MICROBIAL ALTERATIONS OF THE SOIL INFLUENCED BY INDUCED COMPACTION[1]

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SUMMARY

Compaction is one of the most destructive factors of soil quality, however the effects on the microbial community and enzyme activity have not been investigated in detail so far. The objective of this study was to evaluate the effects of soil compaction caused by the traffic of agricultural machines on the soil microbial community and its enzyme activity. Six compaction levels were induced by tractors with different weights driving over a Eutrustox soil and the final density was measured. Soil samples were collected after corn from the layers 0–0.10 and 0.10–0.20 m. The compaction effect on all studied properties was evident. Total bacteria counts were reduced significantly (by 22–30 %) and by 38–41 % of nitrifying bacteria in the soil with highest bulk density compared to the control. On the other hand, fungi populations increased 55–86 % and denitrifying bacteria 49–53 %. Dehydrogenase activity decreased 20–34 %, urease 44–46 % and phosphatase 26–28 %. The organic matter content and soil pH decreased more in the 0–0.10 than in the 0.10–0.20 m layer and possibly influenced the reduction of the microbial counts, except denitrifying bacteria, and all enzyme activities, except urease. Results indicated that soil compaction influences the community of aerobic microorganisms and their activity. This effect can alter nutrient cycling and reduce crop yields.

Index terms: microorganisms, enzymes, corn.

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RESUMO: *ALTERAÇÕES MICROBIANAS DO SOLO INFLUENCIADAS POR COMPACTAÇÃO INDUZIDA*

A compactação é um dos fatores mais agravantes para a qualidade do solo, porém o seu efeito na comunidade e atividade enzimática microbiana não tem sido suficientemente estudado. Seis níveis de compactação foram obtidos pela passagem de tratores com diferentes pesos em um Latossolo Vermelho, e a densidade final foi medida. Amostras de solo foram coletadas nas profundidades de 0–10 e 10–20 cm, após a colheita do milho. O efeito da compactação foi evidente em todos os parâmetros estudados, mas nem sempre foi significativo. A contagem das bactérias totais reduziu significativamente em 22–30 %, e a das nitrificantes, em 38–41 %, no solo com maior densidade em relação ao controle. Contudo, a população de fungos aumentou de 55 a 86 %, e a das bactérias desnitrificantes, de 49 a 53 %. A atividade da desidrogenase diminuiu de 20 a 34 %; a da urease, de 44 a 46 %; e a da fosfatase, de 26 a 28 %. O conteúdo de matéria orgânica e o pH do solo diminuíram na camada 0–0,10 em relação à de 0,10–0,20 m e influíram possivelmente na redução das contagens microbianas exceto das bactérias desnitrificantes, e na atividade das enzimas, menos a da urease. Esses resultados indicam que a compactação do solo teve influência na comunidade de microrganismos aeróbios e na sua atividade. Esse efeito pode alterar a ciclagem de nutrientes e diminuir a produção da planta.

Termos de indexação: microrganismos, enzimas, milho, compactação.

INTRODUCTION

Modern agriculture is based on an intensive use of agricultural equipment for soil handling and fertilizer and pesticide application. Intense traffic by agricultural machinery causes compaction and, as a consequence, changes the soil physically and reduces plant productivity (Lanzanova et al., 2007). The combination of these factors can diminish crop yields compared to uncompacted areas. In addition, macroporosity and total porosity are reduced, increasing soil density and making root penetration more difficult (Giarola et al., 2007). A study with corn seedlings showed that an increased global soil density resulted in reduced length and number of seminal adventitious roots (Rosolem et al., 1999). Although innumerable studies have studied the effect of compaction on soil physical properties and plant development, few studies have focused on the dynamics of microorganism populations and enzymes involved in the soil metabolic processes.

Soil enzymes and microbial activities are sensitive to abrupt environmental changes. Compaction can influence the plant-available nutrients because the number and activity of microorganisms may be diminished. (Lee et al., 1996). Li et al. (2003), for example, showed a reduction in biomass and microbial activity. Populations of bacteria, total fungi, nematodes, and arthropods were significantly larger in control than in compacted soil (Smeltzer et al., 1986). Breland & Hansen (1995) reported an 18 % reduction in the mineralization of organic ^{15}N from clover in compacted soil compared to the control.

