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著者	UTOMO M., NISWATI A., DERMI YATI, WATI M. R., RAGUAN E. F., SYARIF S.
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M. UTOMO*, A. NISWATI, DERMIYATI, M. R. WATI, E. F. RAGUAN and S. SYARIF

Sustainable Soil Management, University of Lampung,
Jl. Sumantri Brojonegoro #1, Bandar Lampung, 35145, Indonesia
*e-mail: mutomo@unila.ac.id

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Abstract

Soil carbon and earthworm are important components in sustainable tropical agro-ecosystem. Long-term experiment was initially conducted in February 1987, at experiment farm of *Politeknik Negeri* Lampung, Sumatra, Indonesia. The soil is a Typic Fragiudult with slope ranging from 6 to 9%, located at 105°13'E, 05°21'S, with elevation 122 m. The experiment was a factorial, randomized complete block design, with 4 replications. Tillage treatments were conservation tillage (no-tillage, NT and minimum tillage, MT), and conventional tillage (CT); while nitrogen fertilization rates were 0, 100 and 200 kg N ha⁻¹. Cropping pattern of the long-term experiment was cereal-legume-fallow rotation. Due to soil compaction in 1997 and 2002, all plots of conservation tillage were plowed. After eleven years of cropping, the soil became acid, therefore; in 2004 all plots were limed with 4 Mg ha⁻¹ of CaCO₃. To determine the effect of long-term no-tillage and N fertilization on earthworm, the soil samples were collected at depth of 0-10 cm in 2001 and 2008; and for soil microbial biomass C and soil organic C, samples were taken at depth 0-20 cm in 2008 after 21 years of continuous experiment.

The only treatment that affected earthworm population in both 14th and 21st year of cropping was tillage treatment. After 14 years of cropping (2001), NT had earthworms averaged of 101m⁻², 46% and 39% ($p<0.05$) higher than CT and MT, respectively. In 21st year of cropping, however, the response showed a little change; NT had earthworm with averaged of 99 m⁻², 251% higher ($p<0.05$) than CT, but 40% lower than MT. In rhizosphere samples, MT had microbial biomass 228 mg C-CO₂ kg⁻¹day⁻¹, or 45.5% higher

($p<0.05$) than NT and 70.7% higher than CT; while 100 kg N ha⁻¹ N treatment had 227 mg C-CO₂ kg⁻¹ day⁻¹, 28% and 103% higher ($p<0.05$) than 0 and 200 kg N ha⁻¹ treatments, respectively. After 21 years of cropping, soil organic carbon of MT in rhizosphere at 0-20 cm soil depth was 16.5 g kg⁻¹, or 9.3% and 13.0% higher ($p<0.05$) than NT and CT, respectively; but in non-rhizosphere was not significantly ($p<0.05$) affected by any treatment.

Introduction

No-tillage is one of few revolutions in agriculture that has greatly impacted agriculture throughout the world (Triplett and Dick, 2008). In fact, no-tillage is an academically modified ancient land preparation technology namely '*slash and burn*' that successfully has been practiced in modern agriculture (Utomo, 2004). Due to the ability to conserve soil and to increase efficiency, worldwide adoption of no-tillage has been rapidly expanded since about 1990, particularly in the United States, South American countries and Africa. While Asia and Europe have been slower to adopt no-tillage, primarily due to their long history of conventional tillage practices. However, reports from Conference of the International Soil Tillage Research indicate this is changing (Derpsch, 1998; Triplett and Dick, 2008). In Indonesia, no-tillage has been initially promoted by few no-tillage researchers in 1980's, and successfully practiced by farmers just in 1990's, particularly in the region with lack of labors, such as in Sumatra, Borneo and Celebes. Yet in 1998, no-tillage has been explicitly stated in national land preparation policy (Utomo, 2004).

