

Increased soil organic carbon stocks under agroforestry: A survey of six different sites in France

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1	Increased soil organic carbon stocks under agroforestry: a survey of six different sites in
2	France
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13	
14	ABSTRACT
15	Agroforestry systems are land use management systems in which trees are grown in
16	combination with crops or pasture in the same field. In silvoarable systems, trees are
17	intercropped with arable crops, and in silvopastoral systems trees are combined with pasture

for livestock. These systems may produce forage and timber as well as providing ecosystem services such as climate change mitigation. Carbon (C) is stored in the aboveground and belowground biomass of the trees, and the transfer of organic matter from the trees to the soil can increase soil organic carbon (SOC) stocks. Few studies have assessed the impact of agroforestry systems on carbon storage in soils in temperate climates, as most have been

undertaken in tropical regions. This study assessed five silvoarable systems and one 23 silvopastoral system in France. All sites had an agroforestry system with an adjacent, purely 24 agricultural control plot. The land use management in the inter-rows in the agroforestry systems 25 26 and in the control plots were identical. The age of the study sites ranged from 6 to 41 years after tree planting. Depending on the type of soil, the sampling depth ranged from 20 to 100 cm and 27 SOC stocks were assessed using equivalent soil masses. The aboveground biomass of the trees 28 was also measured at all sites. In the silvoarable systems, the mean organic carbon stock 29 accumulation rate in the soil was 0.24 (0.09-0.46) Mg C ha⁻¹ yr⁻¹ at a depth of 30 cm and 0.65 30 (0.004-1.85) Mg C ha⁻¹ yr⁻¹ in the tree biomass. Increased SOC stocks were also found in deeper 31 32 soil layers at two silvoarable sites. Young plantations stored additional SOC but mainly in the soil under the rows of trees, possibly as a result of the herbaceous vegetation growing in the 33 rows. At the silvopastoral site, the SOC stock was significantly greater at a depth of 30 to 50 34 35 cm than in the control. Overall, this study showed the potential of agroforestry systems to store C in both soil and biomass in temperate regions. 36

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38 Keywords: Alley cropping, Soil organic carbon storage, Equivalent soil mass, Aboveground
39 biomass, Belowground biomass

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41 **1. Introduction**

42 Soils play an essential role in the global carbon budget (Houghton, 2007). Currently, the land 43 sink (including soil and vegetation) absorbs about 30% of the carbon (C) emitted to the 44 atmosphere through the burning of fossil fuel and cement production (Le Quéré et al., 2014). 45 Since 1850, the depletion of soil organic carbon (SOC) in cultivated lands has transferred about

70 Gt C to the atmosphere (Amundson, 2001; Lal, 2004a). The potential of these SOC depleted 46 47 soils as future C sinks through SOC sequestration has now been recognized (Paustian et al., 1997; Freibauer et al., 2004; Smith, 2004). In France, SOC stocks have been estimated at 3.1-48 3.3 Gt C in the top 30 cm of soils (Arrouays et al., 2001; Martin et al., 2011). Based on the SOC 49 saturation capacity (Hassink, 1997), assuming that the quantity of stable SOC is limited by the 50 amount of fine particles, Angers et al. (2011) found that the median saturation deficit of French 51 arable topsoils was 8.1 g C kg⁻¹ soil. About 70% of French agricultural topsoils are, therefore, 52 unsaturated in SOC and have the potential for additional SOC storage. Increasing SOC stocks 53 is often seen as a win-win strategy (Lal, 2004a; Janzen, 2006) as it allows the transfer of CO₂ 54 55 from the atmosphere to the soil while improving soil quality and fertility (Lal, 2004b).

Several agricultural practices have been developed to increase SOC stocks. For instance, the 56 introduction of cover crops (Constantin et al., 2010; Poeplau and Don, 2015) or grasslands 57 (Conant et al., 2001; Soussana et al., 2004) in the cropping sequence has proven effective. The 58 59 effect of no-till farming on SOC stocks is disputed and highly variable (Luo et al., 2010; Virto et al., 2012; Dimassi et al., 2013) and seems to depend on the amount of C transferred from the 60 crops to the soil (Virto et al., 2012). Agroforestry is a general term for agroecosystems in which 61 trees are intercropped with crops or pasture (Nair, 1993). Silvoarable systems intercrop trees 62 and arable crops and silvopastoral systems combine trees, pasture and livestock. These are 63 recognized as possible land use management systems that can maintain or increase SOC stocks, 64 65 both in tropical (Albrecht and Kandji, 2003) and temperate regions (Peichl et al., 2006; Bambrick et al., 2010; Wotherspoon et al., 2014). However, most studies only consider the 66 67 surface soil layers (to a depth of < 20 or 30 cm) whereas trees grown in agroforestry can be very deep rooted (Mulia and Dupraz, 2006; Cardinael et al., 2015a) and affect deep SOC stocks. 68 A recent study in the Mediterranean region of France showed that an 18-year-old silvoarable 69 70 system with hybrid walnuts intercropped with durum wheat increased SOC stocks by 0.25 \pm

0.03 Mg C ha⁻¹ yr⁻¹ in the 0-30 cm layer and by 0.35 ± 0.04 Mg C ha⁻¹ yr⁻¹ from 0 to 100 cm 71 72 compared to an adjacent agricultural plot (Cardinael et al., 2015b). Furthermore, although trees affect the spatial distribution of organic matter inputs to the soil (Rhoades, 1997), sampling 73 protocols have not always taken account of the potential impact on the spatial distribution of 74 SOC stocks. Some authors showed that SOC stocks were greater in the tree rows than in the 75 inter-rows, and found no gradients within the inter-rows (Peichl et al., 2006; Upson and 76 77 Burgess, 2013). Bambrick et al., (2010) found that the spatial distribution of SOC stocks varied with the time after tree planting. Few studies have estimated SOC storage in agroforestry 78 systems in temperate conditions (Howlett et al., 2011; Mosquera Losada et al., 2011; Upson 79 80 and Burgess, 2013) and these studies sometimes do not have control plots without trees for comparison, making it difficult to evaluate the precise effect of agroforestry on SOC stocks 81 (Pellerin et al., 2013). 82

This study set out i) to quantify organic carbon stocks in soils and in the tree biomass in six agroforestry systems with adjacent agricultural control plots under different soil and climate conditions in France, ii) to study the spatial distribution of SOC stocks as a function of the distance from individual trees and the tree rows and iii) to estimate the SOC stock accumulation rates for these agroforestry systems.

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89 2. Materials and methods

90 2.1 The six agroforestry sites

Each study site had an agroforestry system and an adjacent agricultural control plot. Before tree planting, the agroforestry plot was part of the agricultural plot, with the same soil use and management (crop rotation, fertilization, soil tillage). After tree planting, the soil management of the agroforestry inter-rows and of the agricultural plot remained identical. Rows of trees were planted in the agroforestry fields, with natural or sown grasses between the trees. Five
sites, Restinclières (RE), Châteaudun (CH), Melle (ME), Saint-Jean d'Angely (SJ), and
Vézénobres (VE), were silvoarable systems with no grazing. Only one site, Theix (TH), was a
silvopastoral system with regular grazing. Four sites were owned and managed by farmers and
Restinclières (RE) and Theix (TH) were experimental research sites.



- 103 Figure 1. Location and description of the six study cases under agroforestry systems sampled
- in France.

Table 1 Site characteristics.

Site	Mean annual temperature (°C)	Mean annual rainfall (mm)	allSoil typeSoil depthSoil texture(FAO)(cm)clay/silt/sand (g kg ⁻¹)		ture d (g kg ⁻¹)	Soil pH in water	
					Agroforestry	Control	
СН	11.1	595	Luvisol	0-30	200/700/100	190/710/100	7.0
ME	11.7	810	Luvisol	0-30	240/660/100	260/630/110	5.8
SJ	12.9	850	Luvisol	0-20	560/370/70	500/410/90	7.7
VE	14.5	1037	Fluvisol	0-30	110/410/480	90/370/540	8.3
				30-60	100/440/460	80/370/550	8.3
RE	15.4	873	Fluvisol	0-30	173/406/421	176/413/411	8.0
				30-50	178/416/406	177/421/402	8.1
				50-70	250/501/249	243/507/250	8.2
				70-100	309/582/109	307/586/107	8.3
TH	7.7	800	Andosol	0-20	340/300/360	380/360/260	6.5
				20-50	320/280/400	360/380/260	6.5

110 CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH: Theix.

Table 2 Description of the agroforestry plots.