The main effect of compaction consists in altering the availability of soil aeration due to the loss of biopores and other macropores (Whalley et al., 1995, Lima et al., 2005). Anaerobiosis conditions tend to modify the predominant microbial populations which transform nutrients in the soil. Under controlled conditions, the higher N loss was related to the increase in soil density and saturation as a consequence of denitrification (Torbert & Wood, 1992; Jordan et al., 2003). Soil nitrification is generally catalyzed by aerobic bacteria, whereas denitrification is favored by anaerobic conditions. The increase of anaerobiosis due to excess rainfall and pore saturation resulted in significant N_2O loss from the N fertilizers (Clayton et al., 1997). Denitrifying bacteria were predominant in compacted soil, which increased N_2O emission in potato (Ruser et al., 2006).

Microbial enzymes have essential functions in the soil and have been used to measure the influence of soil management and quality (Bergstrom et al., 1998). Enzyme activity may hinder or foster nutrient cycling and crop yields, apart from causing soil physical and chemical alterations (Martens et al., 1992; Dick et al., 1997). Organic matter is usually seen as an indicator of soil quality and biological transformations, and is a source of information regarding soil management (Riffaldi et al., 2003; Garcia & Nahas., 2007; Araújo & Monteiro, 2007). The objective of this study was to evaluate the effect of compaction caused by agricultural machinery on the soil microbial community and enzyme activity.

MATERIALS AND METHODS

This study was conducted at the Faculdade de Ciências Agrárias e Veterinárias, (FCAV/UNESP) in

Jaboticabal, state of São Paulo (lat. 21 ° 15 ' 29 " S, long. 48 ° 16 ' 47 " W, 614 m asl). The climate, according to Köppen, was classified as Cwa, with hot summers and dry winters. The mean annual medium is 21 °C and rainfall 1,428 mm.

To simulate different compaction levels, tractors weighing 4 t (treatment b) and 11 t (other treatments) were driven across the fields and the resulting soil density (D) was measured. The treatments were: (a) area free of agricultural traffic (D = 1.35); (b) one pass of the tractor (D = 1.62); (c) one pass (D = 1.66); (d) two passes (D = 1.70); (e) four passes (D = 1.73); and (f) six passes (D = 1.74). The triple Master hybrid corn was cultivated in October, 2004, at a spacing of 90 cm between rows, in medium typical dystrophic texture Red Latosol soil with moderate hypoferric kaolinitic (LVd). The crop was fertilized at the beginning with 0.3 Mg ha^{-1} of the compound fertilizer 10–20–20 after plant emergence and later with 0.3 Mg ha^{-1} of ammonium sulfate by covering 28 days after plant emergence. The specific soil characteristics and form of cultivation were reported by Freddi et al. (2007).

Soil samples were collected after the corn harvest at depths of 0–10 and 10–20 cm by a screw auger. Five simple samples were taken from each plot, which were combined in a composite sample, for a total of eight treatment samples: four each from the 0–10 cm and 10–20 cm layer. The samples were sifted through a 2 mm mesh sieve and divided into two fractions: one part was kept in a refrigerator until the microbiological analysis and the other part was air-dried and conserved at room temperature for chemical analysis.

The culture media of Bunt & Rovira (1955) and of Martin (1950) were used to quantify the number of colony-forming units (CFU) of bacteria and total fungi, respectively. The most probable number (MPN) of denitrifying and nitrifying bacteria was determined according to Alexander (1982), using the culture media proposed by Alexander (1965) and Schmidt & Belser (1982), respectively. The incubation time at 30 °C was 24 h for total bacteria, 48 h for total fungi, 4 days for nitrifying bacteria, and 7 days for denitrifying bacteria.

