Tillage effects on soil aggregation and soil organic carbon profile distribution under Mediterranean semi-arid conditions

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Abstract

In rainfed semi-arid agroecosystems, soil organic carbon (SOC) may increase with the adoption of alternative tillage systems (e.g. no-tillage, NT). This study evaluated the effect of two tillage systems (conventional tillage, CT vs. NT) on total SOC content, SOC concentration, water stable aggregate-size distribution and aggregate carbon concentration from 0 to 40 cm soil depth. Three tillage experiments were chosen, all located in northeast Spain and using contrasting tillage types but with different lengths of time since their establishment (20, 17, and 1-yr). In the two fields with mouldboard ploughing as CT, NT sequestered more SOC in the 0–5 cm layer compared with CT. However, despite there being no significant differences, SOC tended to accumulate under CT compared with NT in the 20–30 and 30–40 cm depths in the AG-17 field with 25–50% higher SOC content in CT compared with NT. Greater amounts of large and small macroaggregates under NT compared with CT were measured at 0–5 cm depth in AG-17 and at 5–10 cm in both AG-1 and AG-17. Differences in macroaggregate C concentration between tillage treatments were only found in the AG-17 field at the soil surface with 19.5 and 11.6 g C/kg macroaggregates in NT and CT, respectively. After 17 yr of experiment, CT with mouldboard ploughing resulted in a greater total SOC concentration and macroaggregate C concentration below 20 cm depth, but similar macroaggregate content compared with NT. This study emphasizes the need for adopting whole-soil profile approaches when studying the suitability of NT versus CT for SOC sequestration and CO₂ offsetting.

Keywords: Mediterranean conditions, soil aggregation, soil organic carbon, tillage

Introduction

Soil organic carbon (SOC) sequestration has been defined as any persistent net increase in soil organic C storage (Paustian et al., 1997; Lal, 2004). In agricultural soils, increases in SOC can lead to both improvement in plant productivity due to greater soil fertility and ancillary benefits, such as removal of CO₂ from the atmosphere (Paustian et al., 1998). The impact of agricultural management practices on C sequestration has been widely studied (Follett, 2001; Smith, 2004) and research has focused on a better comprehension of the mechanisms involved in soil organic matter (SOM) stabilization of soils (Six et al., 2002; Krull et al., 2003).

Soil organic matter dynamics are linked with soil aggregate formation and stabilization (Balesdent et al., 2000) and are strongly affected by agricultural management treatments (Six et al., 2002). Tillage not only favours SOM decomposition by aggregate breakdown but also accelerates SOM decomposition by soil microclimate change, crop residue incorporation and greater soil aeration (Peterson et al., 1998; Balesdent et al., 2000). Increased tillage intensity is related to the SOM declining (Rasmussen et al., 1998). Soil aggregation is the process by which particles of different sizes are joined and held together by different organic and inorganic materials (Amézketa, 1999). Soil aggregation is controlled by SOC, biota, ionic bridging, clay and silt content, and the presence of carbonates and gypsum (Amézketa, 1999; Bronick & Lal, 2005). Several models of soil aggregation have been proposed (Tisdall & Oades, 1982; Goss & Kay,

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The latter authors developed a conceptual model of macroaggregate formation and SOC stabilization in relation to tillage. In this model, slower aggregate turnover under no-tillage (NT) compared with conventional tillage (CT) leads to microaggregate formation within macroaggregates and SOC stabilization as a result of occlusion within these microaggregates. Consequently, the formation of soil aggregates results in the physical protection of SOM from decomposer organisms and, thus, the disruption of soil macroaggregates (>250 μm) by tillage is followed by an increase of soil CO₂ emissions (Alvaro-Fuentes et al., 2008a).

In the semi-arid agroecosystems of Spain, variable and small amounts of rainfall result in low crop yields (Cantero-Martínez et al., 2007). In these agroecosystems, traditional management practices have included intensive soil tillage with mouldboard ploughing and deep subsoiling. The adoption of reduced tillage systems could be an option to increase SOC sequestration. In the same region, Alvaro-Fuentes et al. (2009), found greater total SOC and greater formation of stable macroaggregates under NT compared with CT at the soil surface. However, this study did not measure the whole soil profile depth. The depth of sampling is a key issue when differences in SOC levels between tillage systems are being assessed (Angers & Eriksen-Hamel, 2008). However, most of the literature on SOM protection by soil aggregates has been based on studies of the first 20 cm soil depth (e.g. Beare et al., 1994; Six et al., 1999; Zibilske & Bradford, 2007). Surface sampling may lead to overestimation of SOC contents in NT when compared with CT due to SOC accumulation deeper in the profile of tilled soil (Baker et al., 2007). Thus, Balesdent et al. (2000) recommended that the C storage comparisons take into account the first 35 cm of the soil profile due to the SOM translocation to layers deeper than the deepest ploughing event, specifically for mouldboard plough treatments. As recently reported by Angers & Eriksen-Hamel (2008), SOC storage below the soil surface under CT compared with NT may have significant implications in the role of agricultural soils as sinks for atmospheric CO₂ mitigation.

