

## **Changes in water stable aggregate and soil carbon accumulation in a no-tillage with weed mulch management site after conversion from conventional management practices**

Miwa Arai <sup>a</sup>, Yukio Minamiya <sup>a</sup>, Hiroshi Tsuzura <sup>a</sup>, Yoshinori Watanabe <sup>b</sup>, Atsushi Yagioka <sup>c</sup>, Nobuhiro Kaneko <sup>a</sup>, 2014. Changes in water stable aggregate and soil carbon accumulation in a no-tillage with weed mulch management site after conversion from conventional management practices. *Geoderma* 221–222C, 50–60.

<sup>a</sup>Soil Ecology Research Group, Yokohama National University, 79-7 Tokiwadai, Hodogaya, Yokohama 240-8501, Japan

<sup>b</sup> Kinki University, 3-4-1Kowakae, Higashiosaka City, Osaka 577-8502, Japan <sup>c</sup>

<sup>c</sup> College of Agriculture, Ibaraki University, 3-21-1, Chuuo, Ami, Inashiki, Ibaraki, 300-0393, Japan

## Highlight

- > We investigated at a site of no-tillage with weed mulch management (NWM).
- > Weed biomass, water stable aggregate (> 2 mm) and soil C increased with adopting NWM.
- > NWM increased soil C ( $60 \text{ g C m}^{-2} \text{ yr}^{-1}$ ) for 17 years, thus it is an effective method to sequester soil C.
- > C input as slashed weeds and litter were small ( $265\text{-}2317 \text{ g C m}^{-2} \text{ yr}^{-1}$ ) and corresponded to 2- 11 % of C.

Keywords: weed, root, earthworm, chronosequence method, cropland

## Abstract

Soil carbon (C) is crucial for maintaining soil functions, and it increases after conversion of an agricultural field from conventional tillage management to no-tillage management due to decreasing human-induced soil disturbance and the modification of soil structure through ecosystem engineers such as earthworms. To improve soils and prevent degradation, understanding the effects of no-tillage management over time in changing water stable aggregates (WSA) and soil C is important. We investigated the changes in WSA and soil C at a site in Akame, Mie, Japan, operating a no-tillage with weed mulch management (NWM) system over a chronosequence from 0 to 17 years after conversion from conventional tillage practices (NWM for 0, 5, 10, 15, and 17 years). We measured weed aboveground biomass, litter accumulation, and root and earthworm density and biomass, and analyzed the WSA and C of bulk soil and each WSA size fraction. Weed aboveground biomass increased with site age, while litter accumulation, root biomass (soil depth of 0–4 cm), and earthworm density and biomass did not appear to be related to site age. Endogeic earthworm density and biomass tended to increase at year 5 of NWM compared to year 0. The WSA  $\geq 2 \text{ mm}$  and soil C stock in WSA of  $\geq 2 \text{ mm}$

mm increased over time under NWM at a soil depth of 0–15 cm, while the soil C stock of 0.25–1-mm WSA decreased at soil depths of 0–5 cm. The total soil C accumulation rate was  $60 \text{ g C m}^{-2} \text{ yr}^{-1}$  at a soil depth of 0–25 cm over the NWM chronosequence. Therefore, our results indicated that by adopting NWM, C inputs to the soil from weed aboveground biomass, as well as increases in the WSA of  $\geq 2$  mm, might be responsible for soil C sequestration

## 1. Introduction

Soil organic matter (SOM) is important for the maintenance of soil functions, such as nutrient cycling, infiltration, and soil structure (Carter, 2002); however, carbon dioxide emissions from agricultural land have increased since the intensification of agriculture (Lal, 2004), and soil degradation has become a serious problem due to the decrease in soil carbon (C) (Bot and Benites, 2005; Lal, 2009). Soil C is decreasing rapidly due to anthropogenic soil disturbance from tillage and seeding (Luo et al., 2010). Therefore, increasing and maintaining the soil C content is necessary to ensure the sustainable use of croplands. Recently, conservation management practices, including no-tillage, mulching, cover crops, and fertilizer and manure application have attracted attention, as these management practices are known to increase levels of soil C and water stable aggregates (WSA) (Aoyama et al., 1999; Edmeades, 2003; Ludwig et al., 2011).

Soil aggregates are secondary particles bound to mineral particles with organic and inorganic substances (Bronick and Lal, 2005; Kemper and Rosenau, 1986). Soil aggregate formation is influenced by agricultural management, and a relationship exists between soil aggregate stability and certain soil functions, including nutrient supply and retention, and water permeability and retention (Cater, 2002; Holland, 2004). Soil aggregates are formed by the activities of soil animals, roots, and microorganisms, and exhibit varying degrees of water stability, as well as serving as indicators of the soil's structural stability. Tillage practices destroy soil aggregates, but WSA ( $>2$  mm) increase when no-tillage management is adopted (Pinheiro et al., 2004; Shi et al., 2010). Pinheiro et al. (2004)

studied the increases in WSA (>2 mm) and soil C under no-tillage management compared to conventional management. Shi et al. (2010) revealed that WSA (2–10 mm) decreased under tillage management compared to grassland due to repeated tillage. The WSA size distribution can be used to estimate the effects of land use changes on soil structure (Pinheiro et al., 2004). Agricultural management has been shown to alter the stability of macroaggregates (>0.25 mm) that affect soil C dynamics (Six et al., 2000). The maintenance of soil aggregate structure prevents the decomposition of soil C and thus contributes to an increase in soil C.

Changes in soil structure, as well as increases in soil C content and ecosystem engineers, in terms of soil animal abundance and biomass, occur after the conversion from conventional management to no-till management (Kladivko et al., 1997). “Natural farming” (Fukuoka, 1987) is a unique method based on four major principles: no cultivation, no fertilizer, no weeding, and no pesticides. This type of management is different from no-tillage, as it uses weeds for mulching after slashing the aboveground weeds and retains the roots intact in the soil. Luo et al. (2010) reported that the adoption of stubble retention or incorporation significantly increased soil C content for only 25 years. Herbicides have been used worldwide to restrict the growth of weeds under no-tillage management systems (Soane et al., 2012; Stockmann et al., 2013), but weed populations and density increase under no-tillage management compared with conventional management (Cardina et al., 1991; Spandle et al., 1999). Some studies on natural farming (i.e., no-tillage and weed mulch management: NWM) have shown that decreasing human-induced disturbance changed weed biomass and soil organism density and biomass (Miura et al., 2010; Arai et al., 2013). Additionally, changes in weed above- and belowground biomass in agricultural fields are known to influence soil C accumulation. Previous studies have found that soil C under fallow conditions increased or decreased due to variations in the weed above- and belowground input compared to conventional management (Luo et al., 2010; Shimoda and Koga, 2013). Therefore, studies to determine the effects of weed management without external inputs over time on soil C accumulation are necessary. NWM systems are beneficial for earthworms because the level of disturbance decreases and more food is provided as weed mulch than

in conventional management systems. Many studies have indicated that earthworm density and biomass change under different agricultural practices (Chan, 2001; Fonte et al., 2009; Kladivko, 1997). However, few studies have investigated the effects of weeds on earthworm density and biomass under NWM.

NWM sites are usually covered by plant residue at the soil surface, and this soil condition is preferred by earthworms because it contributes to the formation of soil aggregates with fresh C inputs (Arai et al., 2013). Furthermore, above- and belowground biomass increase residue input and root activity, which enhances the formation of soil aggregates and the accumulation of soil C (Arai et al., 2013; Fonte et al., 2012; Luo et al., 2010; Six et al., 2004). The half-life of earthworm casts ranged from 2 to 11 months in pastures (trampled and protected, respectively; Decaëns, 2000); therefore, earthworm casts may affect the soil environment regardless of temporal changes in earthworm activity. Under NWM, the potential exists for an increase in biological factors involved in soil aggregate formation that may contribute to the accumulation of soil C compared to conventional management. Studies on soil aggregates in croplands under different management practices have been reported (Fonte et al., 2009; Riley et al., 2008), but no studies on the changes in soil aggregates, soil C, and biological factors over the NWM chronosequence have been reported. Earthworm biomass increased under no-tillage management in Japan, but earthworm recovery in agricultural fields was not easy after conversion from conventional management to no-tillage management (Arai et al., 2013; Miura et al., 2010). Therefore, studies are needed to determine the earthworm density and biomass under NWM.