Site	Tree species	Age (yrs)	Density (trees ha ⁻¹)	Distance between trees in tree rows (m)	Width of inter-rows (m)	Width of tree rows (m)	Area occupied by tree rows in the AF plot (%)	Crops
СН	Hybrid walnut	6	34	10	24	2	8	wheat, rapeseed
ME	Hybrid walnut	6	35	8	27	2	7	wheat, rapeseed, sunflower
SJ	Black walnut	41	102	7	12	2	14	sunflower, wheat, barley
VE	Hybrid walnut	18	100	10	9	2	18	rapeseed, wheat, potato, garlic
RE	Hybrid walnut	18	110	4-12	11	2	16	durum wheat, rapeseed, chickpea
TH	Wild cherry	26	200	7	No row	No row	No row	ryegrass, fescue

114 CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH: Theix.

The CH silvoarable site was located in Châteaudun (Fig. 1), in the department of Eure-et-Loir 115 (longitude 1°17'58'' E, latitude 48°06'08'' N, elevation 147 m a.s.l.). The mean temperature 116 was 11.1°C and the mean annual rainfall 595 mm (years 2001-2013, INRA CLIMATIK, 117 https://intranet.inra.fr/climatik). The soil was a silty loam Luvisol (IUSS Working Group WRB, 118 2007) (Table 1). Hybrid walnut trees (Juglans regia \times nigra cv. NG23) were planted in 119 February 2008 at a density of 34 trees ha⁻¹. The trees were planted 10 m apart within the rows, 120 with 26 m between rows. A mix of ryegrass (Lolium perenne L.) and tall fescue (Festuca 121 arundinacea Schreb.) was sown in August 2007 in two meter wide strips along the tree rows 122 before the trees were planted. After tree planting, wheat (Triticum aestivum L. subsp. aestivum) 123 124 and rapeseed (Brassica napus L.) were grown in rotation in the control plot and in the interrows (Table 2). The mean fresh grain yield was 7.5-8 t ha⁻¹ for wheat, and 3.8 t ha⁻¹ for rapeseed. 125 All crop residues were left in the field after harvest. The agroforestry inter-rows and the control 126 127 plot were ploughed every three years to a depth of 22 cm and harrowed to 8 cm the other years.

128 The ME silvoarable site was located in Melle (Fig. 1), in the department of Deux-Sèvres (longitude 0°10'37" W, latitude 46°11'54" N, elevation 107 m a.s.l.). The mean temperature 129 was 11.7°C and the mean annual rainfall 810 mm (years 1990-2013, INRA CLIMATIK, 130 https://intranet.inra.fr/climatik). The soil was a silty loam Luvisol (IUSS Working Group WRB, 131 2007) (Table 1). Hybrid walnut trees (Juglans regia × nigra cv. NG23) were planted in 2008 at 132 a density of 35 trees ha⁻¹. The trees were planted 8 m apart within the rows, with 29 m between 133 rows. Sheep fescue (Festuca ovina L.) was sown in 2008 in two meter wide strips along the 134 tree rows before the trees were planted. After tree planting, wheat (Triticum aestivum L. subsp. 135 136 aestivum), rapeseed (Brassica napus L.) and sunflower (Helianthus annuus L.) were grown in rotation in the control plot and in the inter-rows (Table 2). The mean fresh grain yield was 8-137 8.5 t ha⁻¹ for wheat, 3.3 t ha⁻¹ for rapeseed and 2.5 t ha⁻¹ for sunflower. Crop residues were 138 139 usually exported, but this was counterbalanced by the application of manure in both the

agroforestry inter-rows and the control plot (the farmer was unable to specify the application
rates, but they were similar for both plots). Before the spring crop (sunflower), a winter cover
crop was sown to prevent soil erosion and nitrate leaching. This cover crop was a mix of radish
(*Raphanus sativus* L.), phacelia (*Phacelia tanacetifolia* Benth.) and mustard (*Sinapis alba* L.).
The soil was ploughed every year to a depth of 20 cm in both the agroforestry inter-rows and
the control plot. The agroforestry system was established on a moderate slope, while the control
plot was flat.

The SJ silvoarable site was located in Saint-Jean-d'Angély (Fig. 1), in the department of 147 Charente-Maritime (longitude 0°13'57'' W, latitude 46°00'39'' N, elevation 152 m a.s.l.). The 148 mean temperature was 12.9°C and the mean annual rainfall 850 mm (years 1990-2013, INRA 149 CLIMATIK, https://intranet.inra.fr/climatik). The soil was a carbonated silty clay Luvisol 150 (IUSS Working Group WRB, 2007) (Table 1). Black walnut trees (Juglans nigra L.) were 151 planted in 1973 at a density of 102 trees ha⁻¹. The trees were planted 7 m apart within the tree 152 153 rows, with 14 m between rows. The rows of trees were two meters wide, and covered by spontaneous herbaceous vegetation. After tree planting, sunflower (Helianthus annuus L.), 154 wheat (Triticum aestivum L. subsp. aestivum) and barley (Hordeum vulgare L.) were grown in 155 156 rotation in the control plot and in the inter-rows (Table 2). Crop residues were left in the field after harvest. The soil was ploughed every three years to a depth of 10-20 cm in both the 157 agroforestry inter-rows and the control plot. 158

The VE silvoarable site was located in Vézénobres (Fig. 1), in the department of Gard (longitude 4°06'37'' E, latitude 46°00'39'' N, elevation 102 m a.s.l.). The climate was subhumid Mediterranean with a mean temperature of 14.5°C and a mean annual rainfall of 1037 mm (mean 1995-2007, experimental site weather station). The soil was a deep sandy loam alluvial Fluvisol (IUSS Working Group WRB, 2007) (Table 1) originating from deposits from the granitic Cevennes mountain range and was, therefore, not calcareous. Hybrid walnut trees

(Juglans regia \times nigra cv. NG23) were planted in 1995 at a density of 100 trees ha⁻¹. The trees 165 166 were planted 10 m apart with the rows, with 10 m between rows. The tree rows were two meters wide and were covered by spontaneous herbaceous vegetation. In the inter-rows, rapeseed 167 168 (Brassica napus L.) and wheat (Triticum aestivum L. subsp. aestivum) were grown in rotation until 2010 (Table 2). In 2011, the farm changed over to organic farming and potatoes were 169 planted (Solanum tuberosum L.). In 2012 garlic (Allium sativum L.) was grown in the inter-170 rows. In 2013 the inter-rows were left fallow and in 2014 sunflower (Helianthus annuus L.) 171 was sown. The same crops were grown in the control plot, except in 2011 when wheat (Triticum 172 aestivum L. subsp. aestivum) was sown and in 2012 when the control was left fallow. The soil 173 174 was occasionally ploughed to a depth of 20 cm in both the agroforestry inter-rows and the control plot. 175

The RE site was located in Prades-le-Lez, at the Restinclières experimental site (Fig. 1), in the 176 department of Hérault (longitude 04°01' E, latitude 43°43' N, elevation 54 m a.s.l.). A full 177 178 description of this site is given in the study by Cardinael et al. (2015b). The climate was subhumid Mediterranean with a mean temperature of 15.4°C and a mean annual rainfall of 873 179 mm (years 1995-2013, experimental site weather station). The soil was a deep carbonated 180 sandy loam Fluvisol (IUSS Working Group WRB, 2007) (Table 1). Hybrid walnut trees 181 (Juglans regia \times nigra cv. NG23) were planted in 1995 and the density was 110 trees ha⁻¹ at the 182 time of the study (Table 2). The trees were planted 4 to 8 m apart along the rows with 13 m 183 between rows. The two meter wide tree rows were covered by spontaneous herbaceous 184 vegetation. They were mainly intercropped with durum wheat (Triticum turgidum L. subsp. 185 186 durum) but also with rapeseed (Brassica napus L.) and chickpea (Cicer arietinum L.). The soil was regularly ploughed to a depth of 20 cm in both the agroforestry inter-rows and the control 187 plot. 188