The objective of this work was to study the impact of tillage on the distribution of SOC and water-stable aggregate size classes in the soil profile in three tillage experiments with contrasting CT implementation and number of years since the start of the experiment. Given the limited information currently available, our main aim was to investigate the processes of SOM stabilization by aggregates with depth in the soil. We hypothesized that: (i) the adoption of NT will increase SOC primarily in the soil surface; (ii) CT will have greater SOC at lower depths in the soil profile compared with NT; (iii) the greater SOC content in both NT at the surface and CT at depth will increase macroaggregate formation and C occlusion in the macroaggregates in these layers.

### Materials and methods

#### Experimental sites

The study was carried out in three experimental fields located at two sites along the semi-arid Ebro river valley, NE Spain. The sites were: Selvanera (SV-20) where an experimental field was established 20 yr ago to compare various tillage systems; and Agramunt with two experimental fields (AG-1 and AG-17) established 1 and 17 yr ago, respectively, also to compare tillage systems. General soil characteristics of the experimental fields are given in Table 1. In all the field experiments the cropping system was based on growing the winter wheat and barley in alternate years. In AG-1, barley (Hordeum vulgare L.) was grown in the first year. In all the fields, CT and NT were compared. Tillage systems were implemented according to the traditional systems of the area.

In SV-20, the CT treatment consisted of one pass of a subsoiler to the 40 cm depth in August followed by chisel ploughing to 15 cm depth. In AG-17 and AG-1, CT consisted of mouldboard ploughing to 30 cm depth immediately followed by one or two passes with a cultivator to 15 cm depth both in September. The NT treatment was in the same at all the three sites, consisting of a total herbicide application (1.5 L 36% glyphosate per hectare) for controlling weeds before sowing. Planting was performed with a direct drilling disk planter set to 2-4 cm depth in November. The historical

### Table 1 Site and general soil characteristics for the Ap horizon at the three experimental sites

| Site          | Agramunt (AG-1) | Agramunt (AG-17) | Selvanera (SV-20) |
|---------------|-----------------|------------------|------------------|
| Year of establishment | 2006 | 1990 | 1987 |
| Latitude      | 41°48′N | 41°48′N | 41°49′N |
| Longitude     | 1°07′E | 1°07′E | 1°17′E |
| Elevation (m) | 330   | 330   | 475   |
| Annual precipitation (mm) | 430 | 430 | 475 |
| Annual ETo (mm) | 855 | 855 | 800 |
| Soil classification | Fluventic Xerochrept | Xerofluent | Fluventic Xerochrept |
| Ap horizon depth (cm) | 25 | 23 | 28 |
| pH (H₂O, 1,2.5) | 8.4 | 8.5 | 8.3 |
| EC1,5 (dS/m) | 0.17 | 0.15 | 0.16 |
| CaCO₃ eq. (%) | 38 | 40 | 35 |
| Water retention (kg/kg) | 0.22 | 0.16 | 0.16 |
| 33 kPa | 1500 kPa | 0.07 | 0.05 | 0.04 |
| Particle size distribution (%) | 36.5 |
| Sand (2000–50 μm) | 23.9 | 30.1 | 36.5 |
| Silt (50–2 μm) | 55.0 | 51.9 | 46.4 |
| Clay (<2 μm) | 21.1 | 17.9 | 17.1 |

ETo, potential evapotranspiration. §According to the USDA classification (Soil Survey Staff, 1975).
tillage management of these three fields prior to the set up of the experiments was based on intensive tillage with two-three passes of a hoe cultivator to 30 cm depth. At all sites, tillage treatments were arranged in a randomized complete block design with three replicates in SV-20 and AG-1 and four replicates in AG-17. Plot size was (50 × 7) m at SV-20, (50 × 9) m at AG-17 and (48 × 6) m at AG-1.