As for conservation tillage, no-tillage requires at least 30% plant residues as mulch covering soil sur-

face (Lal, 1989). Plant residues serve as substrate that is converted to microbial biomass and soil organic matter, and have the potential to enhance carbon sequestration in agricultural soils (Wright and Hons, 2004). The presence of plant residues will create better microclimate that can enhance soil biota activity, especially earthworms (Lavelle, 1984; Brito-Vega, et al., 2009). However, the responses of earthworm population to no-tillage have been variable, and the impact is dependent on soil factors, climatic conditions and the tillage operations. Results of survey in USA showed that eight of the 14 sites had higher earthworm populations in no-tillage than in conventional tillage, with increases ranging from 25% higher to 10 times higher (Kladivko, 1993). The declines in earthworm population often reported in conventionally tilled soils are associated with undesirable changes in soil environmental condition resulting from excessive tillage (Chan, 2001). Research conducted in dry land farming in the Pacific Northwest, USA showed that earthworm populations under no-tillage were more abundance than under conventional tillage (Wuest, 2001). Brito-Vega et al. (2009) reported that in Mexico earthworm populations under conservation tillage at depth of 10-20 cm were 240 individuals m⁻² in spring and reduced to 60 individuals m⁻² in winter; while under conventional tillage reduced from 192 in spring to 72 individuals m⁻² in winter. The survey on nine years of soybean-barley field under different cropping systems in Japan confirmed that, there were no earthworms in the tilled field without mulch, a few in no-tillage without mulch, but there were increasing number of earthworms in no-tillage with mulch and reached the peak in the ninth year of cropping (Nakamura, et al., 2003).

The increasing numbers of earthworm and other soil biota in no-tillage can contribute to carbon sequestration in agro-ecosystem. Earthworm (*Lumbricus rubellus* and *Aporrectodea caliginosa*) for instance, enhanced total C efflux. The interaction between earthworms and soil microbial processes has important implication for soil C turn over and in turn, C sequestration (Zhang and Hendrix, 1995). Overcoming soil organic matter decline through C sequestration therefore, is important way to sustain agro-ecosystem in the tropics. Shift from high to lower soil disturbance such as conventional tillage to no-tillage, often promotes the accumulation of otherwise labile soil organic carbon that is less available

to microbial attack, controls carbon decomposition rates and increases total microbial biomass (Paustian et al., 1997; Six et al., 2006). Assessment of the impact of long-term no-tillage on soil carbon sequestration conducted in the eastern United States showed that no-tillage increased soil organic carbon in the upper layer of soil, but it did not store soil organic carbon more than conventional tillage for the whole soil profile (Blanco-Canqui and Lal, 2008). Similar to this study, after 20 years of study in Texas, it was showed that soil organic carbon storage under no-tillage for all cropping sequence at 0-5 cm depth was 64% greater than conventional tillage, but at 5-15 cm, organic carbon storage was only 28% higher (Wright and Hons, 2004).

The beneficial effect of long-term no-tillage over conventional tillage has been published mostly in temperate regions, but very few published in tropical region such as Indonesia. The objective of this experiment was to determine the effect of 14th and 21st year of consecutive no-tillage corn-legume rotation on earthworm population and carbon sequestration in a Typic Fragiudults in Sumatra, Indonesia.

Materials and Method

Site characteristics

A consecutive twenty one years of no-tillage experiment (37 crop seasons) was initiated in February 1987, at the experiment farm of *Politeknik Negeri Lampung*, Sumatra, Indonesia (105°13'45.5"- 105°13'48.0"E, 05°21'19.6"- 05°21'19.7"S; 122 m elevation). The soil is a Typic Fragiudult with slope ranging from 6 to 9%. Soil particle sizes composition in the 0-20 cm layer before this long-term experiment (1987) was 160, 320 and 520 g kg⁻¹ of sand, silt and clay, respectively; while bulk density 0.90 Mg m⁻³ and total porosity 65.7%. The other initial soil chemical characteristics are presented on Table 1. The climate of the site is humid tropical with average rainfall of 1800 to 2250 mm yr⁻¹ for 20 years. The site of experiment was previously a *ladang* (a local land rotation with period of fallow), which was abandoned for more than four years and covered by *alang-alang* (*Imperata cylindrica*) grass. Before the 21st of this experiment, the plot was left fallow for a year and weeds were still dominated by *alang-alang* (*Imperata cylindrica*) with average dry matter weight 13.3 Mg ha⁻¹.

