

Full Length Research Paper

Greenhouse assessment of microbial biomass carbon and nitrogen as influenced by compaction, Bradyrhizobium inoculation and nitrogen fertilizer application in maize-soybean cropping systems

Omeke J. O.^{1*}, Yusuf A. A.², Yakubu A. A¹ and Umar F. G.

¹National Agricultural Extension and Research Liaison Services, Ahmadu Bello University, P. M. B. 1067 Zaria, Nigeria.

²Department of Soil Science, Faculty Agriculture/Institute for Agricultural Research, Ahmadu Bello University, P. M. B. 1044 Zaria, Nigeria.

Receive 11 February, 2019; Accepted 25 March, 2019

Soil microbial biomass (SMB) is the main driving force in nutrient cycling and good indicator of soil productivity. A greenhouse experiment was designed to assess the effect of soil compaction, cropping system [sole maize, rotation 1 (inoculated soybean-maize), rotation 2 (un-inoculated soybean-maize) and intercrop 1 (inoculated soybean-maize) and intercrop 2 (un-inoculated soybean-maize)] and nitrogen fertilizer on soil microbial biomass C (SMB-C) and N (SMB-N) and their proportion to soil organic C and total N. SMB-C and SMB-N were higher in un-compacted than compacted soils with percent differences of 2.63 and 6.04% respectively. However, they were 19.32 and 36.36% lower in sole maize compared to rotation 1, 7.83 and 15.36% for rotation 2, 22.19 and 20.06% for intercrop 1 and 14.62 and 12.54% for intercrop 2. The results also showed that the application of 120 kg N ha⁻¹ produced the highest soil microbial biomass as a percent of soil organic carbon, followed by 80 kg N ha⁻¹, while the least value was obtained under zero application of nitrogen. Microbial biomass carbon and nitrogen as a percent of soil total nitrogen was significantly higher up to 80 kg N ha⁻¹ before it decline at 120 kg N ha⁻¹ suggesting better soil productivity improvement at 80 kg N ha⁻¹ under the cropping systems with inoculated soybean. The findings indicate the need for inoculation in soybean-maize cropping systems to improve soil microbial biomass especially under less soil disturbances.

Key words: Compaction, Bradyrhizobium Inoculation, Nitrogen fertilizer, microbial biomass, greenhouse.

INTRODUCTION

The predominant agricultural trends in the past 50 years have been intensive production with increased use of commercial seeds, fertilizers, pesticides, fuel (Tomich et al., 2011) and land use intensification. Consequences such as increased erosion, decreased soil fertility and biodiversity, water pollution and eutrophication, and

alteration of atmospheric and climate processes lead to the urgency to develop new strategies that use the ecological interactions within the agricultural ecosystem (Matson et al., 1997). Soil microbial communities are extremely diverse and the relation between their diversity and function influences soil stability, productivity and

*Corresponding author. E-mail: obia.joe@gmail.com.

resilience; on the other hand, organic matter, water activity, soil fertility, physical and chemical properties influence microbial biomass in soils (Tomich et al., 2011). Because soil biota is influenced by land use and management techniques, changing management practices could have significant effects on the soil microbial properties and processes (Stark et al., 2007).

In recent years, there has been a rapid expansion of mechanized agriculture from land to harvest operation and this has resulted in substantial increases in soil compaction (Balbuena et al., 2000; Startsev and McNabb, 2000; Dias Junior et al., 2008; Silva et al., 2008). Given the short rotation period, fallowing and high frequency of operations it can undermine the productivity of the stands in a near future, across agro-ecological zones. The negative effects of soil compaction on plant growth have been attributed primarily to the restriction on root growth (Sérgio et al., 2011). However, there is some evidence that soil compaction also alters the size, diversity and activity of the microbial community. As a result, there occur changes, for example, in nutrient cycling patterns and their availability to plants (Lee et al., 1996; Sérgio et al., 2011). It has been shown that soil compaction plays an important role in microbial activity since the increase in soil density leads to altered pore size and distribution, lower O₂ and CO₂ diffusion rates and greater abundance of anaerobic micro sites and consequent reduction in the aerobic microbial activity (Jensen et al., 1996a; Tan et al., 2005). These adverse effects of soil compaction on microbial activity seem to result mainly from losses in bio-pores and other macro-pores connectivity (Whalley et al., 1995). Low O₂ concentration (< 2-5%) (Sérgio et al., 2011) and low macro-porosity (< 10%) (Linn and Doran, 1984) cause a reduction in the aerobic microbial activity, and may favor N losses by denitrification (Breland and Hansen, 1996; Jensen et al., 1996a; Ruser et al., 2006) and subsequently lower the productivity of the soil. Accordingly, soil respiration (CO₂ production) is a useful indicator of soil organic matter (SOM) decomposition (Hassink, 1994; Lee et al., 1996) by both, aerobic and anaerobic microbes, which is a clear advantage over techniques based on O₂ uptake (Sergio et al., 2011).

Despite the fact that the soil compaction may negatively affect the cycling of C and N by modifying soil aeration and/or, microbial community structure, there have been a few studies that have dealt with such subject (Torbert and Wood, 1992; Jensen et al., 1996b; Tan et al., 2005). Under field conditions, the microbial biomass carbon (MB-C) in the 10-20 cm soil layer under the tractor track was reduced by 38% by soil compaction, in comparison to the control soil (Dick et al., 1988). In fact, the authors found a significant negative correlation between MB-C and soil density. The changes in microbial activity and denitrification rates in soils as affected by variations in pore space and moisture levels have been extensively researched (Craswell and Martin, 1974;

Myers et al., 1982; Linn and Doran, 1984; Rodrigo et al., 1997; Franzluebbers, 1999; Ruser et al., 2006). Nonetheless, few studies have examined the effects of alterations in soil physical properties on N transformation (Torbert and Wood, 1992; Tan et al., 2005; Ruser et al., 2006). Particularly, there is a lack of information about the consequences of soil compaction on N transformation. Although it seems reasonable to hypothesize that the effects of soil compaction on the microbial community are strong, the few available results indicate the opposite. Linn and Doran (1984) carried out a study under laboratory conditions and found that microbial activity decreased only slightly under soil compaction. Jensen et al. (1996b) observed that no indicator of microbial biomass was significantly affected when total soil porosity was reduced from 0.60 to 0.51 m³ m⁻³ after 21 days of incubation. These findings were attributed to the fact that compaction only altered the larger pores and possibly had no substantial effect on the access of soil microbes to smaller diameter pores. Under intense heavy machinery traffic soil compaction may be severe resulting to changes in soil biomass. Dias Junior et al. (2008) and Silva et al. (2008) found that, depending on the weight and the number of passes of a loaded forwarder, the soil density may increase to values as high as 32%. Thus, it is likely that not only the soil macro-pores, but also the micro-pores are affected by mechanized operations in forest stands so that their impact on soil microbial activity and C and N cycling may be more pronounced than previously thought. The recent findings that a major portion of soil organic matter is stabilized in the entrance of small pores in soils (Kaiser and Guggenberger, 2006) supports this hypothesis.