The TH silvopastoral site was located at the Theix experimental site (Fig. 1), in the department 189 of Puy-de-Dôme (longitude 3°01'39'' E, latitude 45°42'58'' N, elevation 829 m a.s.l.). The 190 mean temperature was 7.7°C and the mean annual rainfall 800 mm (years 1990-2013, INRA 191 CLIMATIK, https://intranet.inra.fr/climatik). The soil was a clay loam Andosol (IUSS 192 Working Group WRB, 2007) (Table 1). Wild cherry trees (Prunus avium L.) were planted in 193 1988 at a density of 200 trees ha⁻¹ on a natural permanent pasture. The trees were planted 7 m 194 apart and the soil was uniformly covered by a permanent pasture, mainly ryegrass (Lolium 195 perenne L.) and fescue (Festuca sp.), in both the control and agroforestry plots (Table 2). There 196 was no distinction between tree rows and inter-rows in terms of soil cover and management. 197 198 The pasture was regularly grazed by sheep in both the control and agroforestry plots.

199

200 2.2 Soil sampling protocol

201 The sampling protocol was defined to allow for the spatial distribution of SOC stocks owing to 202 the presence of trees and rows of trees, with sampling points at varying distances from the trees. The agroforestry designs varied between sites with different distances between the trees within 203 the rows and between the rows. The sampling protocol was flexible to take account of these 204 differences but consistent enough to allow comparisons between sites. A sampling pattern was 205 defined with sampling points in transects around one tree. This was a rectangle with dimensions 206 $\frac{L}{2} \times \frac{d}{2}$, where L is the distance between tree rows and d is the distance between trees in the rows 207 (Fig. 2). This pattern is a quarter of the Voronoi polygon which is the elementary space defined 208 by the half distances between the sampled tree and its neighbors, as commonly used to estimate 209 210 root biomass (Levillain et al., 2011; Picard et al., 2012). At all sites, nine soil samples per pattern were taken at fixed positions around the trees, at 1, 2 and 3 m in the tree row, in the 211 inter-row in front of the tree, and in the inter-row between two trees. If $L \ge 8$ m, soil samples 212

were additionally taken at mid-distance $\frac{L}{2}$, and, if $L \ge 16$ m, soil samples were also taken at $\frac{L}{4}$. If 213 $d \ge 8$ m, soil samples were also taken at $\frac{d}{2}$. This sampling pattern was applied three times in the 214 215 agroforestry plots at all sites. Two sampling patterns were oriented north of the tree rows (if the rows were oriented east-west) or west of the rows (if the rows were oriented north-south) and 216 217 one sampling pattern was oriented south or east, respectively. Thirty-six sampling points were, therefore, defined for the agroforestry plot at the CH site, twenty-four at the SJ site, thirty at the 218 VE site, and twenty-seven at the TH site (Table 3). In the control plots, a simpler sampling 219 pattern was applied in triplicate. This pattern was a rectangle with dimensions $\frac{L}{2} \times \frac{d}{2}$, with soil 220 samples taken at each corner (12 sampling points). 221

222 At the ME site, the agricultural control plot was flat, whereas the agroforestry plot was on a moderate slope. The SOC-rich topsoil in the agroforestry plot might, therefore, have been 223 eroded before the start of the experiment. To take account of this topography difference, six 224 225 soil samples from the middle of the inter-row (two sampling positions for each of the three sampling patterns) were used as an alternate arable control. Because the inter-rows were 27 m 226 227 wide and the 6-year-old trees were only 3 m high, the soil in the middle of the inter-rows had probably not yet been affected by the presence of trees. 30 sampling points were defined in the 228 agroforestry plot and 6 in the control plot (Table 3). 229

The RE site had been the subject of a previous study (Cardinael et al., 2015b) to map SOC stocks at plot scale. The sampling protocol at this site was, therefore, very dense: 100 soil samples were taken from the agroforestry plot and 93 from the control plot (Table 3). Sampling points were located every 5 m along a regular grid (25×25 m), and at 1, 2 and 3 m around nine walnut trees, in the inter-rows and in the tree rows.

The sampling depths were 30 cm at the CH and ME sites, 20 cm at the SJ site, 60 at the VE site, 100 at the RE site and 50 cm at the TH site. At the SJ site, the sampling depth corresponded

to the maximum soil depth. Soil samples were taken every 10 cm depth from the surface, exceptat the RE site (at 10 cm and every 20 cm from 10 cm).



Figure 2. Sampling pattern for the agroforestry sites (except for the RE site). *L* is the distance
between tree rows, *d* is the distance between trees on the rows.

243 2.3 Bulk density measurement

The soil samples were collected in April 2014 at all sites, except at the RE site which was sampled in May 2013. Soil samples were taken every 10 cm from the surface using a 500-cm³ cylinder, except at the RE site where soil samples were taken every 20 cm depth after the top 10 cm. After air-drying in the lab, the soil samples were oven-dried at 105°C for 48 hours, sieved to 2 mm and weighed without coarse particles > 2 mm. The bulk density (g cm⁻³) was calculated as the ratio of the dry mass of fine soil (< 2 mm) to the cylinder volume.

250

251 2.4 Organic carbon analysis

The soil samples were dried at 40°C and ball milled until they passed through a 200 µm mesh 252 253 sieve. The presence of inorganic carbon was tested with HCl. If the soil contained inorganic 254 carbon, carbonates were removed by acid fumigation, as described in Harris et al. (2001). This was the case for samples from the SJ and RE sites. 30 mg of soil were placed in open Ag-foil 255 256 capsules. The capsules were then placed in the wells of a microtiter plate and 50 µL of demineralized water was added to each capsule. The microtiter plate was placed in a vacuum 257 desiccator with a beaker filled with 100 mL of concentrated HCl. The samples were exposed to 258 HCl vapor for 8 h and then dried at 40°C for 48 h. The capsules were then enclosed in a bigger 259 tin capsule. All samples were analyzed for organic carbon concentration using a CHN elemental 260 261 analyzer (Carlo Erba NA 2000, Milan, Italy).

262

263 2.5 SOC stock calculation

The SOC stock at soil sample level (mg C cm⁻³) is defined as the product of the SOC concentration (mg C g^{-1}) and the bulk density (g cm⁻³) and is then calculated for each soil profile

(kg C m⁻²) by summing the SOC stocks in the samples through the profile. For each site, the 266 SOC stocks were calculated on an equivalent soil mass (ESM) basis (Ellert and Bettany, 1995) 267 to enable comparison between all locations (control, tree rows, inter-rows) even where the soil 268 bulk density varied within the same site. SOC stocks in the agroforestry plot (Mg C ha⁻¹) were 269 calculated by adding the tree row and inter-row SOC stocks, weighted by their respective 270 relative surface areas: 271 $SOC \operatorname{stock}_{Agroforestry} = \frac{p \times SOC \operatorname{stock}_{Tree \, row} + (100 - p) \times SOC \operatorname{stock}_{Inter-row}}{100} \quad (1)$ 272 273 where *p* is the percentage of tree row surface area in the agroforestry plot (Table 2). 274 275 The delta SOC stock (Mg C ha⁻¹) at a given depth was expressed as the difference in the SOC 276 stock between the agroforestry and the control plot: 277 $\Delta_{SOC \ stock} = \text{SOC \ stock}_{Agroforestry} - \text{SOC \ stock}_{Control} (2)$ 278 279 The SOC stock accumulation rates under an agroforestry system at a given depth was calculated 280 by dividing the delta SOC stock by the number of years since tree planting. 281 282 283 2.6 Tree aboveground and belowground biomass 284 At each site, 10 to 20 trees were measured to estimate the aboveground biomass. As the trees 285 286 in the farmers' fields could not be felled, the aboveground biomass was estimated by multiplying the volume of the trunk and branches by the wood density, using the global wood 287 density database (Chave et al., 2009). The trunk volume was estimated as the sum of the volume 288 289 of three truncated cones, from the soil surface up to 1.30 m, from 1.30 m to the first branch and

from the first branch to the top of the tree. The trunk diameter was measured 5 cm above the 290 291 soil surface, at 1.30 m (Diameter at Breast Height, DBH) and below the first branch. The total height (H_{tot}) and merchantable height (H) of the trees were also measured. The volume of the 292 first order branches (branches arising directly off the trunk) was also estimated by measuring 293 the diameter of the branches at the trunk and the length of the branches and branch volumes 294 were calculated as cone volumes. For the RE site, three trees were felled to measure the trunk 295 296 and branch biomass directly. The carbon concentrations of the trunk and branches of the Juglans regia \times nigra cv. NG23 were measured. As it was not possible to sample wood from 297 the tree trunks at the other sites, the C concentrations were considered to be the same for Prunus 298 299 avium and Juglans nigra. This simplification was possible because these trees are slow growing species and there is usually little variation in their wood C concentration (462.7 to 499.7 mg C 300 g⁻¹ DM) (Lamlom and Savidge, 2003). It was also assumed that young and old trees had the 301 302 same wood density and C concentration.