The activity of urease, acid phosphatase, and dehydrogenase enzymes was determined according to McGarity & Myers (1967), Tabatabai & Bremner (1969), and Casida Jr. et al. (1964), respectively. Urease activity was expressed as μg NH_4-N g^{-1} dry soil 3 h^{-1}, acid phosphatase as μg p-nitrofenol g^{-1} dry soil h^{-1}, and dehydrogenase as μg triphenylformazan g^{-1} dry soil 24 h^{-1}.

Soil moisture was determined after drying soil samples at 105 °C to constant weight and organic matter by oven incineration at 550 °C for 24 h. The soil pH was measured in a mixture containing 10 cm^3 soil and 25 mL 0.01 mol L^{-1} $CaCl_2.2\,H_2O$.

The experiment was arranged in a split-plot design with four replications. Each experimental plot consisted of five 3-m-long corn rows, where only three rows were evaluated. The different compaction levels were considered in the plots, and the sampling soil depths in the subplots. Results were submitted to variance analysis using software SAS (1990) and means were compared by the Tukey test ($p < 0.05$).

RESULTS

The number of bacteria decreased progressively and significantly (Tukey, $p < 0.05$), up to 30 % at 0–10 cm and 22 % at 10–20 cm depth, when $D_{1.74}$ was compared with the control soil ($D_{1.35}$) (Figure 1a). Fungi counts ($p < 0.05$) were largest in the highest density soils ($D_{1.73}$ and $D_{1.74}$) and decreased in the lower density soils (Figure 1b). The MPN of nitrifying bacteria increased in the $D_{1.62}$ (0–10 cm) and $D_{1.66}$ (10–20 cm) soils, but decreased as soil density increased (Figure 2a). The decrease ($p < 0.05$) in MPN of nitrifying bacteria in $D_{1.74}$ soil varied 38–41 %

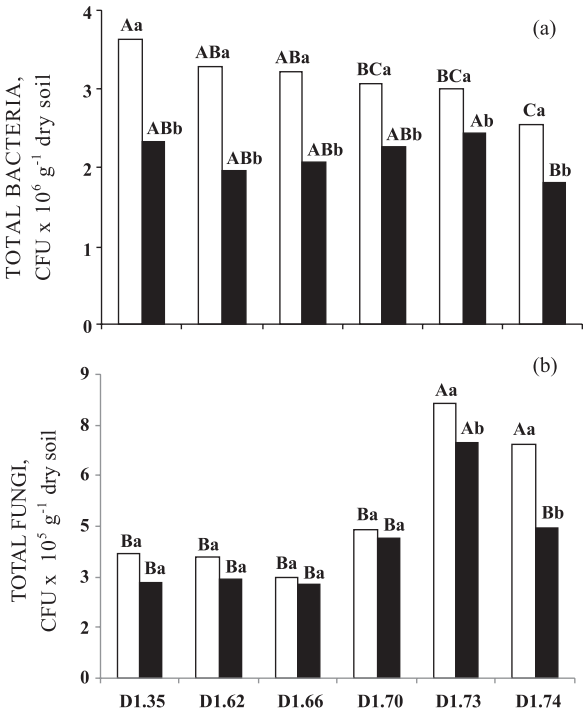

Figure 1. Bacteria (a) and fungi (b) populations observed in the layers 0–10 (▭) and 10–20 cm (■) of the soil. Bars with the same letter (capital letter: density, and lower-case letter: soil depth) are not significant ($p < 0.05$) different. CFU, colony forming units; Bacteria: F test: density (D) = 5.51; depth (P) = 158.39***; DxP = 2.66*. Fungi: F test: D = 53.51***; P = 12.82**; DxP =1.82NS. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; NS, not significant.**