Changes in soil aggregate formation, mediated by biological factors after conversion from conventional management to NWM, may contribute to improvements in soil C accumulation without external inputs. In this study, we focused on weed aboveground biomass, litter accumulation, and root biomass, as well as the density and biomass of earthworms following the adoption of NWM. We investigated 10 plots under NWM over a chronosequence from 0 to 17 years. We examined earthworm density, biomass, and species, weed aboveground biomass, litter accumulation, and root biomass; WSA size distribution and soil C at each WSA size; and changes in soil C abundance over

the NWM chronosequence. We hypothesized that continues NWM practice would increase above- and belowground weed biomass and then WSA size distribution and soil C accumulation would be influenced.

## **2. Materials and methods**

### *2.1. Study site*

The study site was a terraced field with an area of 2.7 ha located in Akame, Mie, Japan (34°35'N, 136°03'E). Monthly means of precipitation and daily temperature ranged from 38.5 to 273.1 mm and 1.9°C to 24.4°C, respectively. Annual averages in precipitation and temperature were 1581.4 mm and 15.9°C, respectively, over the last 30 years (1981–2010). The soils were identified as gravelly yellow soils with mottling, which are classified as Alisols derived from sandy granite according to FAO/UNESCO. The soil physical and chemical parameters, such as pH (H<sub>2</sub>O) that was determined by a 1:2.5 soil: H<sub>2</sub>O suspension and soil texture, are shown in Table 1, and levels of exchangeable base cations (Ca, Mg, and K) were determined by Watanabe et al. (in preparation).

### *2.2. Land management and use*

#### *2.2.1. Natural farming (NWM)*

The study sites are managed by Mr. Yoshikazu Kawaguchi, an expert on natural farming. Natural farming, which includes NWM, was first introduced by Mr. Masanobu Fukuoka (Fukuoka, 1978) and later modified by several other farmers. Natural farming is a simple agricultural practice that involves minimal fertilizer application and weeding and relies on weed or mowed weed cover to prevent the exposure of a bare soil surface. The characteristics of the natural farming site in this study are as follows:

- No-tillage.
- No chemical fertilizer, only one application of rice bran or oil cake at the time of planting.

- No pesticide or herbicide.
- Weed control by mowing, with weeds used as living mulch.

We investigated the sites over a chronosequence of conversion from conventional cropping to natural farming (site age: 5, 10, 15, and 17 years), and sampling plots (0.9 m × 1.8 m) with two replications. Following conventional management, a site left fallow for a year was also investigated, and the site age was set as year 0. These sites had been paddy fields under conventional management prior to the commencement of natural farming management. For details on the management of conventional farming before the fallow period and NWM system was adopted, refer to the next section. A variety of crops were grown with minimum weed control. The crops harvested in the summer were *Zea mays*, *Capsicum annuum* var. *angulosum*, *Zingiber officinale*, *Abelmoschus esculentus*, and *Solanum lycopersicum*, while those harvested in the winter included *Lactuca sativa*, *Allium fistulosum*, *Allium sativum*, *Arctium lappa*, *Brassica campestris*, and *Raphanus sativus* var. *longipinnatus* (Fabaceae).

### 2.2.2. *Conventional farming before no-tillage with weed management and fallow*

The conventional farming had been a paddy field for more than 30 years and was conventionally managed after its conversion from a paddy field. No accurate information was available regarding fertilizer applications and tillage management; however these management sites were believed to have received conventional applications of chemical fertilizer at each cropping (10–60 kg N ha<sup>-1</sup>, 15–35 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, 15–55 kg K<sub>2</sub>O ha<sup>-1</sup>; Standard Reference of Mie Prefecture) and herbicide applications as required. Tillage management and mixing of soil and plant debris from crops and weeds had been conducted.

### 2.3. *Above- and belowground biological activities*

Weed and litter sampling was undertaken from May 19 to 22, 2011. The weed species collected are shown in Table 2. We measured weed aboveground biomass, litter accumulation, and root biomass, as

well as the total C content of weeds above ground and the litter, but we did not measure the total C content of root samples because only small amounts of root samples were collected. We mowed the aboveground weeds (50 cm × 50 cm) and collected litter from the soil surface. Then, soil blocks (400 ml, soil depth: 0–4 cm) were collected for the measurement of root biomass at each site. Weeds above ground, litter, and root samples were washed with tap water to remove soil, then dried in an oven at 60°C. After 72 h, the samples were removed and weighed. Aboveground weed and litter were grained for the analysis of total C content with an organic elemental analyzer (Macro Coder JM1000CN, J-Science Lab Co., Ltd., Minami-ku, Japan). Earthworms were collected by hand-sorting at soil depths of 0–20 cm (50 cm × 50 cm) and by using mustard earthworm repellent at soil depths below 20 cm (ISO, 2006; Muramoto and Werner, 2002). Earthworm species were identified, and wet weights were recorded. Epigeic earthworm species in Japan have 1-year life cycles. *Pheretima hilgendorfi* (Michaelsen, 1892) are juveniles until June, and only adults can be found in July and August (Uchida and Kaneko, 2004). Earthworms collected in the field survey in May 2011 were difficult to identify, and therefore, we collected earthworms again at the same plot on July 30 and 31, 2012, using the same method as in 2011.

#### 2.4. Bulk density, soil carbon, and WSA analysis

We collected soil samples at depths of 0–5, 5–15, and 15–25 cm for the analysis of bulk density, soil C content, and WSA using a 100-ml core sampler over the chronosequence of the NWM (four samples per site). Soil samples were transferred to plastic bags and weighed prior to the analysis of WSA and bulk density. Soil samples used for the analysis of WSA (80 g) were dried and preserved in plastic bags. The remaining soil samples were oven-dried at 105°C and weighed. Roots and debris were removed and the soil was grained and stored in plastic bags until the soil C concentration could be determined. Density measurements were adjusted for roots, debris, and stones. A modified wet-sieving procedure was used to determine the aggregate size distribution and the aggregate stability index (Mikha and Rice, 2004; Yoder, 1936). Dried soil (80 g) was separated using an instrument



similar in principle to the Yoder wet sieving apparatus (DIK-2010, Daiki Co., Saitama, Japan). The apparatus was modified and designed to handle stacked sieves (15 cm diameter × 45 mm height) and to allow for the complete recovery of all particle fractions from individual samples. Four aggregate size classes and soil particle fraction were collected from each site (n = 4 per site): >2, 1–2, 0.25–1, 0.053–0.25, <0.053 mm diameter. Macroaggregates were defined as the >2, 1–2, and 0.25–1-mm fractions, and microaggregates were defined as the 0.053–0.25 mm size fraction. Sieves with mesh openings of >0.053 mm diameter were contained in the oscillation cylinders. Soils were evenly distributed over the nested sieve surfaces. The nested sieve was set at the highest point when the oscillation cylinders were filled with distilled water to a level whereby the bottom sieve (0.053 mm) was completely covered with water without reaching the top screen (2 mm). The soils were submerged, and organic materials and stones were removed in water for 10 min before the start of the wet-sieving action. Samples were then placed on the top sieve of each nest. Distilled water was rapidly added to each cylinder until the soil sample and top screen were covered with water. The apparatus specifications of oscillation time (10 min), stroke length (3.8 cm), and frequency (30 cycles min<sup>-1</sup>) were held constant. Fine floating particles (<0.053 mm) in the distilled water were collected by filtration. Material remaining on each sieve was backwashed into a petri dish and dried at 105°C for 24 h. The dried aggregate from each size class was weighed and stored in a crush-resistant container at room temperature until the soil C concentration was determined for each WSA size fraction. The total C concentration in soils samples was analyzed using an organic elemental analyzer (Macro Coder JM1000CN, J-Science Lab Co., Ltd.). We calculated the soil C stock (g kg soil<sup>-1</sup>) by multiplying each WSA size distribution and the soil C concentration of each WSA size fraction.

The mean weight diameter (MWD) of soil aggregates (Kemper and Rosenau, 1986) was determined as

$$MWD = \sum_{i=1}^n x_i w_i$$

where  $x_i$  is the mean diameter of each class (mm) and  $w_i$  is the weight fraction in each aggregate class.