303 So far as we are aware, there is no allometric equation for estimating the belowground biomass 304 of temperate agroforestry trees and so the equation proposed by Cairns et al. (1997) for 305 temperate forests was used:

$$RB = e^{-1.3267 + 0.8877 \times \ln(AB) + 0.1045 \times \ln(Age)}$$
(3)

where RB is the total root biomass (Mg C ha⁻¹), AB is the aboveground biomass (Mg C ha⁻¹)
and *Age* is the age of the plantation (yr).

309

310 2.7 Statistical analyses

The influence of the sampling location in the inter-rows (in front of a tree or between two trees)on the SOC concentration, bulk density and SOC stock was determined using mixed effects

models. This analysis was done at each site using the *nlme* package (Pinheiro et al., 2013). An 313 314 ANOVA was performed on these models. Mixed effects models were then fitted for each site using the whole soil data set. The SOC concentration, bulk density and SOC stock were 315 316 compared as a function of depth, location (control, tree row, inter-row) and distance from the closest tree. An ANOVA was performed on these models. The SOC stock were compared 317 between tree rows and inter-rows, between inter-rows and the control plot and between the 318 319 agroforestry plot and the control plot. The statistical analyses were performed using R version 3.1.1 (R Development Core Team, 2013), at a significance level of < 0.05. 320

321

322 **3. Results**

323 *3.1 Soil bulk density*

At all sites, the soil bulk density increased significantly with increasing soil depth (Table 3, S1). 324 In the top 30 cm, the bulk density ranged from 0.7 to 1.6 g cm⁻³ depending on the site. There 325 326 was no significant difference in bulk density between the tree row and the inter-row except in the top 10 cm at the ME, SJ and RE sites, where it was lower in the tree row than in the inter-327 row and in the control (Table 3, S1). There was no significant difference between the control 328 and the inter-row at any depth, except at the RE site where the bulk density was higher in the 329 top 10 cm in the control plot (Table 3, S1).. The distance from the closest tree had no significant 330 331 effect on the bulk density except at the SJ site (Table S1). There was no significant difference in the inter-row between samples collected in front of a tree or between two trees at any of the 332 sites or at any depth (p-value ≥ 0.18), except at the ME site (p-value = 0.03) (Table S1). 333

334

		Number of soil samples			Bu	Bulk density (g cm ⁻³)			SOC concentration (mg C g ⁻¹)		
Site	Soil depth (cm)	Tree row	Inter-row	Control	Tree row	Inter-row	Control	Tree row	Inter-row	Control	
	0-10	12	24	12	1.09 ± 0.03	1.10 ± 0.02	1.18 ± 0.02	19.44 ± 1.00	16.44 ± 0.26	14.88 ± 0.38	
CH	10-20	12	24	12	1.12 ± 0.02	1.13 ± 0.02	1.16 ± 0.03	13.58 ± 0.31	14.39 ± 0.34	14.56 ± 0.48	
	20-30	12	24	12	1.15 ± 0.02	1.20 ± 0.01	1.25 ± 0.02	11.76 ± 0.65	12.07 ± 0.48	11.78 ± 0.35	
	0-10	12	18	6	1.04 ± 0.03	1.27 ± 0.02	1.31 ± 0.01	21.30 ± 0.63	13.01 ± 0.19	12.80 ± 0.43	
ME	10-20	12	18	6	1.28 ± 0.02	1.29 ± 0.02	1.37 ± 0.03	13.14 ± 0.26	12.03 ± 0.50	12.02 ± 0.40	
	20-30	12	18	6	1.21 ± 0.01	1.34 ± 0.01	1.35 ± 0.02	10.35 ± 0.21	8.38 ± 0.44	8.68 ± 0.93	
C I	0-10	8	16	12	0.67 ± 0.03	0.76 ± 0.02	0.78 ± 0.01	58.60 ± 1.88	49.49 ± 1.28	32.89 ± 0.33	
21	10-20	8	16	12	0.84 ± 0.03	0.78 ± 0.03	0.88 ± 0.04	35.60 ± 0.82	32.01 ± 0.67	24.86 ± 1.12	
	0-10	12	18	10	1.06 ± 0.04	0.98 ± 0.03	0.91 ± 0.02	17.25 ± 0.49	15.95 ± 0.37	15.00 ± 1.11	
	10-20	12	18	10	1.12 ± 0.02	1.18 ± 0.02	1.24 ± 0.03	13.72 ± 0.40	13.50 ± 0.49	13.19 ± 0.70	
	20-30	12	18	10	1.16 ± 0.03	1.25 ± 0.01	1.31 ± 0.02	11.38 ± 0.30	10.83 ± 0.25	10.89 ± 0.68	
VE	30-40	12	18	10	1.29 ± 0.04	1.39 ± 0.02	1.47 ± 0.04	10.82 ± 0.27	10.31 ± 0.29	8.55 ± 0.78	
	40-50	12	18	10	1.30 ± 0.05	1.37 ± 0.03	1.34 ± 0.03	10.52 ± 0.33	8.25 ± 0.35	5.79 ± 0.69	
	50-60	12	18	10	1.36 ± 0.04	1.39 ± 0.04	1.37 ± 0.06	9.74 ± 0.35	7.16 ± 0.62	5.28 ± 0.86	
	0-10	40	60	93	1.10 ± 0.02	1.23 ± 0.03	1.41 ± 0.01	21.59 ± 0.76	9.78 ± 0.13	9.33 ± 0.06	
	10-30	40	60	93	1.49 ± 0.01	1.60 ± 0.02	1.61 ± 0.00	10.16 ± 0.16	9.57 ± 0.12	8.94 ± 0.05	
RE	30-50	40	60	93	1.71 ± 0.01	1.67 ± 0.02	1.73 ± 0.00	7.29 ± 0.15	6.95 ± 0.11	6.82 ± 0.10	
	50-70	40	60	93	1.73 ± 0.01	1.77 ± 0.01	1.80 ± 0.00	6.07 ± 0.11	5.89 ± 0.07	5.77 ± 0.06	
	70-100	40	60	93	1.68 ± 0.00	1.71 ± 0.00	1.74 ± 0.00	6.49 ± 0.16	6.29 ± 0.06	6.09 ± 0.06	
	0-10		27	10	0.75	5 ± 0.02	0.69 ± 0.02	64.00	± 2.40	67.83 ± 2.45	
	10-20		27	10	0.79	0 ± 0.01	0.75 ± 0.01	46.97	± 1.15	49.31 ± 0.89	
TH	20-30		27	10	0.80	0 ± 0.02	0.73 ± 0.02	38.82	± 0.88	40.56 ± 0.86	
	30-40		27	10	0.82	2 ± 0.01	0.78 ± 0.02	32.90	± 0.70	29.92 ± 0.75	
	40-50	1	19	10	0.80) ± 0.01	0.79 ± 0.03	28.65	± 0.76	22.69 ± 1.25	

Table 3 Mean soil bulk density (g cm⁻³) and mean soil organic carbon (SOC) concentrations (mg C g^{-1}) with associated standard errors.