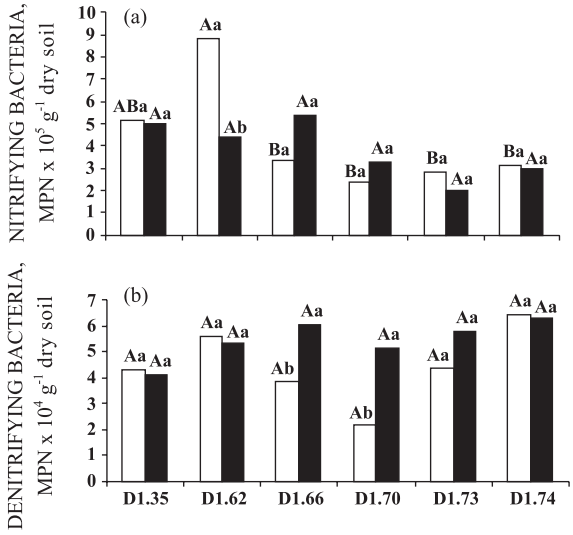

Figure 2. Nitrifying (a) and denitrifying (b) populations found in the layers 0–10 (□) and 10–20 cm (■) of the soil. Bars with the same letter (capital letter: density, and lower-case letter: soil depth) are not significant ($p < 0.05$) different. NMP, most probable number; Nitrifying: F test: density (D) = 5.00*; depth (P) = 0.47NS; DxP = 0.93NS. Denitrifying: F test: (D) = 1.14NS; (P) = 7.09*; DxP = 1.68NS. * $p < 0.05$; NS, not significant.

compared to the control. The MPN of denitrifying bacteria in the 0–10 cm layer rose initially with the increasing soil density; however, decreased in the $D_{1.66}$ and $D_{1.70}$ soils, but later increased (Figure 2b). The MPN growth of denitrifying bacteria in the $D_{1.74}$ soil was 49–53 % in relation to the control, which was the highest ($p < 0.05$) of both soil depths.

When comparing the 0–10 cm layer to the 10–20 cm layer, the mean bacteria and total fungi counts decreased but the denitrifying bacteria increased significantly. The MPN of nitrifying bacteria decreased only 9 %.

Increased soil compaction affected the activity of dehydrogenase, urease, and acid phosphatase enzymes (Figures 3 and 4). Dehydrogenase activity decreased with increased soil density; a significant decrease of 20 % was found in the 0–10 cm and 34 % in the 10–20 cm layer, when the activity in the $D_{1.74}$ soil was compared with the control (Figure 3). Similar results were found with urease activity, which decreased by 46 and 44 %, respectively (Figure 4a). Phosphatase activity decreased significantly with increasing soil density compared to the control; the decrease was greatest in the $D_{1.74}$ soil, 28 % in the 0–10 cm and 26 % in the 10–20 cm layer, compared to the control (Figure 4b).

The means of the soil layers 0–10 and 10–20 cm showed that only dehydrogenase and phosphatase activities decreased significantly (43 and 7 %, respectively).

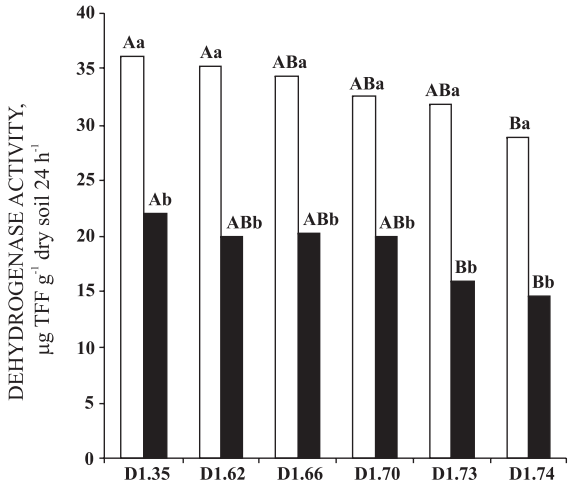

Figure 3. Dehydrogenase activity observed in the layer 0–10 (□) and 10–20 cm (■) of the soil. Bars with the same letter (capital letter: density, and lower-case letter: soil depth) are not significant ($p < 0.05$) different. F test: density (D) = 9.79; depth (P) = 269.95***; DxP = 0.28NS. ** $p < 0,01$; *** $p < 0.001$; NS, not significant. TFF, triphenylformazan.**