## 2.5. Statistical analysis

We assessed the effects of site age (number of years since adopting NWM) on weed aboveground biomass, litter accumulation, root biomass, and earthworm density and abundance using a generalized linear model (GLM). We avoided a potential pseudoreplication problem by analyzing variation in the dependent variables of bulk soil C ( $\text{g C kg}^{-1}$ ), WSA of each size fraction ( $\text{g kg}^{-1}$  soil), soil C stock in WSA ( $\text{g C kg}^{-1}$ ), soil C accumulation ( $\text{g C m}^{-2}$ ), and MWD (mm) using linear mixed-effects models (nlme function in R). Site age (number of years under NWM) was treated as a fixed continuous explanatory variable, while each soil core and each size of WSA per sample ( $n = 4$ ) within each site ( $n = 2$ ) were treated as random effects. All analyses were performed using the R software environment version 2.15.1 for statistical computing (R Development Core Team, 2012).

## 3. Results

### 3.1. Changes in biological factors

Weed aboveground biomass increased over the NWM chronosequence (Fig. 1a;  $P < 0.05$ ). In contrast, litter accumulation and root biomass (soil depth: 0–4 cm) did not differ significantly with site age (Fig. 1b;  $P > 0.05$ ).

Earthworm density and biomass remained more or less constant with site age in 2011 and 2012 (Fig. 2; Table 3a and b). Identifying the earthworm species was difficult because of the presence of juveniles of both Megascolecidae spp. and Lumbricidae spp. in 2011 and 2012. The dominant earthworm species after 5 years of NWM in 2011 were the anecic species *Metaphire megascolidioides* (Goto and Hatai, 1899) and the endogeic species *Eisenia japonica* (Michaelsen, 1892). *E. japonica* was also dominant after 5 years of NWN in 2012. The epigeic species *Metaphire agrestis* and

*Amyntas irregularis* were found in 2012. No significant relationships were observed between earthworm density and biomass and litter accumulation, respectively ( $P > 0.05$ ).

The total C contained in aboveground weeds increased over the NWM chronosequence ( $P < 0.01$ ) and ranged from 62 to 184 g C m<sup>-2</sup> at each site over the 0–17 years. The total C content of the litter did not change with year ( $P > 0.05$ ) and was in the range of 0–266 g C m<sup>-2</sup> at each site over the 0–17 years.

### 3.2. Size distribution of WSA and MWD

For WSA at a soil depth of 0–5 cm, the amount of aggregates >2 mm increased, while the amount of aggregates in the 0.25–1-mm size fraction decreased over the NWM chronosequence (Fig. 3;  $F_{1,8} = 7.24$ ,  $P < 0.05$ ;  $F_{1,8} = 8.85$ ,  $P < 0.05$ ). The aggregate size fractions of 1–2 mm, 0.053–0.25 mm, and <0.053 mm did not differ significantly with site age (soil depth: 0–5 cm) ( $F_{1,8} = 0.83$ ,  $P = 0.389$ ;  $F_{1,8} = 0.156$ ,  $P = 0.156$ ;  $F_{1,8} = 2.67$ ,  $P = 0.141$ ). The fraction of WSA >2 mm tended to increase 5 years after the adoption of NWM (soil depth: 0–5 cm). At a soil depth of 5–15 cm, the proportions of the soil aggregates >2 mm increased, and the 0.053–0.25-mm and <0.053-mm size fractions decreased with site age ( $F_{1,8} = 7.13$ ,  $P < 0.05$ ;  $F_{1,8} = 6.94$ ,  $P < 0.05$ ;  $F_{1,8} = 7.08$ ,  $P < 0.05$ ). The WSA size fractions of 1–2 mm and 0.25–1 mm did not change significantly following the adoption of NWM (soil depth: 5–15 cm) ( $F_{1,8} = 0.239$ ,  $P = 0.638$ ;  $F_{1,8} = 4.09$ ,  $P = 0.078$ ). For WSA at a soil depth of 15–25 cm, all aggregate sizes (>2, 1–2, 0.25–1, and 0.053–0.25 mm) and the <0.053-mm fraction did not change over the NWM chronosequence ( $F_{1,8} = 2.11$ ,  $P = 0.185$ ;  $F_{1,8} = 3.09$ ,  $P = 0.117$ ;  $F_{1,8} = 0.841$ ,  $P = 0.386$ ;  $F_{1,8} = 2.489$ ,  $P = 0.153$ ;  $F_{1,8} = 0.0238$ ,  $P = 0.8811$ ). In samples of plant debris containing roots and organic matter, the amount of WSA at 15–25 cm increased with site age ( $F_{1,8} = 5.97$ ,  $P < 0.05$ ), whereas at soil depths of 0–5 cm and 5–15 cm, levels of WSA did not change with NWM.

The MWD at soil depths of 0–5, 5–15, 15–25 cm were determined using a linear mixed model. MWD increased with time after the adoption of NWM at soil depths of 0–5 and 5–15 cm (Table 2), but not at a soil depth of 15–25 cm (Table 4).

### 3.3. Soil carbon stock in the WSA size class

At a soil depth of 0–5 cm, the soil C stock in WSA >2 mm increased, while the soil C stock in the WSA 0.25–1-mm size fraction decreased, with site age ( $F_{1,8} = 9.99$ ,  $P < 0.05$ ;  $F_{1,8} = 6.87$ ,  $P < 0.05$ ). The soil C stock in the WSA 1–2, 0.053–0.25, and <0.053-mm size fractions did not change with year (soil depth: 0–5 cm) ( $F_{1,8} = 2.63$ ,  $P = 0.144$ ;  $F_{1,8} = 0.0190$ ,  $P = 0.894$ ;  $F_{1,8} = 2.112$ ,  $P = 0.1836$ ). At a depth of 5–15 cm, the soil C stock in WSA >2 mm increased significantly, while in the WSA of all other sizes (1–2, 0.25–1, 0.053–0.25 mm) and <0.053-mm size fraction, the soil C stock did not change over the NWM chronosequence ( $F_{1,8} = 10.8$ ,  $P < 0.05$ ;  $F_{1,8} = 4.99$ ,  $P = 0.0559$ ;  $F_{1,8} = 1.06$ ,  $P = 0.333$ ;  $F_{1,8} = 2.018$ ,  $P = 0.193$ ;  $F_{1,8} = 2.018$ ,  $P = 0.193$ ;  $F_{1,8} = 4.91$ ,  $P = 0.0576$ ). At a soil depth of 15–25 cm, the soil C of all WSA size fractions did not change with NWM ( $F_{1,8} = 2.03$ ,  $P = 0.193$ ;  $F_{1,8} = 4.29$ ,  $P = 0.072$ ;  $F_{1,8} = 0.080$ ,  $P = 0.785$ ;  $F_{1,8} = 0.069$ ,  $P = 0.800$ ;  $F_{1,8} = 0.707$ ,  $P = 0.425$ ).

### 3.4. Soil C accumulation

Bulk density was not influenced at a soil depth of 0–5 and 5–15 cm, but decreased at a soil depth of 15–25 cm in the years after the adoption of NWM. Bulk soil C concentration increased at 0–5 and 15–25 cm, and did not change at a soil depth of 5–15 cm over time after adopting NWM (Table 4).

Soil C at a depth of 0–25 cm increased with site age (Fig. 5;  $F_{1,8} = 7.57$ ,  $P < 0.05$ ). The soil C accumulation rate was  $60 \text{ g C m}^{-2} \text{ yr}^{-1}$ , according to the regression analysis of a linear mixed model. At soil depths of 0–5, 5–15, and 15–25 cm, soil C did not increase over time ( $F_{1,8} = 3.05$ ,  $P = 0.119$ ;  $F_{1,8} = 2.67$ ,  $P = 0.141$ ;  $F_{1,8} = 4.04$ ,  $P = 0.0794$ ).