Thus, intensive agriculture systems with high inorganic fertilizer inputs, however, limit the return of crop residues to the soil (Roper and Ladha, 1995). Maximizing nitrogen content in plant not only entails maximizing the application of organic matter or chemical fertilizer, but also requires that recovery of N is optimized (Yadvinder-Singh, 2010). Nitrogen transformations are occurring during breakdown of soil microorganisms, which are influenced by amount and types of residue, soil physical and chemical properties (Peoples et al., 1995). Studies of MB may be valuable as an indicator for research on the effects of integrated management on soil productivity (Omeke, 2016). This study was carried out to assess, under greenhouse condition, the effects of soil compaction, Brandy rhizobium inoculation in maize-soybean-based cropping systems and nitrogen fertilizer application on MB-C and MB-N in savanna Alfisol of Nigeria.

MATERIALS AND METHODS

Location and soil preparation

Greenhouse experiment was carried out at the Department of Soil

Science, Faculty of Agriculture, Ahmadu Bello University, Samaru, Nigeria. The Greenhouse is located within longitudes 11° 11' N and latitudes 007° 37' E. The bulk soils used for the experiments were taken from 6 points at 0-15 cm depth (top soil) using spade from the field where soybean trial was not conducted over 5 years. The samples were bulked, air-dried and sieved with a 5 mm sieve in readiness for compaction process (Omeke, 2016)

Treatment and experimental design

The treatments consisted of two levels of compaction (compacted and un-compacted soils) as main plot. A total of 120 pots (PVC) with diameter of 15 and 52 cm height were filled with prepared bulked soil of about 15% moisture content to a predetermined depth (50 cm) for compaction process. Approximately 8.1 kg of soil at 15% water content was placed in PVC cylindrical pots, and the bulk density was 1.31 Mg m⁻³ in the non-compacted pots. The compacted pot was prepared by packing soil in a Proctor hammer (5 cm that is, 30 cm drop height, 10.4 kg weight) in three layers by giving a sufficient number of blows using a flat-bottom hammer to reach the target bulk density (approximately 1.50 Mg m⁻³). Soil bulk densities were determined by the core method (Blake and Hartge, 1986). A handy penetrometer gauge was used to obtain uniform penetration force of 2.5 kPa in all the compacted pots. The two bulk densities used for the un-compacted and compacted soils were based on established bulk density range of 1.39 to 1.50 Mg m⁻³ for soils of Samaru, Northern Guinea savannah of Nigeria (Oikeh et al., 1998). Continuous maize, maize-inoculated soybean rotation, maize-un-inoculated soybean rotation, maize-inoculated soybean intercrop and maize-uni-noculated soybean intercrop and N fertilizer rates of 0, 40, 80 and 120 kg N ha⁻¹ were sub plots. Pots were arranged on a bench in the greenhouse according to a randomized complete block design with three blocks (each block contained all 15 treatments x 2 soil compaction x 4 Bradyrhizobium inoculation and nitrogen application rates x 3 replicates).

Inoculation and planting

Two sets of experiments were conducted and terminated at eight weeks respectively. In the first experiment, soybean seeds were divided into two, one part was inoculated with commercial rhizobia inoculants, Legume-fix at 400 g ha⁻¹ (inoculated soybean) while remaining seeds were not inoculated (un-inoculated soybean) as shown in Figure 1. Before inoculation the soybean seeds were sterilized as reported by Vincent (1970). The peat-based inoculants (Legume-Fix) were applied directly on the seed with some quantity of liquid gum Arabic as adhesive to coat the seeds according to user guide produced by the company. The inoculated seeds were allowed to stay for about 10-15 min under shade to avoid effect of direct sun light and for air-drying before planting. Both maize and soybean seeds were sown on the same day in all the pots but un-inoculated soybean pots were sown first to avoid contamination. Four seeds were sown per crops in all the pots and thinned to two per stand (1:1 per stand for intercrop, 2:0 per stand for rotation and 2 per stand for continuous maize) at 10 days after sowing (DAS). The first set of greenhouse experiment was established mainly to create enabling soil environment to carry out effect of the following Bradyrhizobium inoculation in maize-soybean cropping systems; continuous maize, maize-inoculated soybean rotation, maize-un-inoculated soybean rotation, maize-inoculated soybean intercrop and maize-un-inoculated soybean intercrop on MB-C and MB-N in second set of greenhouse. The first set of the experiment was terminated at 8 weeks after sowing (WAS). The crops (maize and soybean) were harvested by cutting of the shoot system at their base without causing any disturbance to the soil and root system within the pots. The second set of the experiment commenced at 2

weeks after first experiment was terminated (harvested). In second set experiment, all the 120 pots both compacted (60 pots) and un-compacted (60 pots) soils were sown only with 6 seeds of maize without soybean and thinned to 2 per stands at 10 days after sowing. The experiment was also terminated at 8 weeks after sowing.

Fertilizer application

The same fertilizer treatment was observed throughout for both the greenhouse and field experiments. P (single super phosphate; SSP) and K (Muriate of potash; MOP) fertilizers were applied to all the pots planted with maize at the rate of 60 kg P₂O₅ ha⁻¹ (1.45 mg kg⁻¹ soil) and 60 kg K₂O ha⁻¹ (0.44 mg kg⁻¹ soil) at planting, respectively. But the sole soybean used for rotation received 40 kg P₂O₅ ha⁻¹ (2.50 mg per kg soil) and 60 kg K₂O ha⁻¹ (0.75 mg kg⁻¹ soil) and no fertilizer was added to the soybean in intercrop pots. The pots containing maize in both sets of experiment received N (urea) fertilizer application at the rates of 0 (0 mg kg⁻¹ soil), 40 (3.26 mg kg⁻¹ soil), 80 (6.52 mg kg⁻¹ soil) and 120 kg N ha⁻¹ (9.78 mg kg⁻¹ soil), which was shared into two: first and second applications were at 2 and 6 WAS, respectively. Fertilizer application was done by band placement method and late in the evening in both sets of the experiment.