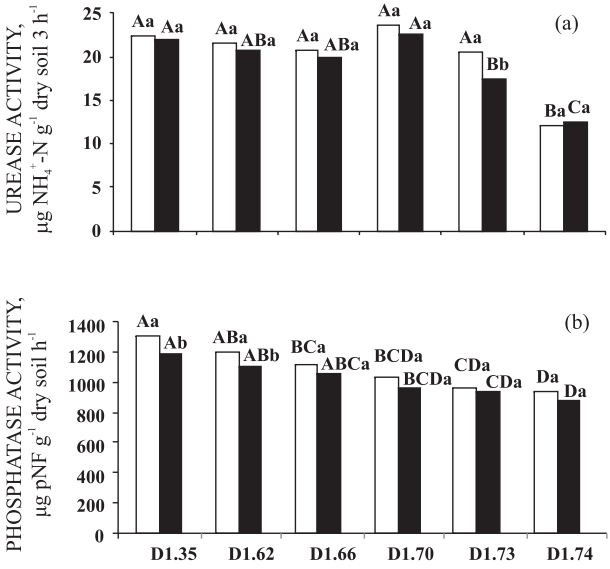

Figure 4. Urease (a) and acid phosphatase (b) activities found in the layers 0–10 (□) and 10–20 cm (■) of the soil. Bars with the same letter (capital letter: density, and lower-case letter: soil depth) are not significant ($p < 0.05$) different. Urease: F test: density (D) = 75.00*; depth (P) = 2.84NS; DxP = 0.79NS. Acid phosphatase: F test: (D) = 15.51***; (P) = 16.76***; DxP = 0.46NS. *** $p < 0.001$; NS, not significant. pNF, p-nitrofenol.**

Figure 5 illustrates that compaction influenced moisture significantly, but not organic matter or soil pH. The moisture content slightly increased with compaction, 6 % in the $D_{1.74}$ soil in relation to the

control ($p < 0.05$) (Figure 5a). While the mean moisture content (Figure 5a) increased 13 % with soil depth, organic matter (Figure 5b) and soil pH (Figure 5c) decreased 3 and 5 %, respectively.

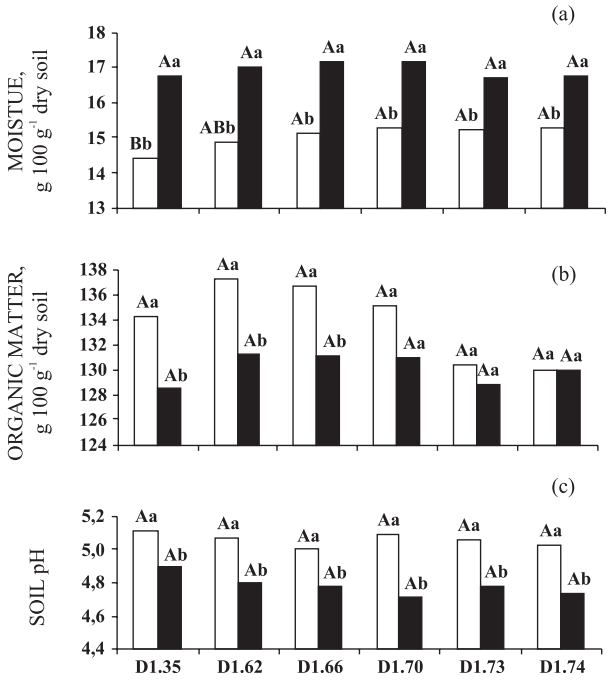

Figure 5. Moisture (a) and organic matter (b) contents and pH (c) found in the layers 0–10 (▭) and 10–20 cm (■) of the soil. Bars with the same letter (capital letter: density, and lower-case letter: soil depth) are not significant ($p < 0.05$) different. Moisture: F test: density (D) = 3.79*; depth (P) = 473.04*; DxP = 2.79*. Organic matter: F test: (D) = 2.58NS; (P) = 10.41**; DxP = 0.69NS. pH: F test: (D) = 2.42NS; (P) = 114.73***; DxP = 0.85 NS. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; NS, not significant.**

DISCUSSION

The community of total and nitrifying bacteria was sensitive to the increase in soil density; however, the fungi and denitrifying bacteria benefited. The effect of compaction in reducing bacteria and fungi populations had been observed by Smeltzer et al. (1986). Therefore, increased soil compaction may have restricted the growth of total and nitrifying bacteria but favored denitrifying bacteria. Some factors may explain the results of this study. As density increased, soil macroporosity (Freddi et al., 2007) and O_2 availability decreased (Pengthamkeerati et al., 2006; Miransari et al., 2007). Limited soil aeration might therefore inhibit the growth of aerobic microorganisms.