## 4. Discussion

### 4.1. Accumulation of soil carbon and WSA

NWM enhances the ecological functions that improve soil C sequestration. Our results indicated that without external inputs, soil C increased with time after adopting NWM. The soil C sequestration

rate at our research site was  $60 \text{ g C m}^{-2} \text{ yr}^{-1}$  at a soil depth of 0–25 cm for 17 years, and this value was similar or relatively faster than values reported in previous studies (Lal et al., 1999; Six et al., 2002; West and Post, 2002). However, in previous study, soil C decreased following the conversion of the site from conventional management to fallow management (Shimoda and Koga et al., 2013). Additionally, the soil C accumulation rate increased with decreasing frequency of fallow management (Campbell et al., 2001), likely because fallow rotations decreased root and microbial activity and the input of organic matter compared to cropping (Alvaro-Fuentes et al., 2008; Franzluebbers et al., 1994). In contrast, the adoption of fallow periods with perennial grass increased soil C compared to fallow periods without perennial grass, and long-term plant diversity restoration practices increased soil C by enhancing the above- and belowground biomass (De Deyn et al., 2011; Luo et al., 2010). Previous studies have reported that soil C sequestration rates increase when cropland management practices change. The conversion from conventional tillage to a no-tillage practice increased soil C sequestration by an average of  $57 \pm 14 \text{ g C m}^{-2} \text{ yr}^{-1}$ , excluding a change to no-tillage in wheat–fallow systems (West and Post, 2002). Lal et al. (1999) estimated that the C sequestration potential through the improved management of cropland from conventional tillage to conservation management was  $24\text{--}40 \text{ g C m}^{-2} \text{ yr}^{-1}$ . A general increase in C levels ( $32.5 \pm 11.3 \text{ g C m}^{-2} \text{ yr}^{-1}$ ) has been observed under a no-tillage system compared with conventional tillage at a soil depth of 0–30 cm in both tropical and temperate soils (Six et al., 2002). The change in soil C was  $-95 \text{ g C m}^{-2} \text{ yr}^{-1}$  after the conversion from grassland to cropland, and  $49 \text{ g C m}^{-2} \text{ yr}^{-1}$  after the conversion from cropland to grassland (Soussana et al., 2004). Changes in soil C were influenced by rotation, tillage operations, and production inputs (Post and West, 2002). We also reported previously that NWM increased soil C compared to no-tillage management, when weeds were controlled by hand-picking in an Andosol field experiment (Arai et al., 2013).

The results of this study indicated that soil C ( $\text{g m}^{-2}$ ) at each soil depth (0–5, 5–15, and 15–25 cm) did not change significantly over time since adopting NWM (data not shown); however, total depth of soil C in the three layers did increase over time under NWM. Previous studies have reported that soil

C increased only at the soil surface and either did not change or even decreased in deep layers under no-tillage management without the application of organic matter. Soil C did not therefore change throughout the total depth of the soil under no-tillage management compared to conventional management. No-tillage farming increases soil organic carbon (SOC) concentrations in the surface layers of some soils, but no-tillage soils do not store more SOC than conventional tillage soils throughout the soil profile at depths of 0 to <60 cm (Blanco-Canqui and Lal, 2008; Blanco-Canqui et al., 2011). Additionally, Luo et al. (2010) reported that after adopting no-tillage, soil C increased by  $315 \pm 242 \text{ g m}^{-2}$  in the top 10 cm of soil, but declined by  $330 \pm 161 \text{ g m}^{-2}$  at a depth of 20–40 cm, indicating that the adoption of no-tillage did not enhance the total soil C stock down to 40 cm. Koga and Tsuji (2009) reported that reduced tillage had no positive effects on overall soil C sequestration (0–30 cm) when compared to conventional tillage in an Andisol field. In contrast, a combination of residue return and manure application exerted a beneficial impact by increasing residue C and reducing the loss of SOC (Koga and Tsuji, 2009).

The reasons for the increase in soil C over time since the adoption of NWM are explained below. First, the increase in soil C at depths of 0–25 cm is probably due to the increase in weed aboveground biomass over time since the adoption of NWM. Weeds in agricultural areas are usually controlled by herbicide applications (Soane et al., 2012). At our study site, the weed management involved mowing, and slashed weeds were applied to the soil surface as mulch, once a month from April to October. The increase in surface residue under no-tillage practice caused soil C to accumulate, and the accumulation rate was rapid compared with the rates observed under plow tillage and ridge tillage practices (Duiker and Lal, 1999). Therefore, an increase in input residues through the use of slashed weeds as mulch contributes to soil C accumulation after conversion from conventional management to NWM. Weed root exudates were also increased by mowing and remained in the soil as a source of C, contributing to the formation of soil aggregates, which in turn enhanced soil C sequestration possibility (Holland et al., 1996; Niwa et al., 2008; Six et al., 2004). Dolan et al. (2006) found that the surface soil (0–20 cm) under a no-tillage system had 30% greater SOC than soil under moldboard plow and chisel plow

tillage treatments. This trend was reversed at a soil depth of 20–25 cm, when significantly more SOC was found in soil under moldboard plow treatments than in soil under a no-tillage treatment. The authors noted that residues buried by inversion might contribute to the observed C increase. The method we used to study roots may not have been deep enough (0–4 cm) to determine differences due to site age; however, we found that plant debris, including roots, only increased under NWM at a soil depth of 15–25 cm. The abundance of soil C at each soil layer did not differ significantly over time under NWM at our study site; however, soil C at a depth of 15–25 cm did not decrease due to increasing organic matter.

The second reason for the increase in soil C was that WSA >2 mm accumulated after conversion from conventional management to NWM. The results of our study showed that soil aggregates (>2 mm) increased over time under NWM at a soil depth of 0–15 cm. Soil aggregates (>2 mm) increased due to both the decreasing frequency of soil disturbance from tillage and seeding, and the increasing input of organic materials (Causarano et al., 2008; Pinheiro et al., 2003). Plaza-Bonilla et al. (2013) reported that differences in WSA between treatments were only found at soil depths of 0–5 and 5–10 cm and that WSA (>2 mm) were found in greater proportions at a soil depth of 0–5 cm in the 11th and 20th years after adopting no-tillage management and conventional management, respectively. In a study by Jiao et al. (2006) on a cropland composed of sandy loam, the adoption of a no-tillage system and treatments of at least 30 Mg wet weight ha<sup>-1</sup> yr<sup>-1</sup> of composted cattle manure increased the proportion of WSA (>2 mm) within 4 years. The results of an analysis of WSA (>2 mm) at a soil depth of 0–5 cm indicated that WSA tended to increase in the 5 years after switching management practices. Under NWM at a soil depth of 0–5 cm, the proportions of WSA in the >2-mm and 0.25–1.0mm size fractions in the first year of management were 14% and 40%, respectively; in the fifth year of management, the corresponding proportions of WSA 0.25–1 mm were 34% and 24%, respectively. Therefore, WSA (>2 mm) might be composed of the 0.25–1-mm size fraction with binding agents, such as plant debris and root exudates. Cater (2002) suggested that the factors enhancing soil C in sandy loam soils were C inputs and soil aggregate formation. Macroaggregates

include microaggregates that do not undergo decomposition by microorganisms (Oades, 1984); therefore, the increased formation of macroaggregates (>2 mm) contributed to the accumulation and maintenance of soil C. At our study site, soil C might have increased due to the soil having sufficient sources of C and the formation of soil aggregates. Six et al. (2004) found that root penetration altered the effects of the soil water regime, root exudation, dead root decomposition, and root entanglement on soil aggregate formation. Root exudates act as binding agents, adhering to soil particles with organic matter, and the turnover of C in exudates is rapid compared to root and shoots (Paterson, 2003). Our results suggested that root biomass did not differ significantly, while the aboveground weed biomass increased following the adoption of NWM. Therefore, root biomass needs to be investigated at greater depths.