- 338 At the TH silvopastoral site, no distinction was made between tree rows and inter-rows (uniform cover), values are for the whole agroforestry plot.
- 339 CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH: Theix.



Figure 3. Soil organic carbon concentration (mg C g^{-1}) at the different sites. Transparent rectangles represent standard errors. At the TH silvopastoral site, no distinction was made between tree rows and inter-rows (uniform cover), values are for the whole agroforestry plot.

345 The SOC concentration decreased significantly with increasing soil depth, except in the ploughed layer, where it was uniform (Table 3, S1). At all sites, the SOC concentration in the 346 top 10 cm was significantly higher in the tree row than in the inter-row (Fig. 3). However, there 347 was no significant difference in the inter-row between samples collected in front of a tree and 348 between two trees (p-value ≥ 0.32) at any site and at any depth. The SOC concentration 349 depended significantly on the distance from the trees only at the oldest site (SJ, p-value < 0.001) 350 (Table S1). At sites CH, SJ and RE, the SOC concentration in the top 10 cm was significantly 351 higher in the inter-rows than in the control plot (Fig. 3, Table 3). At the VE and RE silvoarable 352 sites, the SOC concentration was significantly higher in the inter-row than in the control below 353 30 cm (Fig. 3, Table 3). At the TH silvopastoral site, the SOC concentration below 30 cm was 354 also significantly higher in the silvopasture than in the tree-less pasture (Fig. 3, Table 3). 355

357 *3.3 Soil organic carbon stock*

The SOC stock was mainly influenced by depth and location (Table S1). In the inter-row, there 358 was no significant difference between samples collected in front of a tree and between two trees 359 (p-value ≥ 0.30). The distance from the closest tree had no significant effect on the SOC stock 360 $(p-value \ge 0.5)$ except at the SJ site (p-value = 0.005) (Table S1). In the silvoarable systems, 361 362 the SOC stock was significantly higher in the tree rows than in the inter-rows in the top 10 cm, even in young plantations (CH and ME sites) (Fig. 4). The SOC stock was also significantly 363 higher in the inter-rows than in the control at depths of 10 cm at the CH site, 20 cm at the SJ 364 365 site and 30 cm at the RE site, as happened for SOC concentration (Fig. 4). Unlike, at the VE site, the SOC stock was higher in the inter-rows than in the control below 30 cm (Fig. 4). At 366

the TH silvopastoral site, the SOC stock below 30 cm was higher in the agroforestry plot thanin the control.

369 In the top 30 cm, the delta SOC stock between silvoarable systems and control plots was significantly positive except at the ME and VE sites (Table 4). For the silvoarable sites, the 370 delta SOC stock ranged from 0.5 to 4.5 Mg C ha⁻¹ in the top 30 cm (Table 4), and was about 19 371 Mg C ha⁻¹ in the top 20 cm for the oldest silvoarable system (SJ). At the RE and VE silvoarable 372 sites, the delta SOC stock was significantly positive below 30 cm depth. At the TH silvopastoral 373 site, the delta SOC stock was not significantly different in the top 30 cm (-0.16 \pm 0.25 Mg C 374 ha⁻¹) but was significantly positive for the whole soil profile $(0.49 \pm 0.27 \text{ Mg C ha}^{-1})$ down to 375 60 cm (Table 4). 376

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378 *3.4 Carbon stock in the tree biomass*

The wood density of Juglans regia \times nigra cv. NG23 was 0.62 g cm⁻³, that of Juglans nigra 379 was 0.59 g cm⁻³ and that of *Prunus avium* was 0.54 g cm⁻³. The C concentrations of the trunk 380 and branches of 18-year-old Juglans regia \times nigra cv. NG23 were 445.71 \pm 1.04 and 428.64 \pm 381 1.70 mg C g⁻¹ DM, respectively. At the silvoarable sites, the organic carbon stocks in the 382 aboveground biomass of the trees ranged from 0.02 to 26.64 Mg C ha⁻¹ depending on the tree 383 density and age (Table 5). The aboveground tree C stock was the highest at the silvopastoral 384 site, reaching about 37 Mg C ha⁻¹ The estimated C stocks in the tree belowground biomass 385 ranged from 0.01 to 6.61 Mg C ha⁻¹ at the silvoarable sites and was more than 9 Mg C ha⁻¹ at 386 the TH silvopastoral site (Table 5). 387

388

Site	Cumulativ e ESM (Mg ha ⁻¹)	Approximate soil depth (cm)	Cumulative SOC stock (Mg C ha ⁻¹)			$\Delta_{SOC \ stock}$ (Mg C ha ⁻¹)	SOC stock accumulation rate (Mg C ha ⁻¹ yr ⁻¹)			
	(8)	(000)	Tree row	Inter row	AF	Control	AF – Control	AF/Control	Tree row/Control	Inter-row/Control
	1000	0-10	19.4 ± 1.0	16.4 ± 0.3	16.7 ± 0.3	14.9 ± 0.4	$1.8 \pm 0.5^{*}$	$0.30 \pm 0.08^{*}$	$0.76 \pm 0.18^{*}$	$0.26 \pm 0.08^{*}$
CH	2100	0-20	34.8 ± 1.2	32.5 ± 0.5	32.7 ± 0.5	31.0 ± 0.9	$1.7 \pm 1.0^{*}$	$0.28 \pm 0.17 ^{\boldsymbol{*}}$	$0.63 \pm 0.25^{*}$	$0.25 \pm 0.17^{*}$
	3250	0-30	48.4 ± 1.7	46.6 ± 1.0	46.7 ± 1.0	45.0 ± 1.1	$1.7 \pm 1.4^{*}$	$0.29\pm0.24^{\ast}$	$0.57 \pm 0.33 *$	$0.27\pm0.25^{\ast}$
	1000	0-10	21.2 ± 0.6	13.0 ± 0.2	13.6 ± 0.2	12.2 ± 0.3	$1.4 \pm 0.4*$	$0.24 \pm 0.07*$	$1.50 \pm 0.11^{*}$	$0.14 \pm 0.07*$
ME	2200	0-20	37.2 ± 0.6	27.7 ± 0.5	28.4 ± 0.5	26.4 ± 0.9	$2.0 \pm 1.1^{*}$	$0.33 \pm 0.18^{*}$	$1.79 \pm 0.19^{*}$	$0.22 \pm 0.18^*$
	3500	0-30	51.1 ± 0.8	39.9 ± 0.9	40.7 ± 0.9	40.1 ± 1.7	0.5 ± 2.0	0.09 ± 0.33	$1.83\pm0.32^{\ast}$	$\textbf{-0.04} \pm 0.33$
CT.	700	0-10	40.6 ± 1.1	34.6 ± 0.9	35.5 ± 0.8	23.0 ± 0.2	$12.4 \pm 0.8^{*}$	$0.30 \pm 0.02^{*}$	$0.43 \pm 0.03^{*}$	$0.28 \pm 0.02*$
SJ	1450	0-20	67.7 ± 1.1	59.8 ± 1.0	60.9 ± 0.9	42.1 ± 0.8	$18.8 \pm 1.2^{\texttt{*}}$	$0.46\pm0.03^{\ast}$	$0.62 \pm 0.03^{*}$	$0.43\pm0.03^{\ast}$
	900	0-10	15.5 ± 0.5	14.6 ± 0.4	14.8 ± 0.3	13.5 + 1.0	1.3 + 1.0*	$0.07 + 0.06^{*}$	0.11 + 0.06*	$0.06 + 0.06^{*}$
	2000	0-20	31.2 ± 0.8	29.5 ± 0.8	29.8 ± 0.6	27.9 ± 1.5	$1.9 \pm 1.6^{*}$	$0.11 \pm 0.09^*$	$0.18 \pm 0.09^*$	$0.09 \pm 0.09^*$
	3150	0-30	44.7 ± 1.0	42.4 ± 0.9	42.8 ± 0.8	40.8 ± 2.0	2.0 ± 2.2	0.11 ± 0.12	$0.21 \pm 0.12^*$	0.09 ± 0.12
VE	4400	0-40	58.1 ± 1.2	55.1 ± 1.2	55.7 ± 1.0	51.8 ± 2.5	$3.9 \pm 2.7 *$	$0.22 \pm 0.15^{*}$	$0.35 \pm 0.16^{\ast}$	$0.19 \pm 0.16 ^{\ast}$
	5700	0-50	72.0 ± 1.5	66.8 ± 1.3	67.7 ± 1.1	61.2 ± 3.2	$6.5 \pm 3.4^{*}$	$0.36 \pm 0.19^{*}$	$0.60 \pm 0.20^{*}$	$0.31 \pm 0.19 \texttt{*}$
	7050	0-60	85.3 ± 1.9	77.1 ± 1.6	78.6 ± 1.4	68.6 ± 4.1	$10.0\pm4.3^{*}$	$0.56\pm0.24^{*}$	$0.93 \pm 0.25 *$	$0.48\pm0.25^{\ast}$
	1000	0-10	21.6 ± 1.0	9.8 ± 0.4	11.7 ± 0.3	9.3 ± 0.1	$2.3 \pm 0.4*$	$0.13 \pm 0.02^{*}$	$0.68 \pm 0.05 *$	$0.02 \pm 0.02*$
	4000	0-30	52.8 ± 1.4	37.9 ± 0.6	40.3 ± 0.5	35.8 ± 0.2	$4.5 \pm 0.6^{*}$	$0.25 \pm 0.03^*$	$0.95 \pm 0.08*$	$0.12 \pm 0.03^{*}$
RE	7300	0-50	77.1 ± 1.5	62.0 ± 0.7	64.4 ± 0.6	59.4 ± 0.2	$5.0 \pm 0.6^{*}$	0.28 ± 0.04 *	$0.98 \pm 0.08*$	$0.14 \pm 0.04^{*}$
	10700	0-70	98.1 ± 1.5	82.4 ± 0.7	84.9 ± 0.6	79.7 ± 0.3	$5.1 \pm 0.7^{*}$	$0.29 \pm 0.04^{*}$	$1.02 \pm 0.08*$	$0.15 \pm 0.04*$
	15700	0-100	130.4 ± 1.5	113.7 ± 0.7	116.4 ± 0.7	110.1 ± 0.3	$6.3\pm0.7^{*}$	$0.35\pm0.04^{\boldsymbol{*}}$	$1.13\pm0.09^{\texttt{*}}$	$0.20\pm0.05^{\ast}$
	700	0-10	_	-	44.2 ± 3.4	47.1 ± 1.6	-2.9 ± 3.8	-0.11 ± 0.14	-	-
	1450	0-20	_	-	80.4 ± 5.0	84.1 ± 1.9	-3.7 ± 5.3	-0.14 ± 0.20	-	-
TH	2200	0-30	-	-	110.2 ± 6.1	114.3 ± 2.3	-4.1 ± 6.5	-0.16 ± 0.25	-	-
	3000	0-40	-	-	137.6 ± 6.5	138.2 ± 2.3	-0.5 ± 6.9	-0.02 ± 0.26	-	-
	3800	0-50	-	-	169.3 ± 6.5	156.5 ± 2.7	$12.8 \pm 7.0^{*}$	$0.49 \pm 0.27^{*}$	-	-