Compaction caused by machinery traffic for corn cultivation under conventional tillage in the 0–12 cm layer increased bulk density and penetration resistance, reducing root abundance (Taboada et al., 2008). However, despite the physical consequences of soil compaction, root growth increased in the 0–10 m layer, although corn yields decreased (Freddi et al., 2007). The decreased corn productivity may have been a consequence of the decreased amount of roots and exudation. Since the restriction of the bacterial community depends on the exudates for growth, the transformations of soil nutrients required by the crops would also be limited (Lee et al., 1996). The increase of the denitrifying bacteria community in high-density soils can result in N loss due to the dessimilatory nitrate reduction and decrease N availability for corn. In fact, the higher N_2O emission, observed in compacted forest soil, confirms the stimulation of denitrifying bacteria activity (Teepe et al., 2004). According to Smeltzer et al. (1986), fungi are important agents in soil and the number of populations decreased with greater compaction. However, in this study, the community of fungi benefited from increased density, probably due to adaptation to the new soil conditions (Shestak & Busse, 2005).

The results obtained in this study showed a tendency towards a reduction of the studied enzyme activity, however, a significant decrease ($p < 0.05$) was observed in $D_{1.74}$ soil compared to the control. Dehydrogenase correlated with aeration conditions of the soil, which suggests that the soil compaction or saturation degree influence the activity of this enzyme (Brezezínska et al., 2001). Dick et al. (1988) also detected reduction in the dehydrogenase, phosphatase, arylsulphatase, and amylase activities from 41 to 75 % in compacted soil. Enzyme activities are related to microbial growth (Taylor et al., 2002; Kremer & Li, 2003), therefore, the reduction in the enzyme activity may be a consequence of the decrease in total bacteria counts with increased compaction. The fungi and the denitrifying bacteria, which had increased with increasing soil density, influenced the enzyme activity less because they represent only a small fraction of the total bacterial community. Dehydrogenase catalyzed the electron transfer, and was influenced by the microbial community and indirectly by soil management (Brezezínska et al., 2001). Phosphatase and urease are important enzymes produced by the microorganisms that promote the transformation of organic compounds, which are unavailable to plants in mineral form (Sylvia et al., 2005). Since soil compaction decreases the activity of these enzymes, nutrient availability and plant growth are consequently affected.

The microorganism counts, aside from denitrifying bacteria, as well as the enzyme activity, aside from urease, decreased significantly in the 0–10 cm compared to the 10–20 cm soil layer. This decrease can be attributed to the significant decrease of organic matter content and soil pH between these soil layers. Possibly, the highest content of organic matter in the surface soil layer might be due to the accumulation of corn residues. Organic matter can act as C and energy

source which stimulates heterotrophic microorganisms (Vaidya et al., 2008) and influences enzyme activity. The addition of organic matter increased dehydrogenase activity in corn (Masciandaro et al., 2000). It was also observed that phosphatase activity correlated with organic matter and soil moisture (Amador et al., 1997).

CONCLUSIONS

1. Soil compaction in corn inhibited growth of the total and nitrifying bacteria populations; on the other hand, total fungi and denitrifying bacteria were stimulated.

2. Increased soil density stimulated dehydrogenase and phosphatase activities, but not urease activity.

3. The total bacteria and fungi counts, dehydrogenase and phosphatase enzyme activity, as well as organic matter content and soil pH increased more in the 0–10 cm than in the 10–20 cm soil layer. The inverse was observed for the bacteria counts and soil moisture.

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