#### *4.2. Relationship between inputs of organic matter and soil C accumulation rate*

Although many studies have been conducted, the process of soil C sequestration remains largely unknown. For the effective management of C sequestration, understanding the relationship between inputs of C to soil and its accumulation in the soil C is important. Jarecki and Lal (2003) reported that crop residue as a source of soil C has the potential for soil C sequestration, but they did not provide an annual estimate. An earlier study had reported that under conventional practices, the application of manure as the main C source enhanced WSA accumulation and possibly increased soil C (Aoyama et al., 1999; Whalen et al., 2003). The major soil C source was assumed to be weeds at our study site, and the annual C input from the aboveground weeds and litter, applied once a month from April to October, was estimated to be 265–2317 g C m<sup>-2</sup> yr<sup>-1</sup> within a range of 0–17 years (data for the aboveground weed C concentration are not shown). Carbon in the aboveground weeds and litter represented 2–11% of the soil C at our study site. This estimation did not include root biomass and microbial activity; root exudates are easily decomposable organic matter, and soluble exudates contribute only 1–10% of the total C flow (Paterson et al., 2003). Aoyama and Kumakura (2001)



reported that the application of cattle manure at a rate of  $32,000 \text{ g m}^{-2} \text{ yr}^{-1}$  ( $9278 \text{ g C m}^{-2} \text{ yr}^{-1}$  estimated using a water content of 81.9% and C content of 35.4% by Takama and Hirosawa, 2008) each spring to the surface of a soil increased the soil C at a depth of 30 cm by 30% during a 20-year period. At our study site, soil C had increased by 25% in year 17 compared to year 0. Shimoda and Koga (2013) studied fallow management practices that were previously used for rice and wheat cultivation during 3 years under three different management systems. Their study showed that soil C storage increased by about 1.3 times in former paddies, and decreased by 0.83–0.91 times in former uplands during a 3-year period. They found that belowground production was 2.0–4.7 times (maximum value:  $559 \text{ g m}^{-2}$ ) greater in former paddies than former uplands, suggesting that the lower C input from the annual grasses caused a loss in soil C in the upland fields. Our value for aboveground weed and litter C ( $\text{g m}^{-2}$ ) was low compared with previous studies in which manure was applied; however, the formation of WSA included soil C that originated from weeds, and thus enhanced soil C accumulation possibility.

#### 4.3. *Effect of earthworm activities on WSA*

Earthworm abundance was expected to increase under NWM compared with conventional management, and earthworms were thought to contribute to the formation of soil aggregates and an improved soil environment due to presence of live weeds and decreased levels of disturbance. Previous studies have indicated that earthworm density and biomass increases under no-tillage management compared with conventional management due to the decreased level of soil disturbance (Kladviko, 2001). Earthworm density has been reported to increase by two to nine times under no-tillage management compared with conventional management (Chan, 2001), and the number of three to five species of lumbricid earthworms was  $69 \text{ (m}^{-2}\text{)}$  under a tillage system and  $499 \text{ (m}^{-2}\text{)}$  under a no-tillage system in New Zealand (Springett, 1992). Earthworm density was very low or zero under a no-tillage system, reduced tillage treatment, and conventional tillage treatment in Japan (Arai et al., 2013; Nakamoto et al., 2006). However earthworm abundance and biomass increased under NWM

compared to a no-tillage system (Arai et al., 2013; Miura et al., 2010), and this increase improved soil C accumulation in cropland (Arai et al., 2013). Fonte et al. (2009) conducted a field experiment demonstrating that earthworm biomass and soil C changed according to the management practice used, such as mulch, cover crop, and bare fallow. They found that earthworm biomass, soil aggregate stability, and soil C were influenced by mulch treatment when compared with bare fallow treatment. Decaëns (2000) estimated that the half-life of an earthworm cast in a pasture ranged from 2 to 11 months, indicating that earthworm casts remain in the soil even in the absence of stable earthworm populations.

Our study showed that MWD at a depth of 0–15 cm increased over time following the adoption of NWM. A previous study found that the larger soil aggregate size fractions increased under reduced and no-tillage management because of the reduction in soil disturbance (Jiao et al., 2006). Winsome and McColl (1998) conducted a 6-month incubation experiment on earthworms in microcosms consisting of A-horizon soil and litter, and found that most (90%) of the aggregates in earthworm casts were stable in water, with an MWD of 2.8 mm. In contrast, bulk soil contained 60% WSA, with an MWD of 0.6 mm. Because earthworm casts were mostly WSA (Kawaguchi et al., 2011), the greater MWD values compared to soil without earthworms were due to the increase in soil aggregates in the >2-mm size fraction produced by earthworms (Bossuyt et al., 2004). The results of our study indicated that the MWD at soil depths of 0–5 and 5–15 cm increased after adopting NWM and that earthworms modified the soil structure through their activity. The earthworm (Megascolecidae) casting and activity period in Japan assumed to last from May to August, because high biomass and individuals tended to be found until August (Uchida and Kaneko, 2004). The proportion of earthworm casts in the WSA >2-mm size fraction (0–5 cm) was estimated to be between 0 and 46% at each site, according to our results for earthworm biomass of all species and a casting rate of 0.79 g dry weight g<sup>-1</sup> earthworm fresh weight day<sup>-1</sup> (Kawaguchi et al., 2011). Earthworm biomass explained a relatively small portion of the increase in the WSA > 2-mm size fraction. However, this calculation uses an estimate of cast production by epigeic species because we have no precise record of cast production by Japanese

endogeic and anecic earthworms, and we did not include the soil aggregate dynamics such as mean residence time and breaking rate. Therefore, further studies are required to estimate earthworm contribution to aggregate formation.

## **5. Conclusion**

The results of this study support our hypothesis that aboveground weed biomass increased with adopting NWM, and then WSA size distribution and soil C accumulation would be influenced. However, belowground weed biomass did not be affected by years of NWM. NWM management can accumulate soil C at a relatively high rate ( $60 \text{ g C m}^{-2} \text{ yr}^{-1}$ ). In agricultural fields, weeds are controlled physically or chemically and are not expected to be a C source, while under NWM, weeds are a major C input to soil. The formation of WSA is enhanced due to weed root and earthworm activities, and aggregates will remain in the soil longer due to a lack of soil disturbance. These biological factors may sequester soil C even under low C input conditions. Further studies are necessary to clarify the effects of the roots of weeds and the feeding and casting of earthworms on soil aggregation to fully evaluate ecosystem functioning in agricultural fields.

## **References**

- Alvaro-Fuentes, J., Arrue, J.L., Gracia, R., Lopez, M. V., 2008. Tillage and cropping intensification effects on soil aggregation: temporal dynamics and controlling factors under semiarid conditions. *Geoderma* 145, 390–396.
- Aoyama, M., Kumakura, N., 2001. Quantitative and qualitative changes of organic matter in an Ando soil induced by mineral fertilizer and cattle manure applications for 20 years. *Soil Sci. Plant Nutr.* 47, 241–252.

- Aoyama, M., Angers, D. A., N'Dayegamiye, A., 1999. Particulate and mineral-associated organic matter in water-stable aggregates as affected by mineral fertilizer and manure applications. *Can. J. Soil Sci.* 79, 295–302.
- Arai, M., Tayasu, I., Komatsuzaki, M., Uchida, M., Shibata, Y., Kaneko, N., 2013. Changes in soil aggregate carbon dynamics under no-tillage with respect to earthworm biomass revealed by radiocarbon analysis. *Soil Tillage Res.* 126, 42–49.
- Blanco-Canqui, H., Lal, R., 2008. No-tillage and soil-profile carbon sequestration: an on- farm assessment. *Soil Sci. Soc. Am. J.* 72, 693–701.
- Blanco-Canquia, H., Schlegel, A.J., Heerc, W.F., 2011. Soil-profile distribution of carbon and associated properties in no-till along a precipitation gradient in the central Great Plains. *Agric. Ecosyst. Environ.* 144, 107–116.
- Bossuyt, H., Six, J., Hendrix, P.F., 2004. Rapid incorporation of fresh residue-derived carbon into newly formed stable microaggregates within earthworm casts. *Eur. J. Soil Sci.* 55, 393–399.
- Bot, A., Benites, J., 2005. The Importance of Soil Organic Matter — Key to Drought-resistant Soil and Sustained Food Production. *FAO Soils Bulletin* 80.
- Bronick, C.J., Lal, R., 2005. Soil structure and management: a review. *Geoderma* 124, 3–22.
- Campbell, C.A., Selles, F., Lafond, G.P., Biederbeck, V.Q., Zenter, R.P., 2001. Tillage-fertilizer changes: effect on some soil quality attributes under long-term crop rotations in a thin Black Chernozem. *Can. J. Soil Sci.* 81, 157–165.
- Cardina, J., Regnir, E., Harrison, K., 1991. Long-term tillage effects on seed banks in three Ohio soils. *Weed Sci.* 39, 186–194.
- Carter, M.R., 2002. Soil quality for sustainable land management: organic matter and aggregation interactions that maintain soil functions. *Agron. J.* 94, 38–47.
- Causarano, H.J., Franzluebbbers, A.J., Shaw, J.N., Reeves, D.W., Raper, R.L., Wood, C.W., 2008. Soil organic carbon fractions and aggregation in the Southern Piedmont and Coastal Plain. *Soil Sci. Soc. Am. J.* 72, 221–230.