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Method and analysis

The experiment was a factorial, randomized complete block design, with 4 replications. Plot size was four by six meters. Tillage treatments were conservation tillage (no-tillage, NT and minimum tillage, MT), and conventional tillage (CT); while nitrogen fertilization rates were 0, 100 and 200 kg N ha⁻¹. In NT, weed dominated by *Imperata cylindrica* was sprayed with glyphosate of 4.8 L a.i. per hectare, and the deadly weed covered the soil surface as mulch; in MT, it was the same as NT except soil surface which was slightly tilled; while in CT before plowing *Imperata cylindrica* was removed and the soil was plowed at depth 0-20 cm. Cropping pattern of the long-term experiment was cereal (corn or upland rice)-legume (soybean or cowpea)-fallow rotation (Utomo, et al., 1989). For cereal crop, hybrid corn (*Zea mays* L.) was planted each year at spacing of 75 X 25 cm. Nitrogen fertilizer treatment was not applied when legume was planted. Due to the upper layer soil compaction, all plots of conservation tillage were plowed in 1997 and 2002. After 14 years of cropping, the soil became acid (Table 1); therefore, in 2003 all plots were limed with 4 Mg ha⁻¹ of CaCO₃.

To determine the effect of long-term no-tillage and N fertilization on earthworm, the monolith samples with size of 25 by 25 cm² were collected at depth of

0-10 cm in 2001 and 2008 prior to flowering time; and earthworm was counted by hand sorting. Due to technical problem, however, plots treated with N rate of 100 kg N ha⁻¹ were not sampled. Soil samples for microbial biomass carbon and soil carbon and other chemical characteristics were taken at depth of 0-20 cm within rhizosphere and outside of rhizosphere at harvest time in May 2008. Microbial C content of the incubated soil was determined using the chloroform fumigation-incubation method. Soil organic C was analyzed with Walkley and Black method, soil organic N with Kjeldahl method, and pH with pH meter.

The analysis of variance and means test (LSD 0.05) were run using the statistics package of SAS (Statistical Analysis System).

Results and Discussion

Earthworm population, biomass and cast weight

Earthworms can have significant impacts on soil properties through their feeding, casting and burrowing activity. Their activities are influenced very much by soil management that affects their habitat. With crop residues as mulch, conservation tillage will create conducive habitat to attract earthworm (Kladivko, 1993; Lal, 1989).

Based on analysis of variance, the only treatment

Table 1. Soil pH, organic C and total N at depth of 0-20 cm prior to experiment after long-term (14 years) continuous tillage systems and N fertilization (2001)

Tillage	Treatments N Fertilizer	pH		Organic C (g kg ⁻¹)	Organic N (g kg ⁻¹)
		H ₂ O	KCl		
1987 (initial)	-----	6.8	5.8	16	2.0
2001 (prior to experiment)					
Conventional tillage	0 kg N ha ⁻¹	5.0	4.6	15.2	0.9
	100 kg N ha ⁻¹	4.8	4.3	15.2	1.1
	200 kg N ha ⁻¹	4.7	4.6	16.8	0.8
Minimum tillage	0 kg N ha ⁻¹	5.2	4.2	17.8	1.5
	100 kg N ha ⁻¹	5.0	4.7	17.6	0.9
	200 kg N ha ⁻¹	4.8	4.5	16.8	1.3
No-tillage	0 kg N ha ⁻¹	5.1	4.7	15.2	0.9
	100 kg N ha ⁻¹	5.0	4.6	16.5	0.9
	200 kg N ha ⁻¹	4.7	4.4	16.1	1.0