Soil sampling and laboratory analysis

Composite soil sample was taken from the bulk soil before pots experiment and used for initial soil analysis following standard procedures (IITA, 1989). The soil was sandy loam in texture with the following properties: pH (Water), 5.40; Corg, 5.50 g kg⁻¹; Ntot, 0.46 g kg⁻¹; available P (Bray 1-P), 9.14 mg kg⁻¹; exchangeable cations (cmolkg⁻¹) of Mg²⁺, 0.36; Ca²⁺, 0.80; K⁺, 0.15; and Na⁺, 0.28; exchangeable acidity (cmolkg⁻¹), 1.08; extractable micronutrients (mgkg⁻¹) of Cu, 3.36; Fe, 71.68; Zn, 2.94 and Mn, 96.52; Bacteria; 8.50 x 10⁶ cfug⁻¹ soil, fungi; 3.44 x 10⁶ cfug⁻¹ soil, MB-N; 16.98 mg kg⁻¹, MB-C; 188.68 mg kg⁻¹ and C_{mic} : N_{mic}; 11. Soil samplings were only taken from the second set of experiment at 8 weeks after sowing. Surface soil sampling was done per pot using hand trowel, by cleaning the surface of the pot before and after sampling. The sampled soil was bagged, labelled properly and stored in the refrigerator for laboratory analysis of microbial biomass carbon and nitrogen while the air-dried samples were crushed lightly in preparation for laboratory analysis of selected soil chemical properties. The soil microbial biomass MB-C and MB-N were estimated by the fumigation-extraction method (Brookes et al., 1985; Sparling and West, 1988), using field-fresh, moist 2mm sieved soil sample. The extractable MB-C and MB-N in both fumigated and unfumigated samples were determined. MB-C was estimated by multiplying the difference in extractable C of fumigated and unfumigated samples, using a conversion factor of 2.64 (Vance et al., 1987) whereas MC- N was calculated by multiplying the difference in extractable N of fumigated and unfumigated sample using a conversion factor of 1.46 (Brookes et al., 1985). The soil nitrogen was determined by micro-Kjeldahl digestion method, as described by Bremner and Mulvaney (1982). Organic carbon was measured using the method described by Nelson and Sommers (1982).

Statistical analysis

Data collected were subjected to analysis of variance (ANOVA) using the mixed linear model procedure of SAS, Institute Inc., (2009). Duncan's multiple range test procedures was used when the F-calculated of the ANOVA for each variable was found to be



Figure 1. Soybean seeds before inoculation (left) and inoculated with *Bradyrhizobia* (right).

significant and their interactions were compared by computing least square means and standard errors of difference (SED) at 5 % level of probability.

RESULTS

Effect of soil compaction on biomass carbon (MB-C) and nitrogen (MB-N)

The results in Table 1 show that soil compaction had significant effects only on soil microbial biomass carbon (MB-C) ($P < 0.05$). Compared to compaction soils, MB-C, MB-N and C_{mic}/N_{mic} ratio were consistently higher in uncompacted soils, with percent differences of 2.63% for MB-N, 6.04% for MB-C and 14.29% for C_{mic}/N_{mic} ratio. Generally, the results indicated that the initial values of MB-C, MB-N and C_{mic}/N_{mic} ratio obtained before setting up the greenhouse experiment were higher than those values obtained after the greenhouse experiment except that of MB-N in both soil compaction. A significant interaction in MB-C was observed between soil compaction and cropping systems (Figure 2). Moreover, inoculated soybean-maize rotation with uncompacted soil had higher value of MB-C, compared to other soil compaction and cropping systems combinations.

Effect of bradyrhizobium inoculation on biomass carbon (MB-C) and nitrogen (MB-N)

Also, cropping systems significantly ($P < 0.05$) influenced MB-C and MB-N in the soil (Table 1). Pots with inoculated soybean-maize rotation had higher values of MB-C and MB-N, followed by inoculated soybean-maize intercrop; while the least values were found in pots with continuous sole maize. Value of C_{mic}/N_{mic} ratio was significantly lower under continuous sole maize as compared to other cropping systems. A significant interaction was obtained

between soil compaction and cropping systems on MB-N (Figure 2). Moreover, inoculated soybean-maize rotation with uncompacted soil combination had higher value of MB-C, compared to other soil compaction and cropping systems combinations. The data also show that MB-C value was significantly lower in pots treated with maize/soybean uninoculated and N fertilizer application as compared to those with bradyrhizobium inoculated soybean and nitrogen fertilizer application which tend to increase with increased N fertilizer application to the peak of 80 kg ha^{-1} and decrease (Figure 3). Similar trends obtained in Figure 3 are observed in Figure 4, which indicated significantly higher value of MB-C at 80 kg ha^{-1} under both bradyrhizobium inoculated and un-inoculated soybean/maize cropping systems with N fertilizer application treatments combinations.

Nitrogen fertilizer application on biomass carbon (MB-C) and nitrogen (MB-N)

The effects of N fertilizer on MB-N, MB-C and C_{mic}/N_{mic} ratio were significantly different (Table 1). The values were generally lower in pots without N fertilizer (except for C_{mic}/N_{mic} ratio) than for other levels of N fertilizer application. The results further showed initial increase and decrease with increment in N fertilization rates in both MB-C and MB-N. However, the values of MB-C and MB-N in pots with 0 kg N ha^{-1} showed no significant difference from the result obtained in pots treated with 120 kg N ha^{-1} .

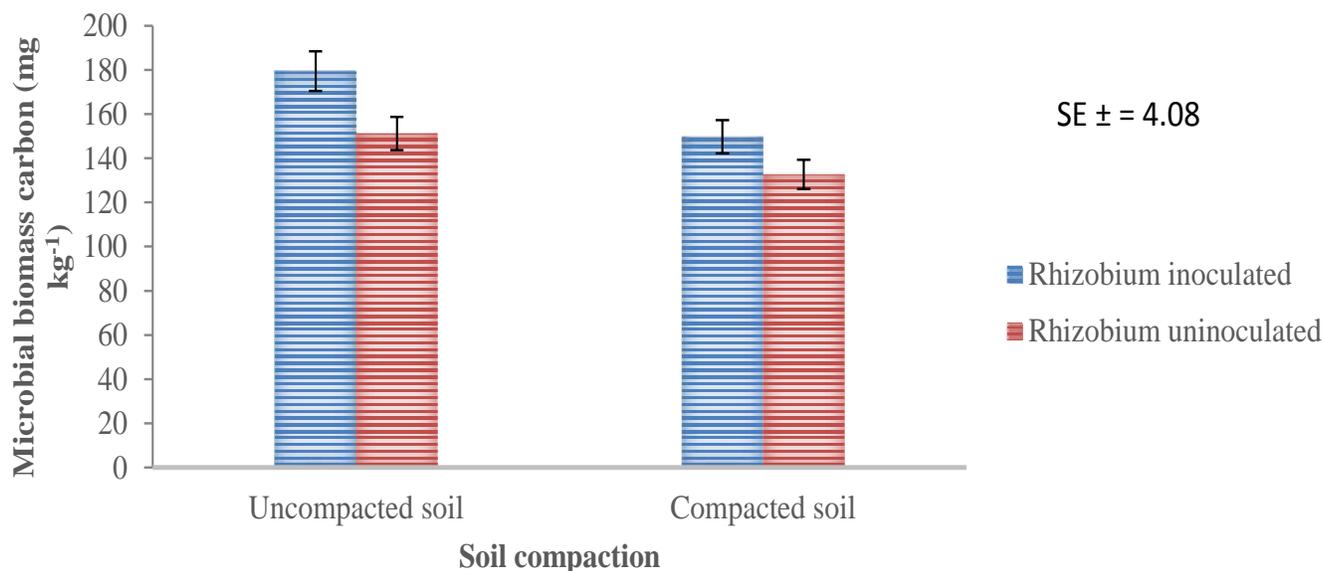
Effect of soil compaction on MB-C and MB-N as a percent of soil carbon and nitrogen