- Chan, K.Y., 2001. An overview of some tillage impacts on earthworm population abundance and diversity — implications for functioning in soils. *Soil Tillage Res.* 57, 179–191.
- De Deyn, G.B., Shiel, R.S., Ostle, N.J., McNamara, N.P., Oakley, S., Young, I., Freeman, C., Fenner, N., Quirk, H., Bardgett, R.D., 2011. Additional carbon sequestration benefits of grassland diversity restoration. *J. Appl. Ecol.* 48, 600–608.
- Decaëns, T., 2000. Degradation dynamics of surface earthworm casts in grasslands of the eastern plains of Colombia. *Biol. Fertil. Soils* 32, 149–156.
- Dolan, M.S., Clapp, C.E., Allmaras, R.R., Baker, J.M., Molina, J.A.E., 2006. Soil organic carbon and nitrogen in a Minnesota soil as related to tillage, residue and nitrogen management. *Soil Tillage Res.* 89, 221–231.
- Duiker, S.W., Lal, R., 1999. Crop residue and tillage effects on carbon sequestration in a Luvisol in central Ohio. *Soil Tillage Res.* 52, 73–81.
- Edmeades, D.C., 2003. The long-term effects of manures and fertilisers on soil productivity and quality: a review. *Nutr. Cycl. Agroecosyst.* 66, 165–180.
- Fonte, S.J., Winsome, T., Six, J., 2009. Earthworm populations in relation to soil organic matter dynamics and management in California tomato cropping systems. *Appl. Soil Ecol.* 41, 206–214.
- Fonte, S.J., Quintero, D.C., Velásquez, E., Lavelle, P., 2012. Interactive effects of plants and earthworms on the physical stabilization of soil organic matter in aggregates. *Plant Soil* 359, 205–214.
- Franzluebbers, A.J., Hons, F.M., Zuberer, D.A., 1994. Seasonal changes in soil microbial biomass and mineralizable C and N in wheat management systems. *Soil Biol. Biochem.* 26, 1469–1475.
- Fukuoka, M., 1987. *The Natural Way of Farming: The Theory and Practice of Green Philosophy.* (P.M. Frederic, Trans.) Rev. ed. Japan Publications, Japan.
- Goto, S., Hatai, S., 1898. New or imperfectly known species of earthworms. No. 1. *Annotationes Zoologicae Japonenses* 2, 65–78.

- Goto, S., Hatai, S., 1899. New or imperfectly known species of earthworms. No. 2. *Annotationes Zoologicae Japonenses* 3, 13–24.
- Holland, J.M., 2004. The environmental consequences of adopting conservation tillage in Europe: reviewing the evidence. *Agric. Ecosyst. Environ.* 103, 1–25.
- Holland, J.N., Cheng, W., Crossley, D.A., 1996. Herbivore-induced changes in plant carbon allocation: assessment of below-ground C fluxes using carbon-14. *Oecologia* 107, 87–94.
- ISO, 2006. 23611–1: Soil Quality — Sampling of Soil Invertebrates. Part 1: Hand-sorting and Formalin Extraction of Earthworms. International Organization for Standardization, Geneva, Switzerland.
- Jarecki, M.K., Lal, R., 2003. Crop management for soil carbon sequestration. *Critical Reviews in Plant Sciences* 22, 471–502.
- Jiao, Y., Whalen, J.K., Hendershot, W.H., 2006. No-tillage and manure applications increase aggregation and improve nutrient retention in a sandy-loam soil. *Geoderma* 134, 24–33.
- Kawaguchi, T., Kyoshima, T., Kaneko, N., 2011. Mineral nitrogen dynamics in the casts of epigeic earthworms (*Metaphire hilgendorfi*: Megascolecidae). *Soil Sci. Plant Nutr.* 57, 387–395.
- Kemper, W.D., Rosenau, R.C., 1986. Aggregate stability and size distribution, In: Klute, A. (Ed.), *Methods of Soil Analysis. Part 1: Physical and Mineralogical Methods*, 2nd ed. Agronomy. Am. Soc. Agron, Madison, Wisconsin, pp. 425–442.
- Kladivko, E.J., 2001. Tillage systems and soil ecology. *Soil Tillage Res.* 61, 61–76.
- Kladivko, E.J., Akhouri, N.M., Weesies, G., 1997. Earthworm populations and species distributions under no-till and conventional tillage in Indiana and Illinois. *Soil Biol. Biochem.* 314, 613–615.
- Koga, N., Tsuji, H., 2009. Effects of reduced tillage, crop residue management and manure application practices on crop yields and soil carbon sequestration on an Andisol in northern Japan. *Soil Sci. Plant Nutr.* 55, 546–557.
- Lal, R., 2004. Soil carbon sequestration to mitigate climate change. *Geoderma* 133, 1–22.
- Lal, R., 2009. Soil degradation as a reason for inadequate human nutrition. *Food Secur.* 1, 45–57.

- Lal, R., Follett, R.F., Kimble, J., Cole, C.V., 1999. Managing U.S. cropland to sequester carbon in soil. *J. Soil Water Conserv.* 54, 374–381.
- Ludwig, B., Geisseler, D., Michel, K., Joergensen, R.G., Schulz, E., Merbach, I., Raupp, J., Rauber, R., Hu, K., Niu, L., Liu, X., 2011. Effects of fertilization and soil management on crop yields and carbon stabilization in soils. A review. *Agron. Sustain. Dev.* 31, 361–372.
- Luo, Z., Wang, E., Sun, O.J., 2010. Soil carbon change and its responses to agricultural practices in Australian agro-ecosystems: a review and synthesis. *Geoderma* 155, 211–223.
- Michaelsen, W., 1891. Oligochaeta des Naturhistorischen Museums in Hamburg. IV. Jb. Hamb. Wiss. Anst. 8, 3–42.
- Mikha, M.M., Rice, C.W., 2004. Tillage and manure effects on soil and aggregate-associated carbon and nitrogen. *Soil Sci. Soc. Am. J.* 68, 809–816.
- Miura, T., Kaneko, N., Komatsuzaki, M., 2010. Earthworm contributes to supply available phosphorus in agricultural soil with no-tillage, weed-cover and low-input management. A case study of nature farming in Ibaraki Prefecture. *Org. Agric. Sci.* 2, 30–39 (in Japanese).
- Muramoto, J., Werner, M.R., 2002. Mustard powder vermifuge: an alternative to formalin expulsion method for earthworm sampling. *Edaphologia* 70, 7–11.
- Nakamoto, T., Tsukumoto, M., 2006. Abundance and activity of soil organisms in fields of maize grown with a white clover living mulch. *Soil Biol. Biochem.* 115, 34–42.
- Niwa, S., Kaneko, N., Okada, H., Sakamoto, K., 2008. Effects of fine-scale simulation of deer browsing on soil micro-foodweb structure and N mineralization rate in a temperate forest. *Soil Biol. Biochem.* 40, 699–708.
- Oades, J.M., 1984. Soil organic matter and structural stability: mechanisms and implications for management. *Plant Soil* 76, 319–337.
- Paterson, E., 2003. Importance of rhizodeposition in the coupling of plant and microbial productivity. *Eur. J. Soil Sci.* 54, 741–750.