Soil sampling and analyses

Soils were sampled in July 2007, directly after crop harvest. There was no rainfall during the sampling period. Soil samples were obtained using a flat spade, from five soil layers from 0 to 40 cm depth (0–5, 5–10, 10–20, 20–30 and 30–40 cm). In each plot, two areas were identified 20 m apart. In each area, a composite sample was collected from three samples randomly selected. The samples were stored in crush-resistant airtight containers and, once in the laboratory, air-dried at room temperature. At the same time, two soil cores were taken per plot and soil depth for soil dry bulk density determination (Grossman & Reinsch, 2002).

Aggregate size separation was carried out on each composite sample using a modified wet sieving method adapted from Elliott (1986). Briefly, 100 g air-dried (8 mm sieved) soil sample was placed on the top of a 2000 μm sieve and submerged for 5 min in deionized water at room temperature. Sieving was done manually, 50 times in 2 min to achieve aggregate separation. A series of three sieves (2000, 250 and 53 μm) was used to obtain four aggregate fractions: (i) large macroaggregates (2–8 mm), (ii) small macroaggregates (0.25–2 mm), (iii) microaggregates (0.05–0.25 mm) and (iv) silt-plus clay-sized particles (<0.053 mm). All aggregate fractions were oven-dried at 50 °C (24 h) in aluminium trays and weighed. Sand content of the aggregate classes (>0.053 mm) was determined by dispersing a 5 g subsample in sodium hexametaphosphate solution (5 g/L) using a reciprocal shaker. Sand correction was performed for each aggregate-size class because it was not considered to be part of the aggregates (Elliott et al., 1991). Total SOC from the bulk soil and from each aggregate size-class were determined using the wet oxidation of the Walkley–Black method described by Nelson & Sommers (1996). Aggregate size classes with a cumulative weight smaller than 5 g were not considered when determining carbon content as there was insufficient material for SOC determination.

The data were analyzed using the SAS statistical software (SAS Institute, 1990). To compare the effects of tillage treatments and soil depths within every experimental field, analysis of variance (ANOVA), for a randomized block design were performed using the procedure general linear model. When significant, differences among treatments and depths were declared at the 0.05 probability level of significance using Duncan’s test.

Results and discussion

Tillage effects on total SOC

From 0 to 40 cm depth in the soil, total SOC concentration ranged between 4.3 and 10.1, 3.6 and 12.87 and 4.5 and 13.6 g/kg at sites AG-1, AG-17 and SV-20, respectively (Table 2). Total SOC concentration was significantly greater under NT than under CT at the AG-1 site for the 0–5 and 5–10 cm depths but only in the first 0–5 cm at the AG-17 site (Table 2). However, when vertical tillage was implemented as CT (SV-20 field) a significantly larger concentration of SOC was found in the 5–10 cm depth compared with NT (Table 2). When soil bulk density was taken into account in calculating total amount of carbon, differences between tillage systems were only found at the 0–5 cm soil depth for

| Soil depth (cm) | AG-1 | AG-17 | SV-20 |
|----------------|------|-------|-------|
|                | CT   | NT    | CT    | NT    | CT   | NT    |
| 0–5            | 5.1 (0.9)*† | 10.1 (1.0) a* | 6.3 (0.4)* | 12.9 (1.4) a | 9.6 (0.2) a | 13.6 (1.9) a |
| 5–10           | 5.3 (0.6)* | 7.7 (0.8) b | 6.3 (0.5) | 7.9 (0.7) b | 8.3 (0.3)* b | 6.9 (0.1) b |
| 10–20          | 6.1 (1.1) | 5.3 (0.3) c | 5.9 (0.2) | 5.4 (0.3) c | 6.5 (0.7) c | 4.9 (0.1) b |
| 20–30          | 5.5 (0.6) | 4.7 (0.2) c | 6.1 (0.3)* | 4.3 (0.5) cd | 5.1 (0.3) d | 4.8 (0.3) b |
| 30–40          | 4.6 (0.2) | 4.3 (0.0) c | 5.7 (0.4)* | 3.6 (0.4) d | 4.5 (0.3) d | 4.8 (0.6) b |

CT, conventional tillage; NT, no-tillage; SOC, soil organic carbon. *Within each tillage system and field, different letters indicate significant differences between depths at \( P < 0.05 \). †Within each field and depth values are significantly different between tillage systems at \( P < 0.05 \).

Values in parentheses are the standard errors of the mean.