Proportion of soil organic C (%) and total N (%) as biomass C and N as influenced by soil compaction are

Table 1. Microbial biomass carbon and nitrogen in soil under greenhouse experiment.

Treatment	MB-N	MB-C	C _{mic} /N _{mic}
	(mgkg ⁻¹)	(mg kg ⁻¹)	Ratio
Soil compaction (SC)			
Compacted soil (1.50 Mg cm ⁻³)	24.46	153.96 ^b	6.29
Uncompacted soil (1.31 Mg cm ⁻³)	25.12	165.96 ^a	6.61
SE±	0.40	7.82	0.53
Cropping system (CS)			
Continuous maize	20.07 ^d	178.53 ^a	8.90 ^a
Rotation-inoculated	28.74 ^a	167.05 ^{ba}	5.81 ^b
Rotation-uninoculated	26.73 ^a	156.06 ^b	5.84 ^b
Intercrop-inoculated	25.87 ^b	155.03 ^b	5.99 ^b
intercrop-uninoculated	22.55 ^c	142.46 ^c	6.32 ^b
SE±	0.26	12.36	0.04
Nitrogen fertilizer application (NFA : kg ha⁻¹)			
0	20.55 ^c	146.35 ^b	7.12 ^a
40	27.92 ^a	164.25 ^a	5.88 ^b
80	25.25 ^b	161.20 ^a	6.34 ^b
120	23.44 ^c	156.50 ^a	6.68 ^b
SE±	0.26	12.36	0.04
Interaction			
SC*CS	NS	*	NS
SC*NFA	NS	*	NS
CS*NFA	**	NS	NS
SC*CS*NFA	NS	NS	NS

MB-C = Microbial biomass carbon, MB-N = Microbial biomass nitrogen, C_{mic}/N_{mic}= Microbial biomass carbon and Microbial biomass nitrogen ratio, NS = Not significant at P<0.05, * = Significant at P<0.05, SE = Standard error.

**Figure 2.** Interaction between soil compaction and Bradyrhizobium inoculations on microbial biomass carbon.

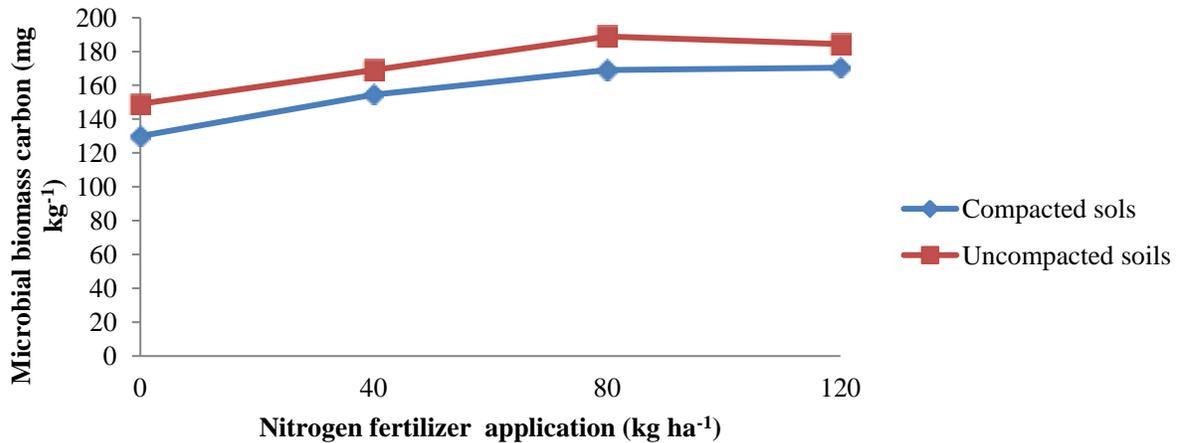


Figure 3. Interaction between nitrogen fertilizer application and soil compaction on microbial biomass carbon.

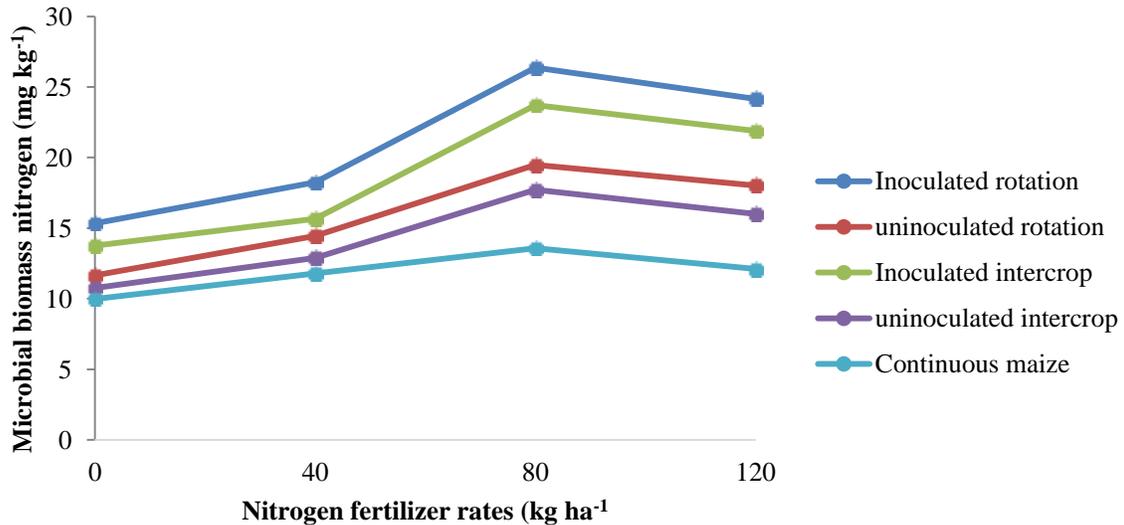


Figure 4. Interaction between nitrogen fertilizer application and Bradyrhizobium inoculation on microbial biomass nitrogen.

organic C and total N as biomass C and N (Table 2) showed a significant difference between the two soil compactions. The biomass as a percent of soil carbon and nitrogen under un-compacted soil were observed to be higher than those obtained in compacted pots with difference of 7.47 and 4.79%.