- Pinheiro, E.F.M., Pereira, M.G., Anjos, L.H.C., 2004. Aggregate distribution and soil organic matter under different tillage systems for vegetable crops in a Red Latosol from Brazil. *Soil Tillage Res.* 77, 79–84.
- Plaza-Bonilla, D., Cantero-Martínez, C., Viñas, P., Álvaro-Fuentes, J., 2013. Soil aggregation and organic carbon protection in a no-tillage chronosequence under Mediterranean conditions. *Geoderma* 193–194, 76–82.
- R Development Core Team, 2012. *A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna.
- Riley, H., Pommeresche, R., Eltun, R., Hansen, S., Korsæth, A., 2008. Soil structure, organic matter and earthworm activity in a comparison of cropping systems with contrasting tillage, rotations, fertilizer levels and manure use. *Agric. Ecosyst. Environ.* 124, 275–284.
- Shi, X.M., Li, X.G., Long, R.J., Singh, B.P., Li, Z.T., Li, F.M., 2010. Dynamics of soil organic carbon and nitrogen associated with physically separated fractions in a grassland- cultivation sequence in the Qinghai–Tibetan plateau. *Biol. Fertil. Soils* 46, 103–111.
- Shimoda, S., Koga, N., 2013. Rapid change in soil C storage associated with vegetation recovery after cessation of cultivation. *Soil Sci. Plant Nutr.* 59, 27–34.
- Six, J., Elliott, E.T., Paustian, K., 2000. Soil macroaggregate turnover and micro-aggregate formation: a mechanism for C sequestration under no-tillage agriculture. *Soil Biol. Biochem.* 32, 2099–2103.
- Six, J., Feller, C., Denef, K., Ogle, S.M., Sa, J.C.D., Albrecht, A., 2002. Soil organic matter, biota and aggregation in temperate and tropical soils — effects of no-tillage. *Agronomie* 22, 755–775.
- Six, J., Bossuyt, H., Degryze, S., Denef, K., 2004. A history of research on the link between (micro) aggregate, soil biota, and soil organic matter dynamics. *Soil Tillage Res.* 79, 7–31.
- Soane, B.D., Ball, B.C., Arvidsson, J., Basch, G., Moreno, F., Roger-Estrade, J., 2012. No-till in northern, western and south-western Europe: a review of problems and opportunities for crop production and the environment. *Soil Tillage Res.* 118, 66–87.



- Soussana, J.F., Loiseau, P., Vuichard, N., Ceschia, E., Balesdent, J., Chevallier, T., Arrouays, D., 2004. Carbon cycling and sequestration opportunities in temperate grasslands. *Soil Use Manag.* 20, 219–230.
- Spandl, E., Durgan, B.R., Forcella, F., 1999. Foxtail (*Setaria* spp.) seedling dynamics in spring wheat (*Triticum aestivum*) are influenced by seeding date and tillage regime. *Weed Sci.* 47, 156–160.
- Springett, J.A., 1992. Distribution of lumbricid earthworms in New Zealand. *Soil Biol. Biochem.* 24, 1377–1381.
- Stockmann, U., Adams, M.A., Crawford, J.W., Field, D.J., Henakaarchchi, N., Jenkins, M., Minasny, B., McBratney, A.B., de Courcelles, V.R., Singh, K., Wheeler, I., Abbott, L., Angers, D.A., Baldock, J., Bird, M., Brookes, P.C., Chenu, C., Jastrow, J.D., Lal, R., Johannes Lehmann, J., O'Donnell, A.G., Parton, W.J., Whitehead, D., Zimmermann, M., 2013. The knowns, known unknowns and unknowns of sequestration of soil organic carbon. *Agric. Ecosyst. Environ.* 164, 80–99.
- Takama, Y., Hirose, M., 2008. The proper technique for continuous application of organic material judging from physicochemical properties of soil, crop yield, and the impact on surrounding areas. *Bull. Tochigi Agr. Exp. Stn.* 63, 35–45 in Japanese.
- Uchida, T., Kaneko, N., 2004. Life history of Megascolecidae earthworms in forest soils at Kanagawa, Japan. *Edaphologia* 74, 35–45 in Japanese.
- Watanabe, et al. Chronosequential changes of the soil physicochemical properties in No-Till and Organic farming (nature farming) technology under upland crop field in western Japan in preparation.
- West, T.O., Post, W.M., 2002. Soil organic carbon sequestration rates by tillage and crop rotation: a global data analysis. *Soil Sci. Soc. Am. J.* 66, 1930–1946.
- Whalen, J.K., Hu, Q., Liu, A., 2003. Manure applications improve aggregate stability in conventional and no-tillage systems. *Soil Sci. Soc. Am. J.* 67, 1842–1847.

Winsome, T., McColl, J.G., 1998. Changes in chemistry and aggregation of a California forest soil worked by the earthworm *Argilophilus papillifer* Eisen (Megascolecidae). *Soil Biol. Biochem.* 30, 1677–1687.

Yoder, R.E., 1936. A direct method of aggregate analysis of soils and a study of the physical nature of soil erosion losses. *J. Am. Soc. Agron.* 28, 337–351.

## Figures

**Fig. 1.** (a) Aboveground weed biomass ( $n = 2$ ), (b) litter accumulation ( $n = 2$ ), and (c) root biomass (soil depth of 0-4 cm,  $n = 2$ ). A line of (a) shows liner regression ( $y = 12x + 219$ ).

**Fig. 2.** (a) Earthworm density ( $n = 2$ ) and (b) biomass ( $n = 2$ ) in 2011 and in 2012.

**Fig. 3.** Changes in size distribution of water stable aggregate at 10 sites of increasing site age at the no-tillage and weed mulch management after conversion from conventional management. Symbols represent the mean values of the two site of each site age ( $n = 2$ ).

**Fig. 4.** Change in soil C stock of water stable aggregate size fraction at ten sites of increasing site age at the no-tillage with weed mulch management after conversion from conventional management. Symbols represent the mean values of the two site of each site age ( $n = 2$ ).

**Fig. 5.** Change in soil C at ten sites of increasing site age at the no-tillage and weed mulch management after conversion from conventional management that was analyzed by applying liner mixed model (soil C ( $\text{g C m}^{-2} \text{ yr}^{-1}$ ) =  $60 A + 4092 + B$  (A: site age (year), B: random effect)). Symbols represent the mean values of the two site of each site age ( $n = 2$ ). A line shows that soil C ( $\text{g C m}^{-2} \text{ yr}^{-1}$ ) =  $60 A + 4092$  (A: site age (year)).