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the AG-1 and AG-17 experimental fields, and at the 5–10 cm soil depth in AG-1 (Table 3) with ca. 50% less total SOC content in the CT treatment compared with NT. Throughout the entire sampling profile (0–40 cm), no differences were found between tillage systems in any of the experimental fields (Table 3). This fact could be partly explained with the slight differences on carbon inputs between tillage treatments. On average, the production of carbon inputs for the whole experimental period was 6341 and 4919 kg/ha in the AG-1, 6637 and 6097 kg/ha in the AG-17 and 9191 and 9616 kg/ha in the SV-20, for the NT and CT treatments, respectively.

Several studies have reported SOC accumulation under NT in surface soil (Franzluebbers, 2002; Deen & Kataki, 2003; Hernanz et al., 2009). Under NT crop residues are deposited on the soil surface where a slower decomposition occurs because of drier conditions and reduced mineral nutrient availability (Kern & Johnson, 1993; Paustian et al., 1997). By contrast, at deeper depths, tillage mixes organic residues with soil (Balesdent et al., 2000; Franzluebbers, 2004).

It was notable that after 1 yr, the NT treatment at the AG-1 field showed twice as much SOC as the CT treatment at the 0–5 cm depth (Table 3). This difference between NT and CT in SOC for the 0–5 cm soil depth was the same as that

Table 3 Soil organic carbon (SOC) content under conventional tillage (CT) and no-tillage (NT) in Agramunt (AG-1 and AG-17, 1- and 17-yr-old experiment, respectively) and Selvanera (SV-20) sites

| Site   | Soil depth (cm) | Bulk density (g/cm³) | SOC (kg/ha) | Bulk density (g/cm³) | SOC (kg/ha) |
|--------|-----------------|----------------------|-------------|----------------------|-------------|
|        | CT              | NT                   |             | CT                   | NT          |
| AG-1   | 0–5             | 1.34                 | 3371*       | 1.29                 | 6499        |
|        | 5–10            | 1.33                 | 3498*       | 1.45                 | 5694        |
|        | 10–20           | 1.33                 | 8152        | 1.44                 | 7610        |
|        | 20–30           | 1.32*                | 7274        | 1.47                 | 6875        |
|        | 30–40           | 1.33                 | 6044        | 1.42                 | 6115        |
|        | 0–40            | 28 340               |             | 32 794               |             |
| AG-17  | 0–5             | 1.41                 | 4460*       | 1.31                 | 8267        |
|        | 5–10            | 1.41*                | 4417        | 1.53                 | 5986        |
|        | 10–20           | 1.38*                | 8169        | 1.53                 | 8288        |
|        | 20–30           | 1.37*                | 8409        | 1.55                 | 6655        |
|        | 30–40           | 1.48                 | 8481        | 1.49                 | 5429        |
|        | 0–40            | 33 938               |             | 34 627               |             |
| SV-20  | 0–5             | 1.20                 | 5795        | 1.16                 | 7937        |
|        | 5–10            | 1.40                 | 5780        | 1.47                 | 5069        |
|        | 10–20           | 1.54                 | 10 048      | 1.59                 | 7849        |
|        | 20–30           | 1.48                 | 7599        | 1.59                 | 7545        |
|        | 30–40           | 1.53                 | 6825        | 1.59                 | 7611        |
|        | 0–40            | 36 048               |             | 36 010               |             |

CT, conventional tillage; NT, no-tillage; SOC, soil organic carbon.
*Within each field and depth values are significantly different between tillage systems at \( P < 0.05 \).

Figure 1 Water-stable aggregate size distribution at 0–5, 5–10, 10–20, 20–30 and 30–40 cm soil depths as affected by tillage (CT, conventional tillage; NT, no-tillage) in Agramunt (AG-1, 1-yr-old experiment). Error bars represent standard errors. For the same tillage system, different lower case letters indicate significant differences between depths at \( P < 0.05 \). *Indicate significant differences between tillage systems within the same soil depth at \( P < 0.05 \).
observed in the AG-17 field, after 17 yr under NT (Table 2). The historical tillage management before the imposition of this experiment consisted of tillage system which could over years lead to an accumulation of SOC in the topsoil. This SOC on the soil surface was lost after just one pass of mouldboard ploughing in the CT treatment since no SOC redistribution at depth was reflected in the observed values (Tables 2 and 3). Similar SOC loss has been observed by other authors when ploughing a previously minimum-tilled land (Stockfisch et al., 1999).