390	Table 4 Soil organic carbo	n stock (Mg C ha ⁻¹) and SOC stock accumulation	n rate (Mg C ha ⁻¹ yr ⁻¹).
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Associated errors are standard errors. Approximate depths are presented here to give a better understanding of the ESM for a given site but do not correspond to the precise mass of the profile, which may vary between tree rows, inter-rows and the control (Ellert and Bettany, 1995). At the TH silvopastoral site, no distinction was made between tree rows and inter-rows (uniform cover). Significantly different (p-value < 0.05) delta SOC stock ($\Delta_{SOC \ stock}$) and additional SOC storage rate are followed by *. ESM: Equivalent Soil Mass, AF: Agroforestry. CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH: Theix.

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Figure 4. Soil organic carbon stock (kg C m⁻³) at the different sites. Bars represent standard errors. Approximate depths are presented but refer to
 equivalent soil mass. At the TH silvopastoral site, no distinction was made between tree rows and inter-rows (uniform cover), values are
 for the whole agroforestry plot.



Figure 5. Total organic carbon stock (Mg C ha⁻¹) of the different sites. AF: agroforestry, C: agricultural control. SOC: Soil organic carbon, ABG:
Aboveground, BLG: Belowground. CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH:
Theix. Studied depths vary between sites: 30 cm for CH, 30 cm for ME, 20 cm for SJ, 60 cm for VE, 100 cm for RE and 50 cm for TH.
Different lowercase letters indicate a significant (p-value < 0.05) difference of SOC stock between AF and C plots per site, and different
uppercase letters indicate a significant difference (p-value < 0.05) in the total organic carbon stock between AF and C plots per site.

409 *3.5 Total carbon stock of the different systems*

At the silvoarable sites, the total C stock (SOC + biomass) ranged from about 50 Mg C ha⁻¹ to 125 Mg C ha⁻¹, and reached 220 Mg C ha⁻¹ at the TH silvopastoral site (Fig. 5). The total C stock was always higher in the agroforestry systems than in the control plots. In the young plantations (CH and ME), the total C stock was mainly SOC, with tree C stock accounting for less than 0.01% of the total C stock. At oldest sites, up to 75% of the difference between total C stock in the agroforestry systems and control plots was explained by the tree biomass (Fig. 5).

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418 3.6 Organic carbon accumulation rate in soil and tree biomass

The mean SOC stock accumulation rate in the top 30 cm in the silvoarable systems was 0.18 Mg C ha⁻¹ yr⁻¹ (0.09 to 0.29 Mg C ha⁻¹ yr⁻¹). This rate reached 0.24 Mg C ha⁻¹ yr⁻¹ when the SJ silvoarable site and its shallow soil (20 cm) was taken into account. At the RE site, the SOC stock accumulation rate was 0.25 Mg C ha⁻¹ yr⁻¹ in the top 30 cm, and 0.35 Mg C ha⁻¹ yr⁻¹ in the top 100 cm, with a SOC stock accumulation rate of about 0.1 Mg C ha⁻¹ yr⁻¹ in the 30-100 cm layer (Table 4). Tree rows contributed about 20% to 50% to the SOC stock accumulation rate although they covered only 7% to 18% of the agroforestry surface area.

The C accumulation rate in the tree biomass in CH and ME young plantations was negligible (0.004 and 0.02 Mg C ha⁻¹ yr⁻¹, respectively) (Table 5). In the older and denser silvoarable sites, this rate ranged from 0.62 to 1.85 Mg C ha⁻¹ yr⁻¹, and was 1.76 Mg C ha⁻¹ yr⁻¹ at the TH silvopastoral site (Table 5).

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+52 Fuble 5 files, above ground and below ground carbon blocks at the various bite	432	Table 5 Tree characteristics	, aboveground and	belowground car	bon stocks at the	various sites.
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Site	Age (yr)	DBH (cm)	Height of merchantable timber (m)	Total height (m)	C stock of merchantable timber (kg C tree ⁻¹)	ABG tree C stock (kg C tree ⁻¹)	ABG tree C stock (Mg C ha ⁻¹)	Estimated BEG tree C stock (Mg C ha ⁻¹)	Estimated total tree C stock accumulation rate (Mg C ha ⁻¹ yr ⁻¹)
СН	6	2.6 ± 0.2	1.45 ± 0.04	2.12 ± 0.11	0.44 ± 0.06	0.49 ± 0.07	0.017 ± 0.002	0.01 (0.01-0.01)	0.004 ± 0.0004
ME	6	5.5 ± 0.3	1.13 ± 0.03	3.18 ± 0.13	1.18 ± 0.12	2.07 ± 0.19	0.073 ± 0.007	0.03 (0.03-0.04)	0.02 ± 0.001
SJ	41	29.9 ± 1.3	3.11 ± 0.23	13.18 ± 0.10	41.44 ± 2.36	194.56 ± 14.94	19.85 ± 1.52	5.55 (3.28-9.38)	0.62 ± 0.10
VE	18	31.7 ± 1.5	4.17 ± 0.18	15.52 ± 0.36	56.85 ± 3.77	266.44 ± 19.90	26.64 ± 1.99	6.61 (4.00-10.95)	1.85 ± 0.27
RE	18	25.5 ± 1.4	4.49 ± 0.39	11.21 ± 0.65	46.23 ± 2.47	98.93 ± 7.80	10.88 ± 0.86	2.99 (1.89-4.72)	0.77 ± 0.11
TH	26	30.7 ± 1.4	4.10 ± 0.23	14.70 ± 0.32	53.80 ± 1.76	183.46 ± 2.66	36.69 ± 0.53	9.13 (5.34-15.63)	1.76 ± 0.25