Effect of cropping system on biomass as a percent of soil carbon and nitrogen

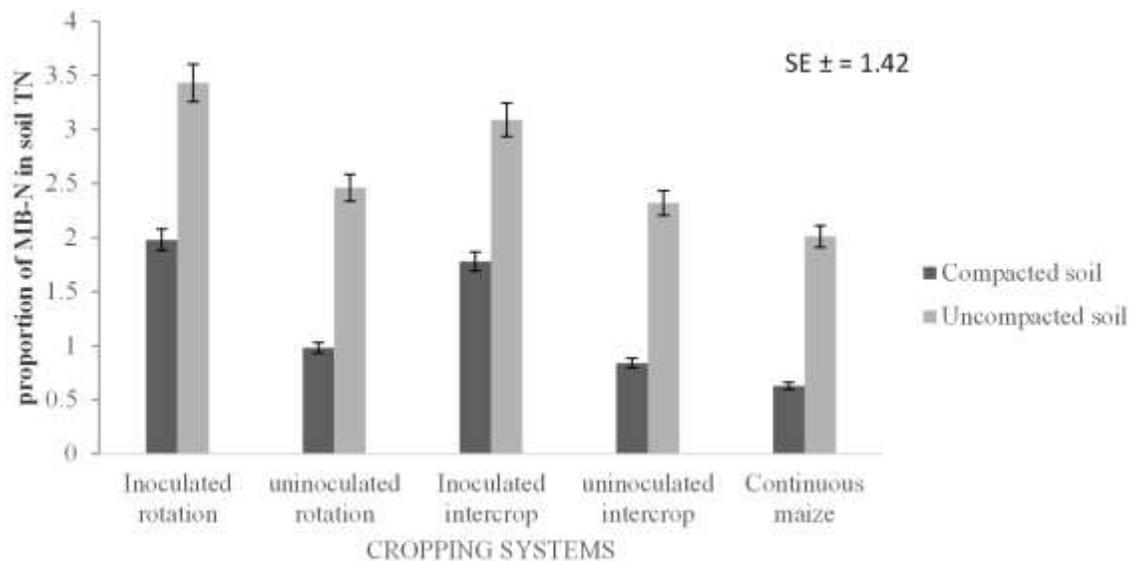
Rhizobium inoculation in maize-soybean-based cropping systems showed a significant effect ($P < 0.05$) on biomass as a percent of soil carbon and total nitrogen (Table 2). Among the rhizobium inoculation in maize-soybean-based

cropping systems, biomass as a percent of soil carbon and total nitrogen were highest in maize following maize/inoculated soybean intercrop, followed by maize following inoculated soybean rotation; it was lowest for maize following maize (continuous sole maize). Biomass as a percent of soil carbon and total nitrogen obtained under continuous sole maize was lower than others with difference of 19.32 and 36.36% for rotation inoculated, 7.83 and 15.36% for rotation un-inoculated, 22.19 and 20.06% for intercrop inoculated and 14.62 and 12.54% for intercrop un-inoculated respectively. Significant interaction was observed between soil compaction and cropping systems on proportion of MB-N in soil total nitrogen (Figure 5) which was significantly higher under un-compacted soil in combination with rhizobium

Table 2. Proportion of soil organic C and total N as biomass C and N as influenced by soil compaction, Bradyrhizobium inoculation and nitrogen fertilizer.

Treatment	Biomass as a percent of soil	
	Organic C (%)	Total N (%)
Soil compaction (SC)		
Compacted soil (1.50 Mg cm ⁻³)	4.15 ^b	3.76 ^b
Uncompacted soil (1.31 Mg cm ⁻³)	4.46 ^a	3.94 ^a
SE±	0.04	0.02
Cropping system (CS)		
Continuous maize	3.83 ^c	3.19 ^c
Rotation-inoculated	4.57 ^a	4.35 ^a
Rotation-uninoculated	4.13 ^b	3.68 ^b
Intercrop-inoculated	4.68 ^a	3.83 ^b
intercrop-uninoculated	4.39 ^b	3.59 ^b
SE±	0.26	
Nitrogen fertilizer rate (NRate: kg/ha)		
0	3.21 ^d	3.29 ^c
40	4.08 ^c	3.55 ^b
80	4.35 ^b	4.04 ^a
120	5.16 ^a	3.88 ^b
SE±	0.12	0.16
Interaction		
SC*CS	NS	*
SC*NRate	NS	NS
CS*NRate	**	**
SC*CS*NRate	NS	NS

OC = Organic carbon, TN = Total nitrogen, MBC = Microbial biomass carbon, MBN = Microbial biomass nitrogen, NS = Not significant at P<0.05, * = Significant at P<0.05, SE = Standard Error.

**Figure 5.** Interaction between nitrogen soil compaction and cropping systems on % microbial biomass carbon in soil organic carbon.

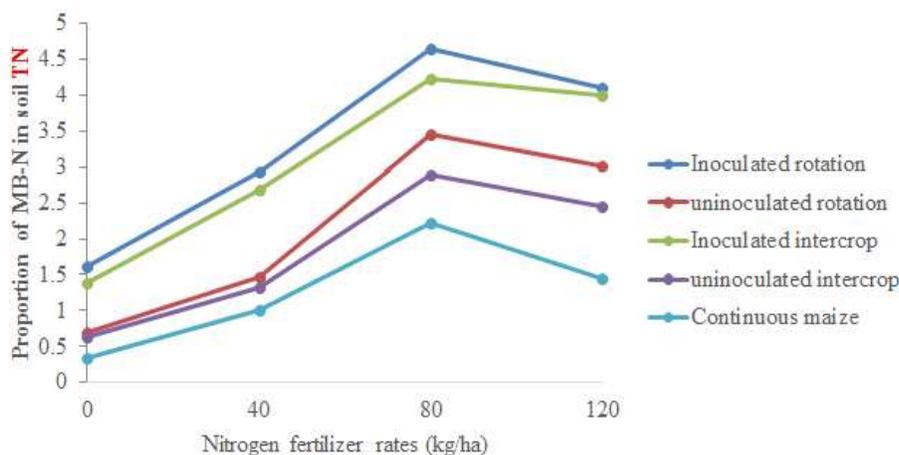


Figure 6. Interaction between nitrogen fertilizer application and cropping systems on % microbial biomass carbon in soil organic carbon.

inoculated soybean-maize rotation as compared to other combinations.