In our study, mouldboard ploughing resulted in SOC accumulation to greater depth in CT than in NT in the AG-17 field. This accumulation balanced the smaller amount of SOC under CT near the soil surface and, as a result, SOC stocks under CT and NT in the whole studied depth (0–40 cm) were similar (Table 3). Recently, Angers & Eriksen-Hamel (2008) compiled SOC data from both full inversion tillage and NT soil profiles located in different long-term experiments. These authors observed that, when comparing with NT, soil under CT accumulated SOC below the 20 cm depth. In our study, despite individual values for a given horizon being significant different between treatments, there was a trend towards SOC accumulation in CT compared with NT for the 20–30 and 30–40 cm depths in the AG-17 field (Table 3) with 25–50% higher SOC content in CT compared with NT. However, at the other two sites (SV-20 and AG-1) there was no such trend. As stated in the Material and Methods, at the SV-20 site, primary tillage involved subsoiling to a depth of 40 cm. The lack of differences in SOC content between CT and NT below 5 cm depth in SV-20 may be attributed to the fact that this tillage implement mixed the residues more uniformly through the top 15 cm and did not bury straw at the plough sole. This fact could have a significant effect not only on the amount of crop residues incorporated into the soil but also on the edaphic conditions and related SOC mineralization resulting from tillage.

Tillage effects on aggregate-size classes

In AG-1, after 1 yr under experimentation, differences in water-stable macroaggregates between tillage treatments were found in all the soil depths except at the 0–5 cm (Figure 1). At 5–10 cm depth, more small and large macroaggregates were found in NT compared with CT. However, below 10 cm, more soil macroaggregates were found in CT compared with NT (Figure 1). Differences in total SOC concentration between tillage treatments were only measured at 0–5 and 5–10 cm depths (Table 2). Therefore, the greater amount of macroaggregates found in CT compared with NT below 10 cm depth were not accompanied by greater total SOC concentration. Also, in AG-1, the amounts of large and small macroaggregates decreased significantly with soil depth in NT but not in CT (Figure 1).
In the AG-17 field, significant differences were found between tillage systems in the large and small macroaggregate fractions at 0–5 and 5–10 cm soil depths and in the large macroaggregate fraction at 10–20 cm depth (Figure 2). At 0–5 and 5–10 cm depth, the proportion of large macroaggregates ranged from 0.03 to 0.05 g/g dry soil in CT and from 0.17 to 0.31 g/g dry soil in the NT treatment (Figure 2). In the surface soil layer under NT, more macroaggregates were found compared with CT, corresponding to greater SOC content, as found in other studies (Beare et al., 1994; Zibilske & Bradford, 2007; Álvaro-Fuentes et al., 2008b, 2009). This trend did not continue at depth, where greater total SOC found under CT compared with NT at 20–30 and 30–40 cm depths were not accompanied by greater soil macroaggregates. No differences were found between tillage systems for any fraction in the SV-20 field (Figure 3).

In the three fields, except at the soil surface at AG-17, differences in total SOC concentration between treatments were not accompanied by differences in soil macroaggregates (Table 2). Chaney & Swift (1984) suggested that total SOC may not be sufficient to explain differences in aggregate stability and that certain SOM fractions may play a more important role.

### Tillage effects on C concentration in the water-stable aggregate fractions

Not enough large macroaggregates were obtained for subsequent C concentration analyses for most of the depths and tillage treatments of the three experimental sites (Tables 4–6). At AG-1, the average sand-free C concentration of the small macroaggregates, macroaggregates and silt-plus clay-sized particles were 19.5, 8.6, 4.5 and 22.5, 10.2, 4.9 g/kg dry soil in CT and NT, respectively (Table 4). The C concentration increased with the aggregate size as found in similar studies (Elliott, 1986; Oades & Waters, 1991). Significant differences between tillage systems were only found at the 0–5 and the 5–10 cm depths for the microaggregate fraction (Table 4).

At 0–5 cm depth of AG-17, greater macroaggregate and microaggregate C concentrations were observed in NT compared with CT. However, at the 20–30 and 30–40 cm depths the opposite trend was observed, with greater

### Table 4 Soil organic carbon (SOC) and sand-free soil organic carbon (SOC sand-free) concentrations in different water-stable aggregate classes under conventional tillage (CT) and no-tillage (NT) in Agramunt (AG-1, 1-yr-old experiment, respectively) at 0–40 cm soil depth