433 Errors represent standard errors. Number of measured trees: CH=24, ME=20, SJ=10, VE=10, RE=9 except for biomass measurements where n=3,

434 and TH=10. Values in brackets represent the 95% prediction interval for estimating the belowground biomass (Cairns et al., 1997). ABG:

435 Aboveground, BEG: Belowground. CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH: Theix.



Figure 6. SOC stock accumulation rates as a function of plantation age. Values are for the
approximate top 30 cm, except for the SJ site (approximate top 20 cm, maximum soil
depth).

441 **4. Discussion**

442 *4.1 Spatial variation of SOC stock in silvoarable systems*

The sampling protocol was designed to take account of the spatial distribution of SOC stocks as a function of distance from the trees. Sampling in the inter-rows in front of a tree or between two trees did not affect the estimation of SOC stocks. The protocol could, therefore, be simplified for instance by sampling only in front of a tree or by sampling along the diagonal of the sampling pattern, which was equivalent to a quarter of the Voronoi polygon (Levillain et al., 2011). Field sampling would then be less costly and less time-consuming.

The distance from the trees had no effect on SOC stocks in the inter-rows, except at the oldest 449 450 SJ site. At this 41-year-old site, the width of the cropped alley had been reduced over the past 10 years owing to light competition, which might explain the gradient of SOC stocks observed. 451 At the RE site, Cardinael et al., (2015b) suggested that close to the trees, organic C input coming 452 from tree fine root senescence (Cardinael et al., 2015a; Germon et al., 2016), exudates and 453 leaves might be compensated by a decrease in organic C input from crop residues owing to 454 lower yields (Dufour et al., 2013). The same hypothesis might apply at the VE site, where no 455 SOC stock gradient was found in the inter-rows (same tree density, same tree species and tree 456 age as the RE site). Consequently, fewer soil samples could be taken to estimate SOC stock in 457 the inter-rows. However, these two 18-year-old sites had a high tree density, the distance 458 between two tree rows (11 m and 13 m) being almost the same as the mean tree height (15 m 459 and 11 m). It is possible that a SOC stock gradient may appear with time in the inter-rows in 460 low-density plantations with a large distance between two tree rows (> 30 m). This gradient 461 effect could also depend on the tree species. This hypothesis could be tested in the future at the 462 463 CH and ME sites.

At all the silvoarable sites, the SOC stock was higher in the tree rows than in the inter-rows and 464 in the control plot, especially in the topsoil layer (0-10 cm). Tree rows therefore had a 465 considerable effect on SOC storage, contributing up to 50% of the additional SOC storage at 466 silvoarable plot scale for only a small surface area. There were two main sources of organic 467 matter returned to the soil in the tree rows: carbon from the trees (litter, fine roots and exudates) 468 and carbon from the herbaceous vegetation. At the RE site, the aboveground and belowground 469 biomass of the herbaceous vegetation in the tree rows was 2.13 Mg C ha⁻¹ and 0.74 Mg C ha⁻¹, 470 respectively (unpublished data). The C input to the soil from this vegetation in the tree rows 471 could, therefore, be up to 2.9 Mg C ha⁻¹ yr⁻¹. The spaces between the trees along the tree rows 472 473 could be considered comparable to grass strips or natural grassland because of the herbaceous cover and the lack of soil tillage. Converting annual crop cultivation to grassland was shown to 474 be very efficient in terms of SOC storage by Conant et al., (2001), Arrouays et al., (2002), and 475 Soussana et al., (2004) with SOC stock accumulation rates ranging from 0.49 Mg C ha⁻¹ yr⁻¹ to 476 1.01 Mg C ha⁻¹ yr⁻¹ in the top 30 cm. Based on their results and on the high SOC stocks also 477 478 measured in the topsoil in tree rows of young plantations with small tree biomass, we suggest that a major part of the SOC storage in the tree rows is due to the herbaceous vegetation. There 479 was no clear difference between sown and natural herbaceous vegetation in the tree rows, 480 481 although the highest SOC stock accumulation rate was obtained for sown grass (ME site, 1.3 Mg C ha⁻¹ yr⁻¹). However, the management of these tree rows seems to be a key factor for 482 increasing the SOC storage capacity of silvoarable systems. Several studies showed that 483 including legumes in the composition of grasslands increased herbage productivity (Tilman et 484 al., 2001; Marquard et al., 2009; Prieto et al., 2015) and SOC storage (Steinbeiss et al., 2008; 485 Lange et al., 2015). 486

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In the five silvoarable systems studied, the mean SOC stock accumulation rate in the top 30 cm 490 was 0.24 (0.09-0.46) Mg C ha⁻¹ yr⁻¹. This estimate for silvoarable plots with an average age of 491 17.8 -yr, is slightly lower than previously suggested for 20-yr-old agroforestry systems in 492 France (0.30 (0.03-0.41) Mg C ha⁻¹ yr⁻¹) by Pellerin et al. (2013) based on a literature review 493 494 but it is of the same order of magnitude. The SOC stock accumulation rate was also slightly lower than those reported by Oelbermann et al. (2006) for a 13-yr-old Canadian alley cropping 495 system combining hybrid poplars and wheat, soybean and maize grown in rotation (0.30 Mg C 496 ha⁻¹ yr⁻¹ in the top 20 cm and 0.39 Mg C ha⁻¹ yr⁻¹ in the top 40 cm). As well as, Peichl et al. 497 (2006) reported a SOC stock accumulation rate of 1.04 Mg C ha⁻¹ yr⁻¹ in the top 20 cm for a 498 499 13-yr-old hybrid poplar and Norway spruce-barley agroforestry system. Overall, our estimated 500 SOC stock accumulation rate is slightly lower than most published results (Lorenz and Lal, 2014; Kim et al., 2016). However, as reported by Cardinael et al. (2015b), our study estimated 501 502 SOC storage in silvoarable systems using the equivalent soil mass, which gives more accurate results when soil bulk density is modified by changes in land use (Ellert and Bettany, 1995; 503 Ellert et al., 2002), as was the case in these systems, especially in the tree rows. Furthermore, 504 505 most fields in our study were owned and managed by farmers. Although this fact may generate 506 some uncertainties, it has the advantage of taking account of a broad variety of practices that are commonly used by farmers. 507

At the two 18-year-old silvoarable sites (RE and VE) there was a significant increase in deep SOS stocks (below 30 cm). At the VE site this might be partially due to a slightly higher sand content in the control plot than in the agroforestry plot below 30 cm. At the RE site, this increase might result from a high density of deep tree fine roots (Mulia and Dupraz, 2006; Cardinael et al., 2015a). Although the SOC stock accumulation rate was lower than in topsoil layers, deep soil layers might then be able to store a large amount of SOC over a longer period owing to better SOC stabilization conditions (Rasse et al., 2005). However, little is known about the
effect of fresh organic matter input on deep soil layers and some authors found that this might
stimulate the mineralization of old organic matter (Fontaine et al., 2004, 2007).