that affected ($p < 0.05$) earthworm population both in 14th and 21st years was tillage treatment. After 14 years of cropping (2001), NT had earthworms averaged of 101 m⁻², 46% and 39% higher than CT and MT, respectively. After 21 years of cropping, however, the response showed a little change; NT had earthworm averaged of 99 m⁻², 251% higher than CT, but 40% lower than MT. It turned out that in 14th year of cropping, NT had the highest earthworm population among other tillage system, but in 21st year of cropping NT had similar effect with either MT or CT (Table 2). Similar effects were shown in long-term (25 years) dry-land tillage practices on earthworms in southern Alberta, Canada (Claperton *et al.*, 1997), and in 30 years experiment of tillage system in the Pacific Northwest, USA (Wuest, 2001). The lower of earthworm population in CT was associated with undesirable changes in the soil environmental conditions resulting from excessive tillage (Chan, 2001). In this experiment, all plant residues in CT treatment were removed and soil was plowed; while in conservation tillage all plant residues remained on the soil surface as mulch. The present organic plant residues with respect of conservation tillage (NT and MT) had significant effect on the population of earthworm (Kladivko, 1993; Chan, 2001; Brito-Vega, 2009). Tillage system such as conservation tillage which leaves surface residue is one of the important ways to influence the earthworm population. The presence of plant residues will create better microclimate that can enhance earthworm activity (Lavelle, 1984; Brito-

Vega, *et al.*, 2009). Conservation tillage has higher earthworm populations than conducting moldboard system, due to increased food supply and mulch protection. With residues on surface, the food supply is available to the earthworms for a longer time than residues removed (Kladivko, 1993). Previous plant residue weight which was used as mulch for conservation tillage (NT and CT) in this experiment averaged of 13.3 Mg ha⁻¹; created better habitat for the earthworm. Nutrient contents of crop residues were N 10.1 g kg⁻¹, P 1.1 g kg⁻¹, K 2.5 g kg⁻¹, Ca 3.8 g kg⁻¹, Mg 1.4 g kg⁻¹, C 322.6 g kg⁻¹ and C/N 31.9. Under conservation tillage systems, earthworms can potentially play an important role than under conventional tillage in the functioning of the farming systems because of their abilities to modify soil physical environment and nutrient cycling. The earthworms genus identified in this experiment were *Pheretima* and *Lumbricus*.

Contrary to earthworm population, although earthworm biomass and cast weight under NT were relatively higher than other treatments, it seems that both earthworm biomass and cast weight were not significantly ($p < 0.05$) affected by any treatment both in 14th (2001) and in 21st (2008) long-term experiments. The relatively higher soil strength due to long-term conservation tillage at 0-10 cm soil depth tended to decrease the size of earthworms with respect to NT and MT, resulted in the decreased of their biomass. Less responses of N fertilization to earthworm biomass and cast weight were also shown in this experiment

Table 2. Effect of long-term (14 and 21 years) continuous tillage systems and N fertilization on earthworm population, biomass and cast weight at soil depth 0-10 cm

Treatments	Population		Biomass		Cast weight	
	2001*	2008	2001*	2008	2001*	2008
	(indiv.m ⁻²)		(g m ⁻²)		(g m ⁻²)	
Conventional tillage	69.33 b	28.40 b	6.16 a	0.16 a	7.13 a	12.57 a
Minimum tillage	72.89 b	167.15 a	6.11 a	6.69 a	5.34 a	105.36 a
No-tillage	101.33 a	99.56 ab	6.97 a	1.53 a	7.77 a	121.81 a
0 kg N ha ⁻¹	78.22 a	119.12 a	6.86 a	2.23 a	5.28 a	40.09 a
100 kg N ha ⁻¹	85.33 a	---	6.56 a	---	5.09 a	---
200 kg N ha ⁻¹	83.55 a	77.62 a	5.83 a	3.36 a	9.86 a	119.73 a

* In 2001, all plots of conservation tillage were plowed, and N treatments were not applied. Values within a column followed by the same letter are not significantly different at 0.05 levels

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(Table 2), but there were no negative effects of N fertilizer to the earthworm's activities.

Carbon sequestration

As labile component of soil organic C, soil microorganisms can contribute to microbial biomass C (Wang, et al., 2001), and in turn contribute to C sequestration. Carbon sequestration was measured as soil microbial biomass C and soil organic C both in rhizosphere and non-rhizosphere at depth 0-20 cm of the 21st year of cropping (at harvesting time in May 2008).

