC loss in CTR at UZF was due to seasonal exposure to oxidative losses (SOC 536 537 mineralization) through disruption of soil macroaggregates by tillage as alluded by Bai et al., (2019); Cambardella and Elliott, (1993) and Lal, (2018). We underscore that maize-cowpea 538 539 rotation under NT improved SOC accumulation in the top soil due to reduced soil disturbance and alternate OC inputs of high (cowpeas) and low quality (maize). High quality OC inputs 540 have a positive priming effect (Chen et al., 2014) which have been shown to be preferentially 541 stabilized in the soil due to a higher carbon use efficiency of soil microbes (Cotrufo et al., 2013; 542 Kopittke et al., 2018). This explains significant improvement in SOC stocks under the 543 combination of NT and alternate high- and low-quality OC inputs (maize-cowpea rotation) to 544 the soil in medium to heavy textured soils at UZF and vice versa at DTC. 545

# 546 <u>4.3. Role of soil texture in SOC accumulation</u>

547 Soil texture is widely recognized to influence SOC stocks (Sun et al., 2020) through physical and chemical protection of SOC against microbially mediated decomposition (Chivenge et al., 548 549 2007; Mtambanengwe et al., 2004). In our study, the main difference between the two study sites is soil texture in the top soil (0-30 cm), where DTC had light textured (sandy loams) soils 550 and UZF had medium to heavy textured (sandy-clay-loams) soils (Figure 1). These soil textural 551 552 differences explain why there were no differences in SOC stocks, changes and accumulation rates between NTM, NTR and NTMR at DTC regardless of higher cumulative OC inputs in 553 NTM and NTMR (Figure 4). The direct SOC inputs in the top soil at DTC, where SOC was 554

555 more concentrated (Table S2, Figure 3) was subject to mineralization because of low clay content and thus low protection by soil micro-aggregates (Chivenge et al., 2007; 556 Mtambanengwe et al., 2004; Sun et al., 2020), such that the differences in OC inputs had little 557 558 effect. Light textured soils have large pores which cannot protect SOC against microbial decomposition (Mtambanengwe et al., 2004; Christensen, 1987; Sun et al., 2020; Kravchenko 559 and Guber, 2017). Additionally, the low clay content meant less surface area for SOC 560 adsorption (Han et al., 2016; Churchman et al., 2020) which is another mechanism for SOC 561 protection from mineralization. In contrast, there were differences between NTM and NTR at 562 563 UZF in the top soil layers and intermediate between NTM and NTMR. Cumulative OC inputs in NTMR (12.4 Mg C ha<sup>-1</sup>) were about 75 % of cumulative OC inputs in NTM (16.2 Mg C ha<sup>-1</sup>) 564 1) (Figure 4) after 8 seasons. The added C, especially from maize stover mulch, most likely 565 was protected by clay particles as well as formation of organo-mineral complexes (Malepfane 566 et al., 2022; Chivenge et al., 2007; Jephita et al., 2023) which protects SOC from mineralization 567 (Dunjana et al., 2012; Shumba et al., 2020; Button et al., 2022; Rumpel et al., 2012; Sanaullah 568 569 et al., 2016).

570

# 571 **5.** Conclusions

572 Our study has shown the overarching importance of mulching and of combining at least two 573 CA principles to improve top SOC stocks. No tillage (NT) alone could not increase SOC stocks, 574 and even led to a slight decrease compared to CT, due to lower crop productivity in NT and 575 therefore reduced OC inputs to the soil. Nevertheless, whole profile (0-100 cm) SOC stocks 576 were the same between all the treatment. Our study also showed that sampling the entire soil 577 profile is necessary for a more accurate view of SOC accumulation potential among different 578 cropping systems.

# 580 6. Data availability

581 582	All data are freely available on the CIRAD data repository <u>https://doi.org/10.18167/DVN1/VPOCHN</u> (Shumba et al., 2023a).
583	
584	7. Author contributions
585	CT designed, established and maintained the experiments since 2013; RCa, RCh were involved
586	in soil sampling campaigns; AS, RCa performed the statistical analyses, graphics and drafting
587	the manuscript; AS, RCa, RCh, MC, JS, CT reviewed and edited the manuscript.
588	
589	8. Competing interests
590	One of the co-authors is a member of the editorial board of the SOIL journal. The other authors
591	have no competing interests to declare.
592	
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