There was no change in the SOC stock accumulation rates with time in the silvoarable systems 517 (Fig. 6) but very old sites (> 40 year old) were under-represented in this study. It is therefore 518 519 difficult to assess the possible effect of tree age on the SOC accumulation rate. Tree growth increases organic litter production with time but competition with the intercrop also increases, 520 potentially causing a decrease in crop yields such as cereals (Dufour et al., 2013). In a recent 521 meta-analysis, Kim et al., (2016) found a slight decrease in the SOC stock accumulation rates 522 in very old agroforestry systems, which was attributed to the soil reaching a new SOC stock 523 equilibrium. Based on technical limits (soil depth, water holding capacity, field size), Pellerin 524 et al., (2013) and Chenu et al., (2014) estimated that about 4 M ha of arable land could be 525 converted to silvoarable systems in France. Given the estimated SOC stock accumulation rate 526 in this study, this would mean that $3.6 \ 10^5$ to $1.84 \ 10^6$ Mg C could be stored annually in the soil. 527

528

529 *4.3 Carbon storage in silvopastoral systems*

The silvopastoral system set up on an andosol on permanent grassland (Tables 3 and 4) had no 530 531 more additional SOC in the top 30 cm than grassland without trees. This site had been under pasture for decades before tree planting. It had a high SOC concentration (about 65 mg C g⁻¹ at 532 0-10 cm) and the soil was possibly at a steady state so that it could not store additional SOC, at 533 least in fine soil fractions (Hassink, 1997). On a Patagonian andosol, Dube et al., (2012) also 534 535 found that there was no significant difference in the SOC stocks in the top 40 cm of a silvopastoral system compared to a natural pasture. At our site, there was a significant effect of 536 the silvopastoral system on SOC concentration and stock in the 30-50 cm layer: the SOC 537

concentration in the silvopastoral system was about 29 mg C g⁻¹ while in the grassland control 538 it was only about 23 mg C g⁻¹. It is possible that these deep soil layers in grasslands might be 539 less SOC-saturated than topsoil layers and that roots from agroforestry trees could, therefore, 540 541 contribute to additional SOC storage at depth. Haile et al. (2010) also found that trees affected deep SOC storage in silvopastoral systems. The biomass production of pastures in silvopastoral 542 systems is usually less sensitive to shade than that of annual crops such as cereals grown in 543 silvoarable systems (Moreno et al., 2007a, b; Moreno, 2008), except for N2 fixing species 544 (Carranca et al., 2015). Furthermore, grass under the tree cover can have a longer growing 545 season (Puerto et al., 1990) and forage quality can be improved under tree canopies (Cubera et 546 al., 2009). Therefore, silvopastoral systems might support a higher tree density than silvoarable 547 systems (Benavides et al., 2009; Devkota et al., 2009), resulting in higher C stocks in the tree 548 biomass (> 35 Mg C ha⁻¹ in this case). 549

550

551 *4.4 Carbon storage in the tree biomass*

The C stock in the tree biomass in the young plantations was negligible but, in the old 552 plantations, C storage was greater in the tree biomass than in the soil (Fig. 5). The C 553 accumulation rate in the tree biomass was higher in the old plantations than in young 554 plantations. This is explained by the much higher total leaf area of old trees compared to very 555 young trees and, therefore, by a higher photosynthesis capacity (Stephenson et al., 2014). 556 557 However, estimates of the tree root biomass may be underestimated by the forest allometrics used. The architecture of agroforestry trees is different from forest trees owing to a lower 558 intraspecific competition and to pruning. Moreover, agroforestry trees have been shown to be 559 560 very deep rooted owing to soil tillage and to competition with intercrops (Mulia and Dupraz, 2006; Cardinael et al., 2015a). 561

Carbon stock in the tree biomass is not usually considered as a long-term C sink in the same way as the SOC stock but the residence time of C in the harvested biomass depends on the fate of wood products and can be as long as many decades for timber wood (Profft et al., 2009; Bauhus et al., 2010), which was the case for the trees grown at the sites studied. Branches could be used as a substitute for fossil fuel to produce energy (Kürsten, 2000; Cardinael et al., 2012) or be returned to the soil as ramial chipped wood amendments (Barthès et al., 2010).

568

569 5. Conclusion

This study showed the potential of agroforestry systems to increase carbon stock in both the 570 571 soil and tree biomass under different pedo-climatic conditions in France. The sampling protocol 572 evaluated the spatial distribution of SOC stock and the results showed that it could be simplified for future studies. SOC stocks accumulated mainly in the tree rows and mainly in the top 30 cm 573 574 of soil, but at deeper soil layers in two silvoarable sites, as well. Further studies are required to 575 gain a better assessment of the effect of agroforestry on deep SOC stock. Allometric equations should be developed for trees grown in temperate agroforestry systems to reduce the uncertainty 576 of tree root biomass estimates. Very old sites (> 40 years old) were under-represented in our 577 dataset and long-term experimental agroforestry sites are required to assess the effect of trees 578 579 on soil carbon over long periods.

580

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		Soil organic carbon content		Bulk density		Soil organic carbon stock	
Site		F-value	Pr(>F)	F-value	Pr(>F)	F-value	Pr(>F)
СН	Depth	64.982	< 0.0001	10.956	0.0001	22.341	< 0.0001
	Location	2.246	0.137	3.153	0.079	1.890	0.173
	Distance	0.394	0.532	0.266	0.607	0.379	0.540
	Depth×Location	8.078	0.0006	0.672	0.513	6.908	0.002
	Depth×Distance	0.576	0.564	0.296	0.744	0.570	0.568
	Location×Distance	0.227	0.635	0.226	0.636	0.472	0.494
	Depth	140.956	< 0.0001	20.473	< 0.0001	24.004	< 0.0001
	Location	130.363	< 0.0001	78.246	< 0.0001	116.989	< 0.0001
ME	Distance	0.012	0.911	7.257	0.008	0.016	0.900
ME	Depth×Location	51.699	< 0.0001	15.888	< 0.0001	45.731	< 0.0001
	Depth×Distance	1.627	0.202	1.910	0.154	2.895	0.063
	Location×Distance	0.004	0.949	0.162	0.688	0.144	0.705
	Depth	370.623	< 0.0001	7.285	0.0104	284.905	< 0.0001
	Location	35.543	< 0.0001	0.356	0.554	33.719	< 0.0001
CI	Distance	15.183	0.0004	0.691	0.411	8.827	0.005
21	Depth×Location	6.719	0.014	6.305	0.017	9.250	0.004
	Depth×Distance	4.101	0.0501	7.985	0.008	10.264	0.002
	Location×Distance	0.987	0.327	1.534	0.223	0.728	0.399
	Depth	110.547	< 0.0001	39.920	< 0.0001	19.071	< 0.0001
	Location	24.017	< 0.0001	5.956	0.016	23.272	< 0.0001
VE	Distance	0.001	0.980	0.674	0.413	0.083	0.773
VЕ	Depth×Location	2.801	0.019	1.998	0.082	2.243	0.053
	Depth×Distance	0.086	0.994	0.917	0.472	0.151	0.980
	Location×Distance	0.278	0.599	0.095	0.758	0.075	0.785
	Depth	703.719	< 0.0001	391.32	< 0.0001	723.666	< 0.0001
RE	Location	223.367	< 0.0001	23.90	< 0.0001	66.935	< 0.0001
	Distance	2.229	0.1387	2.12	0.1491	2.353	0.1283
	Depth×Location	272.736	< 0.0001	10.04	< 0.0001	68.377	< 0.0001
	Depth×Distance	2.338	0.0173	0.68	0.7137	1.775	0.0784
	Location×Distance	4.425	0.0380	1.25	0.2666	3.285	0.0731

Table S1 ANOVA on the linear mixed-effects (LME) model for SOC content, bulk density and SOC stock in the agroforestry plots as a function

of depth, location (inter-row or tree row), distance to the closest tree, and interactions between these.

	Depth	89.206	< 0.0001	2.739	0.033	59.624	< 0.0001
TH	Location	0.040	0.842	0.577	0.449	0.032	0.859
	Distance	1.511	0.222	6.966	0.010	0.446	0.506
	Depth×Location	0.673	0.612	0.817	0.517	0.622	0.648
	Depth×Distance	0.225	0.924	0.750	0.560	0.341	0.850
	Location×Distance	0.235	0.629	1.663	0.200	0.001	0.975

803 CH: Châteaudun, ME: Melle, SJ: Saint-Jean-d'Angély, VE: Vézénobres, RE: Restinclières, TH: Theix.

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