| Soil depth (cm) | Water-stable aggregate classes (mm) | AG-1 |          |          |
|----------------|------------------------------------|------|----------|----------|
|                |                                    | SOC (g/kg) | SOC (g/kg) sand-free |
|                |                                    | CT  | NT       | CT  | NT       |
| 0–5            | >2                                 | –   | 14.9 (1.4) | –   | 21.2 (4.7) |
|                | 0.250–2                            | 9.1 (2.7)* | 14.8a (1.5) | 19.2 (3.1) | 27.7 (2.8) |
|                | 0.050–0.250                        | 3.8 (2.0) | 8.5a (0.8) | 6.3 (3.2)* | 14.5a (0.8) |
|                | <0.053                             | 4.6 (0.2) | 6.3a (0.6) | 4.6 (0.2) | 6.3a (0.6) |
| 5–10           | >2                                 | –   | 11.3 (1.1) | –   | 14.0 (1.4) |
|                | 0.250–2                            | 9.6 (3.2) | 13.3a (2.0) | 21.0 (2.7) | 23.7 (1.5) |
|                | 0.050–0.250                        | 4.7 (1.0)* | 7.6b (0.7) | 8.3 (0.8)* | 13.1a (0.1) |
|                | <0.053                             | 4.7 (0.3) | 5.6b (0.3) | 4.6 (0.3) | 5.6b (0.3) |
| 10–20          | >2                                 | 8.7 (4.6) | –          | 10.1 (5.2) | –          |
|                | 0.250–2                            | 10.2 (3.1) | 8.1b (2.2) | 21.8 (2.7) | 22.2 (1.8) |
|                | 0.050–0.250                        | 5.1 (0.8) | 4.9c (0.5) | 10.3 (0.3) | 7.9b (0.8) |
|                | <0.053                             | 4.7 (0.2) | 4.6c (0.2) | 4.7 (0.2) | 4.6c (0.2) |
| 20–30          | >2                                 | 6.6 (3.4) | –          | 7.8 (3.9) | –          |
|                | 0.250–2                            | 10.2 (2.2) | 7.9b (2.4) | 19.9 (1.4) | 20.8 (1.0) |
|                | 0.050–0.250                        | 5.2 (0.3) | 4.7c (0.4) | 9.5 (0.6) | 8.1b (0.1) |
|                | <0.053                             | 4.5 (0.1) | 4.2cd (0.3) | 4.5 (0.1) | 4.2cd (0.3) |
| 30–40          | >2                                 | –   | –          | –   | –          |
|                | 0.250–2                            | 7.4 (1.1) | 7.0b (1.3) | 15.8 (0.8) | 18.1 (2.3) |
|                | 0.050–0.250                        | 4.6 (0.7) | 4.6c (0.3) | 8.5 (1.7) | 7.4b (0.5) |
|                | <0.053                             | 4.2 (0.3) | 4.0d (0.4) | 4.2 (0.3) | 4.0d (0.4) |

CT, conventional tillage; NT, no-tillage; SOC, soil organic carbon. *Within each tillage system and water-stable aggregate fraction, different letters indicate significant differences between depths at $P < 0.05$. **Within each depth and water-stable aggregate fraction values are significantly different between tillage systems at $P < 0.05$. †Values in parentheses are the standard errors of the mean.
aggregate C concentration in CT compared with NT (Table 5) similar to the trend found in the total SOC (Table 2).

At AG-17, sand correction affected greatly the differences in the C concentration for the small macroaggregate fraction between tillage systems. For instance, in the small macroaggregates in the 0–5 cm soil layer, aggregate C concentration without correcting for sand content was significantly greater in NT compared with CT. However, when corrected, C concentration in small macroaggregates was similar in both tillage treatments (Table 5). In our experiment, the use of inversion tillage (i.e. mouldboard ploughing) for several years led to either the redistribution of the sand particles within the soil profile or the erosion of the clay and silt particles from the soil surface resulting in a greater sand proportion in CT compared with NT. Caravaca et al. (2004) pointed out that the intensively tilled soils of semi-arid Mediterranean areas are likely to become degraded by losing their organo-mineral fine particle-size fractions. In the SV-20 field, a significantly greater C concentration was found in aggregates from the 10–20 cm where higher SOC was found in the small macroaggregates, microaggregates and sand-plus clay-sized particles (Table 6). The CT treatment with subsoil tillage left enough sample weight to determine the C concentration of the large microaggregates in both NT and CT but only for the 0–5 and 5–10 cm soil depths (Table 6). The greater C concentration in the small macroaggregate fraction found at the soil surface under the NT treatment compared with CT (i.e. AG-1 and AG-17) was in agreement with other similar studies (Beare et al., 1994; Zibilske & Bradford, 2007; Álvaro-Fuentes et al., 2008b). In Mediterranean Spain, aggregate stability has been shown to increase with the reduction of tillage intensity in the soil surface (Hernanz et al., 2002; Álvaro-Fuentes et al., 2008b), likely due to greater SOC stabilization of microaggregates within macroaggregates (Álvaro-Fuentes et al., 2009). Six et al. (1998, 1999) proposed a model in which slower macroaggregate turnover rate under NT was the main mechanism for C sequestration. Consequently, SOC accumulated in the top 5 cm was occluded by soil macroaggregates in NT due to lower aggregate turnover rates. However, in the AG-17 field with mouldboard