Effect of N fertilizer on biomass as a percent of soil carbon and nitrogen

Results of the effects of various N fertilizer application rates on biomass as a percent of soil carbon and total nitrogen are presented in Table 2. Comparison data on the various fertilizer treatments showed that biomass as a percent of soil carbon and total nitrogen increased significantly, as compared to that of control, which increased with additional increase in N. The application of 120 kg N ha⁻¹ produced the highest biomass as a percent of soil carbon, followed by 80 kg N ha⁻¹, while the least value was obtained under zero application of kg N ha⁻¹. Whereas, biomass as a percent of soil total nitrogen was significantly higher at 80 kg N ha⁻¹ which shows increasing trend with nitrogen application rates and decreased at 120 kg N ha⁻¹. Interaction results obtained between cropping systems and N fertilizer application rates show significantly higher values of MB-C and MB-N proportion in soil organic carbon (Figure 6) and total nitrogen contents (Figure 7) under rhizobium inoculated soybean-maize rotation at all levels of N application as compared to other cropping systems and N rates combinations. Under all the cropping systems, values of biomass of carbon and nitrogen proportion to organic carbon and total nitrogen contents of the soil increase with N rates to a peak of 80 kg N ha⁻¹ and decrease with additional N application.

DISCUSSION

Significantly, higher microbial biomass carbon (MB-C) and nitrogen (MB-N) under un-compacted soil could be

attributed to the beneficial effects of uncompacted soil on the accumulation of soil microbial biomass C and N (Spedding et al., 2004; Omeke, 2017). The current study found that compacted soil lower microbial biomass C and N as compared with uncompacted soil, due to negative effects of compaction process on soil biological activity. This suggests that compacted soil could create unfavourable soil condition for microbial activity and greater protection of soil organic matter due to the formation of small pores and poor aeration that lower microbial biomass C and N in soil. Study conducted by Omeke (2016) in the same field where soil sample was collected for the greenhouse experiment claimed that OC needed a longer period to respond to cultivation, as compared to SMB-C. This could explain the slight variation found among the soil compactions with percent difference of 6.04% for MB-C, 2.63% for MB-N, 14.29% for $C_{mic}:N_{mic}$ ratio and none significant difference observed for OC. Therefore, the soil's quick turnover rate facilitates the changes of MB-C and MB-N in the short-term (Wang et al., 2012) which may serve as a potential indicator of soil productivity. The greater average difference of $C_{mic}:C_{org}$ and $N_{mic}:N_{tot}$ ratios observed in un-compacted soil as compared with compacted soils may be related to residual effect of un-compacted soil on accumulation of organic carbon. Results obtained from cultivated soils show a higher proportion of MB-C and MB-N in soil OC and TN in un-compacted soils than compacted soils, with values ranging from 0.2-4.4 (Souza et al., 2003) are within the ranged obtained in this study. The variations in the $C_{mic}:C_{org}$ and $N_{mic}:N_{tot}$ relationship reflect the pattern of residues deposition, efficiency of the microbial C and N conversion, losses of C and N from the soil, and stabilization of organic C and N in the mineral fractions of the soil (Carter et al., 1994; Sparling, 1992) and intensity of compaction process (Omeke, 2016).

The variations of MB-C, MB-N and C_{mic}/N_{mic} observed among the various cropping systems with or without

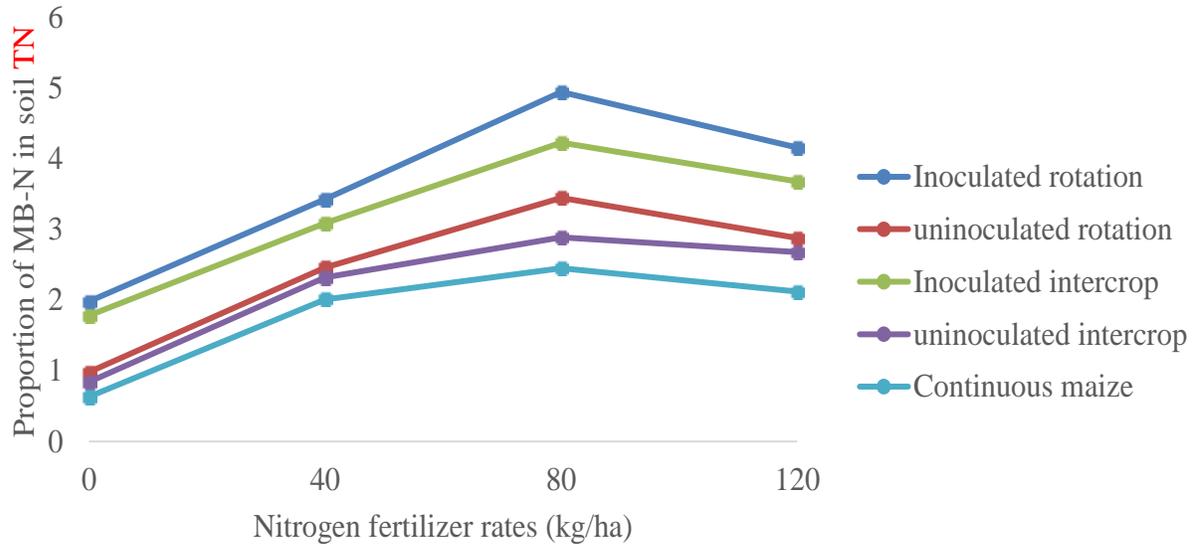


Figure 7. Interaction between nitrogen fertilizer application and cropping systems on % microbial biomass nitrogen in soil total nitrogen.

rhizobium inoculation could be ascribed to greater production of organic source through underground biomass and low C/N ratio of soybean residues, compared to continuous sole maize. Agronomic practices that favoured the accumulation of organic matter in soil increases both microbial biomass and its proportion in total soil organic matter (Adeboye, 2009). But the higher microbial biomass N observed in inoculated soybean-maize rotation than those without inoculated soybean and continuous sole maize could be attributed to greater N contribution through BNF and root N supported by lower C/N ratio of soybean root and nodule biomass. The significant interaction observed for compacted soils and cropping systems on SMBC and C_{mic}/N_{mic} ratio implies that the inclusion of inoculated soybean in the cropping systems under less soil disturbances would provide a good soil environment that facilitates microbial biomass production in soil. The high utilization of N by maize (compared to soybean) during the growth stage reduced N availability and consequently lowered MB-N in soil under continuous sole maize. For cropping systems' sustainability, soil microbial biomass is the active component of the soil organic pool, which is responsible for organic matter decomposition, affecting soil nutrient content and, consequently, primary productivity in most biogeochemical processes in soil ecosystems (Haney et al., 2001). Variability of $C_{mic}:C_{org}$ and $N_{mic}:N_{tot}$ proportion obtained among the cropping systems with or without rhizobium inoculations may indicate the differences in contribution of OC and TN to the soil (Omeke, 2017). It has been suggested that values of the proportion of $C_{mic}:C_{org}$ express a balance point and can range from 2.3 for monocultures to 4.4 for crop rotations (Carter et al., 1994) which is comparable to those reported in this

study. Larger or small values may indicate carbon or nitrogen accumulation or losses; this proportion is used as a good indicator for alterations of soil carbon and nitrogen content as a function of soil management. Therefore, measuring microbial biomass in soil would give clear understanding effects of land preparation in combination with integration of inoculated soybean in the maize-soybean-based cropping and N rates for sustainable soil productivity (Omeke, 2018).