Based on analysis of variance it was showed that microbial biomass C both in rhizosphere and non-rhizosphere was significantly ($p<0.05$) affected either by tillage system or by N fertilization, but not affected by their interactions. It turned out that both in rhizosphere and in non-rhizospher, MT had microbial biomass C higher than other tillage system, while NT tended to have soil microbial biomass higher than CT, but in fact its effect is statistically the same as CT.

Generally, conservation tillage with high surface residue results in higher C sequestration than CT. Every season, all previous plant residues in conservation tillage were used as mulch covering the soil surface. Previous plant residue weight which was used as mulch for conservation tillage (NT and MT) in the 21st year experiment averaged of 13.3 Mg ha⁻¹, while in CT there was no surface mulch. Additions of plant residues on the surface and less soil disturbance will increase soil organic C particularly in upper layer of

the soil. Conversely, with no mulch on the surface and because of soil disturbance, conventional tillage will decrease soil organic C due to erosion and decomposition. Loss of soil organic C caused by tillage has been estimated to be 16 to 77% (Mann, 1986). Nitrogen fertilization with medium rate (100 kg N ha⁻¹) had the highest ($p<0.05$) microbial biomass C both in rhizosphere and in non-rhizosphere, while N fertilization with the highest rate (200 kg N ha⁻¹) had the lowest microbial biomass C.

Different from microbial biomass C, the only treatment that affected ($p<0.05$) soil organic C in rhizospher was tillage system. Minimum tillage had the highest soil organic C, but it was not different from NT (Table 3). It seems that soil microbial biomass C and organic C in rhizosphere are consistently higher than those in non rhizosphere. This is related to the higher root exudates as substrate for microorganism activity resulting in more microbial biomass (Wang, et al., 2001), and higher soil organic matter with respect to rhizosphere. In contrast, soil organic C in non-rhizosphere at depth of 0-20 cm was not significantly ($p<0.05$) affected by any treatment, even by tillage system. This was attributable to a dilution effect of sampling depth on soil organic carbon. In fact, there is a mark stratification of soil organic matter with soil under long-term NT (Blevins et al., 1984; Unger, 1991). After 20 years of study in Texas it was showed that soil organic carbon storage under no-tillage for all cropping sequence at 0-5 cm depth was 64% greater than conventional tillage, but at 5-15 cm,

Table 3. Effect of long-term (21 years) continuous tillage systems and N fertilization on soil microbial biomass C and soil organic C at soil depth 0-20 cm (2008)

Treatments	Soil Microbial Biomass C		Soil Organic C	
	Rhizosphere	Non-Rhizosphere	Rhizosphere	Non-Rhizosphere
	(mg C-CO ₂ kg ⁻¹ day ⁻¹)		(g C kg ⁻¹)	
Conventional tillage	156.91 b	48.78 b	14.6 b	12.6 a
Minimum tillage	227.64 a	74.80 a	16.5 a	13.8 a
No-tillage	182.11 b	56.10 b	15.1 ab	13.3 a
0 kg N ha ⁻¹	177.24 b	62.61 b	15.7 a	13.2 a
100 kg N ha ⁻¹	277.23 a	75.61 a	15.9 a	14.2 a
200 kg N ha ⁻¹	112.20 c	41.46 c	14.6 a	12.2 a

Values within a column followed by the same letter are not significantly different at 0.05 levels

organic carbon storage was only 28% higher (Wright and Hons, 2004). Clap *et al.* (2000) also reported that a 14% increase was observed in soil organic in the top 15 cm of NT soil, but it decreased in the 15-30 cm depth.

Soil Organic N and pH

Soil N and pH are among factors that can influence earthworm and soil C dynamic (Kladivko, 1993; Brito-Vega, *et al.*, 2009). Soil organic N and pH were measured at depth 0-20 cm in the 21st years of cropping as those of microbial biomass C were done. Soil organic N and soil pH both in rhizosphere and non-rhizosphere were significantly ($p < 0.05$) affected by tillage system or by N fertilization treatment; except for soil organic N in non-rhizosphere that was only affected by N fertilization (Table 4). It turned out that in rhizosphere MT system had soil organic N higher than CT, but had no difference from NT. Similar responses of soil organic N to tillage system were reported by Zibilske *et al.* (2002), and Brito-Vega, *et al.* (2009). Significant effect of the treatment is attributable to mineralization N from plant residues which returned it back each year. In this experiment, decomposition rate of plant residue under conservation tillage is 65% season⁻¹ (Utomo, *et al.*, 1989), a potential released of N from plant residues under MT and NT system is about 87 kg N ha⁻¹yr⁻¹.