### Table 5 Soil organic carbon (SOC) and sand-free soil organic carbon (SOC sand-free) concentrations in different water-stable aggregate classes under conventional tillage (CT) and no-tillage (NT) in Agramunt (AG-17, 17-yr-old experiment, respectively) at 0–40 cm soil depth

| Soil depth (cm) | Water-stable aggregate classes (mm) | SOC (g/kg) | SOC (g/kg) sand-free |
|----------------|------------------------------------|------------|----------------------|
|                |                                    | CT         | NT                   |
| 0–5            | >2                                 | 12.1 (4.0) | –                    |
|                | 0.250–2                            | 11.6 (1.2)*| 19.5 (2.4)*          |
|                | 0.050–0.250                        | 5.4a (0.3)*| 9.7a (0.9)           |
|                | <0.053                             | 5.1 (0.2)* | 7.4a (0.8)           |
| 5–10           | >2                                 | 10.3 (0.4) | –                    |
|                | 0.250–2                            | 12.9 (1.4) | 14.3b (1.3)          |
|                | 0.050–0.250                        | 5.5a (0.6) | 6.7b (0.5)           |
|                | <0.053                             | 5.0 (0.1)  | 5.7b (0.4)           |
| 10–20          | >2                                 | 11.0 (0.1) | –                    |
|                | 0.250–2                            | 13.1 (1.2) | 10.1c (0.6)          |
|                | 0.050–0.250                        | 5.3a (0.6) | 4.6c (0.2)           |
|                | <0.053                             | 5.1 (0.2)  | 4.5c (0.4)           |
| 20–30          | >2                                 | –          | –                    |
|                | 0.250–2                            | 13.3 (1.5)*| 6.8d (1.5)           |
|                | 0.050–0.250                        | 5.6a (0.4) | 3.9cd (0.6)          |
|                | <0.053                             | 4.8 (0.3)  | 4.2cd (0.6)          |
| 30–40          | >2                                 | –          | –                    |
|                | 0.250–2                            | 11.4 (1.3)*| 5.2d (1.1)           |
|                | 0.050–0.250                        | 4.6b (0.3)*| 3.2d (0.3)           |
|                | <0.053                             | 4.9 (0.4)  | 3.7d (0.7)           |

CT, conventional tillage; NT, no-tillage; SOC, soil organic carbon. *Within each tillage system and water-stable aggregate fraction, different letters indicate significant differences between depths at $P < 0.05$. †Within each depth and water-stable aggregate fraction values are significantly different between tillage systems at $P < 0.05$. Values in parentheses are the standard errors of the mean.
ploughing after 17 yr, greater SOC concentration was found in the small macroaggregates under CT below the first 20 cm depth (Table 5). As mentioned before, below 20 cm depth, 25–50% higher total SOC content was observed in CT compared with NT at AG-17 (Table 3). When mixing crop residues at depth (full inversion tillage case), residue C is located closely in contact with mineral soil particles (Angers & Eriksen-Hamel, 2008). In a laboratory study, Stemmer et al. (1999) compared the distribution of corn-derived organic matter between different residue locations. The authors concluded that organic matter is more protected within inorganic compounds when mixed into the soil than when left over the soil surface. As a result, they observed greater fraction of SOC moved into the stable SOC pool in tilled than in untilled soil. Therefore, the silt-plus clay-sized particles may play an important role on the protection of SOC, especially in deeper horizons (Poirier et al., 2009). Moreover, more adverse microenvironmental conditions for the decomposers in the plow layer in the CT treatment may restrict the mineralization of SOC (Poirier et al., 2009). However, macroaggregate content was similar between tillage treatments below 20 cm depth. Slower decomposition of crop residues incorporated by tillage to depth (Olchin et al., 2008) could lead to the build up of soil aggregates by fresh C incorporated in the CT treatment, resulting in greater macroaggregate C concentration, but without differences in macroaggregate fraction contents between treatments. As suggested recently by Angers & Eriksen-Hamel (2008), mechanisms controlling C stabilization deep in the soil profile require further investigation.