Fig. 1

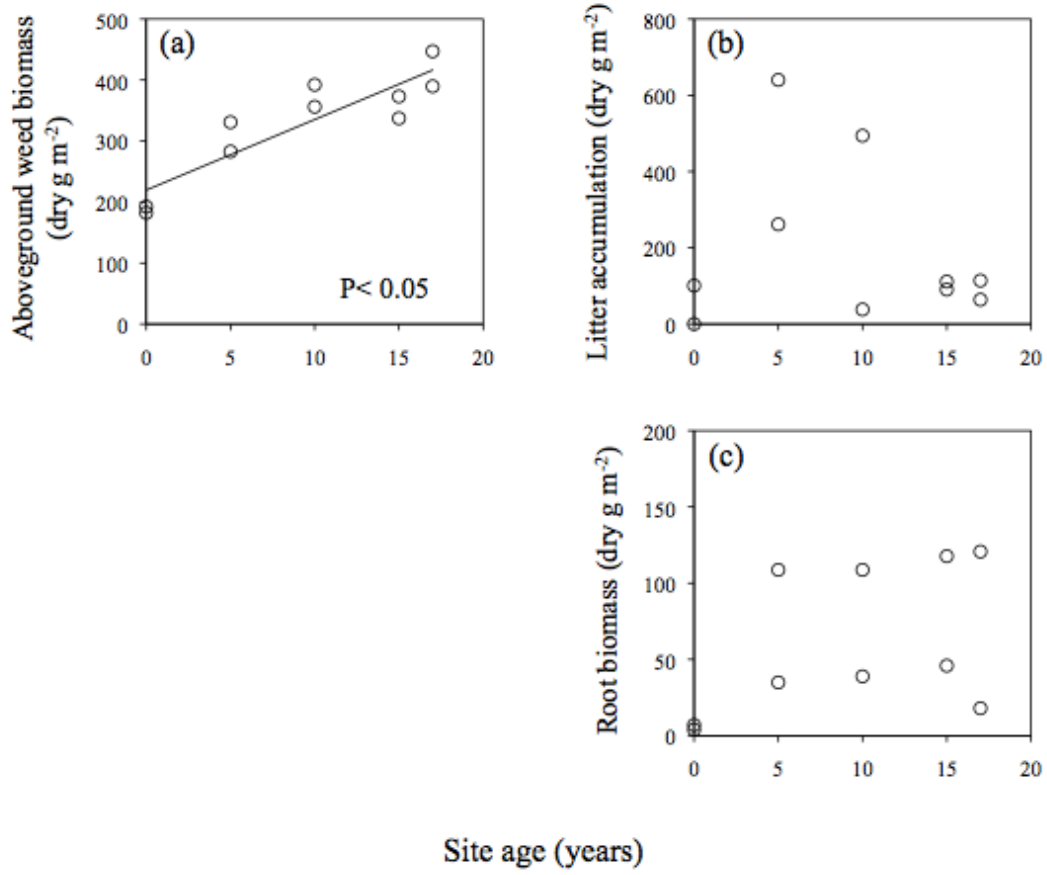


Fig. 2

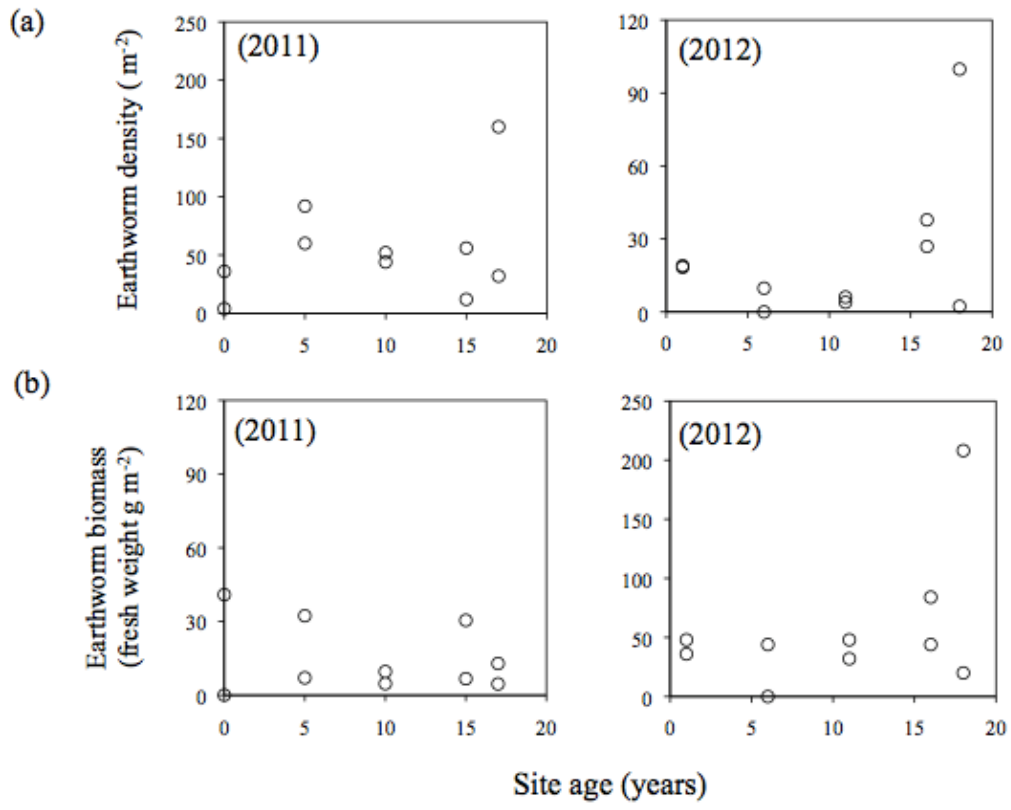


Fig. 3

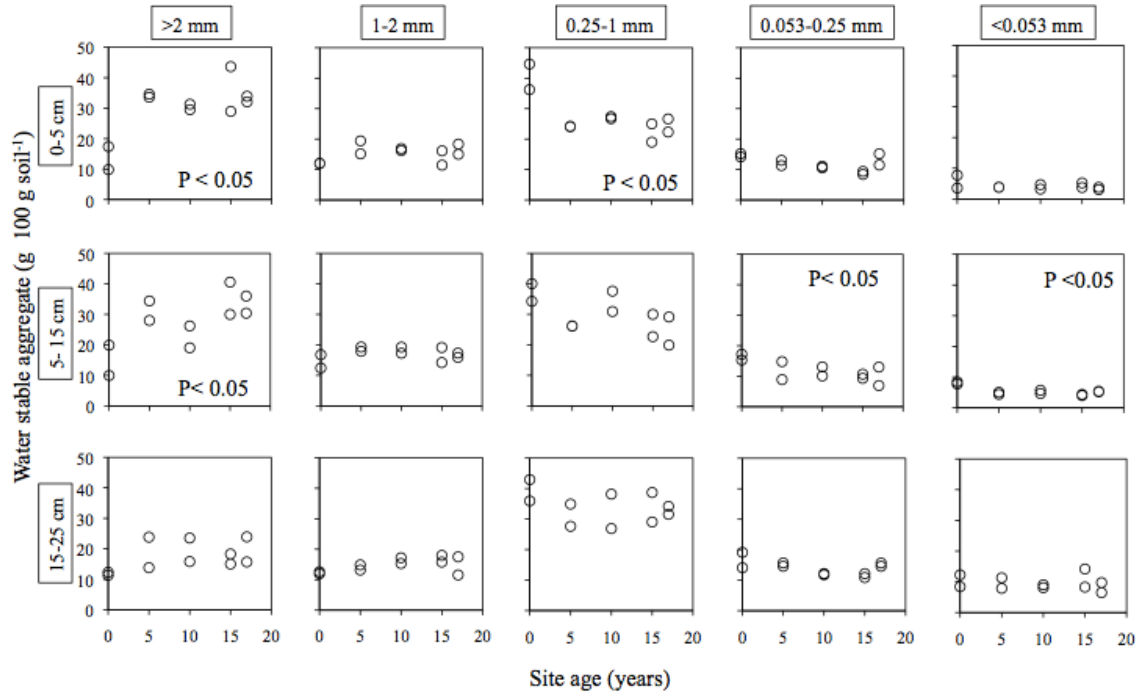


Fig. 4

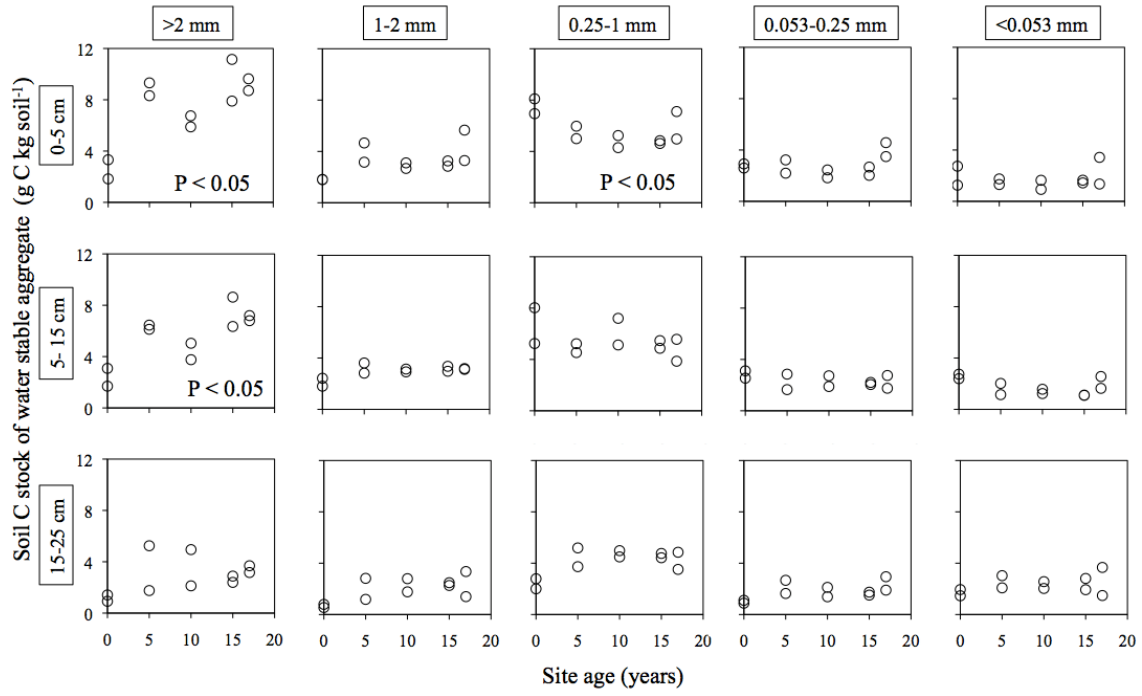


Fig. 5