Values of MB-C and MB-N were generally lower under control (0 kg N ha^{-1}) than other N fertilizer treatments pots, which significantly ($P < 0.05$) increase and decrease with N fertilizer rates. This suggests that a decline in N status of the soil would negatively influence microbial biomass status of the soil, and vice versa. Also, nitrogen fertilizer application contributed to more availability of N due to low N status of the soil (Table 1). Improvement of soil N enhances generation of organic carbon source from the growing and decaying of plant residues like dead root and sloughed root cap cells as well as N release from root exudates (Adeboye, 2009). Generally, N treatment at 80 kg N ha^{-1} confirmed to be best among other N treatments for soil biomass improvement, meaning that below or above this N rate (80 kg N ha^{-1}) would lower nutrients cycling for crop production. This implies that N fertilizer applied within the rhizosphere stimulated growth of gram-negative bacteria more than fungi in the soil (Omeke et al., 2016). Bacterial growth is favoured by the crop-root rhizo-deposition, rich in amino acids (Vinzke et al., 2004) and soluble sugars (Jensen, 1996) with substantial nitrogen content. Influenced by all these factors, bacteria multiply faster in population in soil under N fertilizer application and may contribute to increasing soil microbial N and C in the

rhizosphere due to their lower C/N ratio as compared to plots without N treatment (Omeke et al., 2016; Omeke, 2017).

Conclusion

Compacted soil had lower values of MB-C and MB-N as compared to un-compacted pots, with a percentage contrast of 1.83% for MB-C and 19.67% for MB-N. The values of MB-N, MB-C and their percentage in soil organic carbon and total nitrogen were significantly lower in continuous sole maize than other cropping systems with soybean; significantly higher in inoculated soybean-maize rotation. Generally, pots without nitrogen fertilizer treatments (0 kg N ha^{-1}) had significantly lower values of MB-C and MB-N proportion in soil organic carbon and total nitrogen. The results revealed that un-compacted soil in combination with maize-soybean cropping systems with rhizobium-inoculated soybean promotes microbial biomass activity as compared to other treatments combination. The study also demonstrated that integration of inoculated soybean in soybean-maize-based cropping systems in combination with nitrogen fertilizer application enhances microbial biomass status of the soil, which appears to be better at 80 kg N ha^{-1} . This suggests that those cropping systems with rhizobium inoculated soybean under minimum soil disturbance have best improved soil conditions (quality/health) for sustainable crop production, by restoring microbial carbon proportion in soil organic carbon and nitrogen content better than the other treatments combination. Therefore, findings advocate the need for inclusion of bradyrhizobium inoculation in legume-maize-based cropping systems to improve soil microbial biomass especially under less soil disturbances in combination with 80 kg N ha^{-1} rate of application.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENT

The author appreciates the support from Institute for Agricultural Research, Ahmadu Bello University Zaria, Nigeria, towards the success of this research as staff in training.

REFERENCES

- Adeboye A (2009). Microbial biomass and water soluble carbon as affected by cereal/legume rotation in a Guinea savanna Alfisol of Nigeria. *Nigeria Journal of Soil Science* 19(1):70-80.
- Balbuena H, Terminiello M, Claverie A, Casado, P, Marlats R (2000). Compactación del suelo durante la cosecha forestal. *Evolución de las*
- propiedades físicas. *Revista Brasileira de Engenharia Agrícola e Ambiental* 4:453-459.
- Breland A, Hansen S (1996). Nitrogen mineralization and microbial biomass as Affected by soil compaction. *Soil Biology and Biochemistry* 28:655-663.
- Bremner JR, Mulvaney CS (1982). Nitrogen-total In: Page A.L. (Ed). *Methods of Soil Analysis, Part 2*, ASA, Madison WI, pp. 595-624.
- Brookes PC, Landman A, Pruden G, Jenkinson DS (1985). Chloroform fumigation and the release of soil nitrogen: a rapid extraction method to measure microbial biomass nitrogen in soil. *Soil Biology and Biochemistry* 17:837-842.
- Carter MR, Angers DA, Kunelius HT (1994). Soil structural forms and stability and organic matter under cool-season perennial grass. *Soil Science Society of America Journal* 58:123-130.
- Craswell ET, Martin AE (1974). Effects of moisture content on denitrification in clay soil. *Soil Biology and Biochemistry* 6:127-129.
- Dias Junior MS, Silva SR, Santos NS, Araujo Junior CF (2008). Assessment of the soil compaction of two Ultisols caused by logging operations. *Revista Brasileira de Ciência do Solo* 32:2245-2253.
- Dick RP, Myrold DD, Kerle EA (1988). Microbial biomass and soil enzyme activities in compacted and rehabilitated skid trail soil. *Soil Science Society of America Journal* 52:512-516.
- Franzuebbers AJ (1999). Microbial activity in response to water-filled pore space of variably eroded southern Piedmont soils. *Applied Soil Ecology* 11:91-101.
- Haney RL, Frenzlubbars AJ, Hons FM, Hossner LR, Zuberer DA (2001). Elucidation of source and turnover of water soluble and microbial biomass carbon in agricultural soils. *Soil Biology and Biochemistry* 133:1501-1507.
- Hassink J (1994). Effects of soil texture and grassland management on soil organic C and N and rates of C and N mineralization. *Soil Biology and Biochemistry* 26:1221-1231.
- Jensen LS, Mcqueen DJ, Shepherd TG (1996a). Effects of compaction on N-mineralization and microbial-C and -N. I. Field measurements. *Soil Tillage Research* 38:175-188.
- Jensen LS, Mcqueen DJ, Ross DJ, Tate KR (1996b). Effects of soil compaction on N-mineralization and microbial-C and -N. II. Laboratory simulation. *Soil Tillage Research* 38:189-202.
- Jensen ES (1996). Rhizodeposition of N by pea and barley and its effect on soil N dynamics. *Soil Biology and Biochemistry* 28:65-71.
- Kaiser K, Guggenberger G (2006). Sorptive stabilization of organic matter by microporous goethite: sorption into small pores vs. surface complexation. *European Journal of Soil Science* 58:45-59.
- Lee WJ, Wood CW, Reeves DW, Entry JA, Raper RL (1996). Interactive effects of wheel-traffic and tillage system on soil carbon and nitrogen. *Communications in Soil Science and Plant Analysis* 27:3027-3043.
- Linn DM, Doran JW (1984). Effects of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and non-tilled soils. *Soil Science Society of America Journal* 48:1267-1272.
- Matson PA, Parton WJ, Power AG, Swift MJ (1997). Agricultural Intensification and Ecosystem Properties. *Science* 277:504-509.
- Myers RJK, Campbell CA, Weier KL (1982). Quantitative relationship between net nitrogen mineralization and moisture content of soils. *Canadian Journal of Soil Science* 62:111-114.
- Nelson DW, Sommers LE (1982). Total Carbon, Organic Carbon and Organic Matter. In: *Methods of Soil Analysis: Part 3 Chemical and Microbiological Properties*, Bigham J.M. (Ed.). ASA, CSSA, SAAJ, Madison, WI. pp. 961-1010.
- Oikeh SO, Chude VO, Carsky RJ, Weber GK, Horst WJ (1998). Legume rotation in the moist tropical savanna: Managing soil nitrogen dynamics and cereal yields in farmers' fields. *Experimental Agriculture* 34:73- 83.
- Omeke JO (2017). Microbial biomass carbon and soil organic carbon ratio as influenced by rhizobium inoculated soybean-maize cropping systems in greenhouse experiment. *Proceeding of the 51st Annual Conference of The Agricultural Society of Nigeria Abuja 2017*. pp. 1012-1020
- Omeke JO (2016). Effect of tillage, cropping systems and nitrogen fertilizer application on the productivity of a savanna Alfisol, Nigeria. PhD Thesis, Ahmadu Bello University Zaria, Nigeria.
- Omeke JO, Yusuf AA, Uyovbisere OE, Abu ST (2016). Assessment of microbial biomass carbon and nitrogen under tillage, cropping