Nitrogen fertilization with rate 200 kg N ha⁻¹ had the highest soil organic N both in rhizosphere and

in non-rhizosphere, while no N fertilization had the lowest soil organic N (Table 3). It means, 200 kg N ha⁻¹ had residual N effect higher than with no N fertilization. Referring to initial soil organic N content in 1987 (Table 1), both conservation tillage and higher N rate can retain more soil organic N from depletion than other treatments. Long-term loss reductions and greater retention of N fertilizer due to N immobilization in conservation tillage may improve crop N use efficiency by subsequent re-mineralization of the N in better synchrony with crop need (Utomo, *et al.*, 1989; Zibilske *et al.*, 2002).

Different from soil organic N response, CT had the highest ($p < 0.05$) pH both in rhizosphere and in non-rhizosphere, while NT had the lowest pH but similar to MT; and 200 kg N ha⁻¹ had the lowest pH than other N rate. It indicates that, either conservation tillage or N fertilization has acidifying effect higher than CT or with no N fertilization. In long-term crop production, however, any tillage has similar acidifying effect. The surface acidifying effect of long-term no-tillage, especially when it is combined with high N rates, is consistent with data in temperate region reported by Thomas and Frye (1984). Data on Table 4 show that after 14 years of cropping, pH of soil reduced from 6.2 (initial pH in 1987) to 4.7 (after 14 years of cropping) in any tillage with 200 kg N ha⁻¹, while any combination with 0 kg N ha⁻¹ reduced to pH around 5.0. This is the reason why in 2004 all plots were limed with 4 Mg CaCO₃ ha⁻¹.

Table 4. Effect of long-term (21 years) continuous tillage systems and N fertilization on soil organic N and soil pH at soil depth 0-20 cm (2008)

Treatments	Soil Organic N		Soil pH	
	Rhizosphere	Non-Rhizosphere	Rhizosphere	Non-Rhizosphere
	(g kg ⁻¹)			
Conventional tillage	1.5 b	1.4 a	6.4 a	6.5 a
Minimum tillage	1.8 a	1.6 a	6.2 ab	6.3 b
No-tillage	1.6 ab	1.5 a	6.1 b	6.2 b
0 kg N ha ⁻¹	1.4 b	1.3 b	6.4 a	6.5 a
100 kg N ha ⁻¹	1.6 ab	1.5 ab	6.2 a	6.4 a
200 kg N ha ⁻¹	1.8 a	1.7 a	6.0 b	6.1 b

Values within a column followed by the same letter are not significantly different at 0.05 levels

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Conclusion

After 14 years of cropping (2001), NT had earthworms averaged of 101 m⁻², 46% and 39% higher ($p < 0.05$) than CT and MT, respectively. In 21st year of cropping, however, the response showed a little change; NT had earthworm averaged of 99 m⁻², 251% higher than CT, but 40% lower ($p < 0.05$) than MT. The earthworm genus identified was *Pheretima* and *Lumbricus*.

In rhizosphere samples, MT had microbial biomass 228 mg C-CO₂ kg⁻¹ day⁻¹, or 45.5 % higher ($p < 0.05$) than NT and 70.7% higher than CT; while 100 kg N ha⁻¹ N treatment had 227 mg C-CO₂ kg⁻¹ day⁻¹, 28% and 103% higher than 0 and 200 kg N ha⁻¹ treatments, respectively.

After 21 years of cropping, soil organic C of MT in rhizosphere at 0-20 cm soil depth was 16.5 g kg⁻¹, or 9.3% and 13.0% ($p < 0.05$) higher than NT and CT, respectively. At depth of 0-20 cm in non-rhizosphere, there was less response of soil organic C to NT.

Long-term continuous no-tillage corn-legume rotation in Indonesia has potentially to increase earthworm activity and to sequester C better than conventional tillage.

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