### Conclusions

In the rainfed semi-arid agroecosystems studied, we observed under NT an increase in SOC content only in the surface layer, whereas we observed an increase in SOC concentration below 20 cm depth under the CT mouldboard treatment, when this tillage system had been performed for 17 yr. However, at the 0–40 cm soil depth, NT compared with CT did not show any differences in total SOC content. After 17 yr under NT, the highest SOC in the 0–5 cm depth under NT was accompanied by both higher macroaggregate content and higher aggregate C concentration. But after only 1 yr under NT, in the 0–5 cm depth the greater SOC content was not accompanied either by differences in macroaggregate content or macroaggregate C concentration. Mouldboard

### Table 6

Soil organic carbon (SOC) and sand-free soil organic carbon (SOC sand-free) concentrations in different water-stable aggregate classes under conventional tillage (CT) and no-tillage (NT) in Selvanera (SV-20, 20-yr-old experiment, respectively) at 0–40 cm soil depth

| Soil depth (cm) | Water-stable aggregate classes (mm) | SOC (g/kg) | SOC (g/kg) sand-free |
|----------------|------------------------------------|------------|---------------------|
|                | CT       | NT          | CT       | NT          |
| 0–5            | >2       | 14.0 (1.8)† | 19.0 (1.9) | 27.1 (8.2) | 30.7 (7.9) |
|                | 0.250–2  | 15.5a (0.6) | 20.0a (2.2) | 34.0 (4.4) | 33.8a (4.5) |
|                | 0.050–0.250 | 9.6a (0.5) | 11.3a (1.3) | 20.0a (1.1) | 24.5a (3.5) |
|                | <0.053   | 8.4a (0.6) | 10.1a (1.3) | 8.4a (0.6) | 10.1a (1.3) |
| 5–10           | >2       | 7.0 (3.5) | 13.5 (0.8) | 28.4 (16) | 22.6 (2.9) |
|                | 0.250–2  | 14.6a (4.9) | 12.3b (1.8) | 21.5 (11) | 26.1b (2.8) |
|                | 0.050–0.250 | 8.5ab (0.4) | 7.4b (0.4) | 17.8a (0.6) | 17.6b (1.3) |
|                | <0.053   | 7.7a (0.3) | 6.8a (0.3) | 7.7a (0.3) | 6.8b (0.3) |
| 10–20          | >2       | –          | –         | –         | –         |
|                | 0.250–2  | 10.5b (0.4)* | 5.8a (0.5) | 26.5 (2.5)* | 17.4c (0.4) |
|                | 0.050–0.250 | 7.0bc (0.3)* | 4.6c (0.3) | 13.6b (0.9)* | 10.0c (0.2) |
|                | <0.053   | 7.7a (0.5)* | 5.7a (0.0) | 7.7b (0.5)* | 5.7b (0.0) |
| 20–30          | >2       | –          | 4.2 (2.8) | –         | 16.4 (9.4) |
|                | 0.250–2  | 6.3c (0.6) | 4.4c (0.6) | 23.6 (3.4) | 13.2c (0.8) |
|                | 0.050–0.250 | 5.1d (0.4) | 4.2c (0.1) | 11.2bc (0.7) | 9.3c (0.3) |
|                | <0.053   | 6.2b (0.7) | 5.2a (0.4) | 6.2b (0.7) | 5.2b (0.4) |
| 30–40          | >2       | –          | –         | 9.2 (9.2) | 11.9 (12) |
|                | 0.250–2  | 5.6c (1.3) | 4.3c (0.4) | 11.4 (1.1) | 13.0c (1.5) |
|                | 0.050–0.250 | 5.5cd (0.8) | 4.4c (0.3) | 10.0c (0.7) | 8.8c (1.0) |
|                | <0.053   | 6.0b (0.5) | 4.8b (0.6) | 6.0b (0.5) | 4.8b (0.6) |

CT, conventional tillage; NT, no-tillage; SOC, soil organic carbon. *Within each tillage system and water-stable aggregate fraction, different letters indicate significant differences between depths at \( P < 0.05 \). †Within each depth and water-stable aggregate fraction values are significantly different between tillage systems at \( P < 0.05 \). ‡Values in parentheses are the standard errors of the mean.
ploughing resulted in greater total SOC concentration and macroaggregate C concentration below 20 cm than NT, but vertical tillage did not. The generally accepted notion that tillage reduction is the most beneficial practice for SOC sequestration must be considered with caution. This study emphasizes the complex relationship among tillage, soil aggregation and SOC stabilization in soil depth and so it will require further investigation to clarify the relative importance of these mechanisms.

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