- systems and N fertilizer rate in the savanna Alfisol of Nigeria. *Academia Journal of Agricultural Research* 4(5):258-267.
- Peoples MB, Herridge DF, Ladha JK (1995). Biological nitrogen fixation: an efficient source of nitrogen for sustainable agricultural production. *Plant and Soil* 174(1-2):3-28.
- Rodrigo A, Recous S, Neel C, Mary B (1997). Modelling temperature and moisture effects on C-N transformations in soils: comparison of nine models. *Ecological Modelling* 102:325-339.
- Roper MM, Ladha JK (1995). Biological N₂ fixation by heterotrophic and phototrophic bacteria in association with straw. *Plant and Soil* 174(1-2):211-224.
- Ruser R, Flessa H, Russow R, Schmidt G, Buegger F, Munch JC (2006). Emission of N₂O, N₂ and CO₂ from soil fertilized with nitrate: effect of compaction, soil moisture and rewetting. *Soil Biology and Biochemistry* 38:263-274.
- SAS Institute Inc. (2009). SAS/STAT Users' guide. Version 8, 6th edn. Statistical Analysis Institute, Cary, NC.
- Sérgio RS, Ivo RS, Nairam FB, Eduardo de SM (2011). Effect of compaction on microbial activity and carbon and nitrogen transformations in two Oxisols with different mineralogy. *Revista Brasileira de Ciência do Solo* 35:1141-1149
- Silva SR, Barros NF, Costa LM, Leite FP (2008). Soil compaction and eucalyptus growth in response to forwarder traffic intensity and load. *Revista Brasileira de Ciência do Solo* 32:921-932.
- Sparling GP, West AW (1988). Modification to the fumigation-extraction technique to permit simultaneous extraction and estimation of soil microbial C and N. *Communications in Soil Science and Plant Analysis* 19:327-344.
- Sparling GP (1992). Ratio of microbial biomass carbon to soil organic carbon as a sensitive indicator of changes in soil organic matter. *Australian Journal of Soil Resources* 30(2):195-207.
- Spedding TA, Hamel C, Mehuys GR, Madramootoo CA (2004). Soil microbial dynamics in maize-growing soil under different tillage and residue management systems. *Soil Biology and Biochemistry* 36:499-512.
- Stark C, Condrón LM, Stewart A, Di HJ, O'Callaghan M (2007). Effects of past and current crop management on soil microbial biomass and activity. *Biology and Fertility of Soils* 43:531-540.
- Startsev AD, McNabb DH (2000). Effects of skidding on forest soil infiltration in west-central Alberta. *Canadian Journal of Soil Science* 80:617-624.
- Tan X, Chang SX, Kabzems R (2005). Effects of soil compaction and forest floor removal on soil microbial properties and N transformations in a boreal forest long term soil productivity study. *Forest Ecology Management* 217:158-170.
- Tomich TP, Brodt S, Ferris H, Galt R, Horwath WR, Kebreab E, Leveau JHJ, Liptzin D, Lubell M, Merel P, Michelsmore R, Rosenstock T, Scow K, Six J, Williams N, Yang L (2011). Agroecology: A Review from a Global-Change Perspective. *Annual Review of Environment and Resources* 36:193-222.
- Torbert HA, Wood CW (1992). Effects of soil compaction and water-filled pore space on soil microbial activity and N losses. *Communications in Soil Science and Plant Analysis* 23:1321-1331.
- Vance ED, Brookes PC, Jenkinson DS (1987). An extraction method for measuring microbial biomass C. *Soil Biology and Biochemistry* 19:703-707.
- Vincent JM (1970). A manual for the practical study of root-nodule bacteria. IBP Handbook of methods. No.15. Blackwell Scientific Publication, Oxford.
- Vinze FSP, Feigl BJ, Piccolo MC, Lorival FL, Marcos SN, Cerri CC (2004). Root systems and soil microbial biomass under no-tillage system. *Scientia Agricola* 61(5):529-537.
- Wang JJ, Li XY, Zhu AN, Zhang XK, Zhang HW, Liang WJ (2012). Effects of tillage and residue management on soil microbial communities in North China. *Plant Soil Environment* 58(1):28-33.
- Whalley WR, Dumitru E, Dester AR (1995). Biological effect of soil compaction; V.2: In protection of the soil environment by avoidance of compaction and proper soil tillage. *Proceedings of the International Conference, Melitopol, Ukraine, August 23rd-30th, 1993*; 21-24.
- Yadvinder-Singh MS (2010). Options for effective utilization of crop residues, Directorate of Research, Punjab Agricultural University, Ludhiana, India.