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## SPECIAL ISSUE ARTICLE

# Long-term surface application of dairy liquid manure to soil under no-till improves carbon and nitrogen stocks

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**Abstract**

Manure application and no-till management improve soil quality, crop yields and C inputs, but little specific knowledge is available regarding the combined effect of long-term dairy liquid manure (DLM) application and no-till management on soil organic matter status and soil structure in topsoils. Here, we assess the impacts of surface-applied DLM for a 10-year period on soil total organic carbon (TOC) and nitrogen (TN) concentrations and stocks, on organic matter fractions and on soil physical-structural attributes in a no-till crop rotation system on clayey Ferralsol under no-till crop rotation management in a subtropical climate. Four rates of DLM (0, 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>) were applied in a randomized block design with four replications. Soil samples were collected to calculate TOC and TN stocks to 100-cm depth and organic matter fractions in the 0–5-cm layer, and physical indicators of soil quality: bulk density, macroporosity and microporosity and aggregate stability. The largest DLM rates (120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>) added 3.8–5.8 Mg C ha<sup>-1</sup> year<sup>-1</sup> as manure, increased crop residue input by 0.6 Mg C ha<sup>-1</sup> year<sup>-1</sup>, and increased TOC stock by 17% and TN stock by 27% in the top 10 cm of soil, at annual accumulation rates that averaged 0.72 Mg C ha<sup>-1</sup> year<sup>-1</sup> and 86 kg N ha<sup>-1</sup> year<sup>-1</sup>. Stocks of TOC and TN in the sand-POM (particulate organic matter) fraction under the 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> rate increased by 49 and 63%, and in the silt-MOM (mineral-associated organic matter) fraction by 30 and 47%, respectively. In the clay-MOM fraction, the change in TOC was not significant, but the TN increased by 16%. Soil physical-structural attributes (bulk density, porosity and aggregates) were improved by DLM application in the 10-cm layer and followed the soil carbon and nitrogen increments. Overall, the DLM amendment applied on the soil surface of no-till proved to be a strategy to ameliorate soil organic matter status, an important factor for agronomic and environmental benefits, besides improving crop biomass production.

**Highlights**

- Long-term application of liquid manure increased concentration and stock of SOC and TN in the 0–10-cm layer.
- Increases of 0.72 Mg C ha<sup>-1</sup> year<sup>-1</sup> and 86 kg N ha<sup>-1</sup> year<sup>-1</sup> were observed in the 10-cm topsoil with manure.

- SOC and TN increased in the sand-POM and silt-MOM size fractions but not the clay-MOM fraction.
- Improvements to soil physical-structural attributes were related to organic matter changes.

**KEYWORDS**

carbon sequestration, conservation agriculture, liquid manure, physical fractionation, soil organic carbon, soil physical-structural attributes

## 1 | INTRODUCTION

One of the major worldwide concerns with animal production is the safe disposal of the generated manure. Subtropical southern Brazil has significant animal production in confinement systems, including dairy cattle. The manure produced by these systems has been applied on the soil surface of no-till agricultural fields. This has been the typical soil management system in the region for about 40 years. However, little specific knowledge is available regarding the combined effect of long-term dairy liquid manure (DLM) application and no-till management on soil organic matter status and soil structure in topsoils.

It is well known that manure application to cropland provides an array of benefits for soil quality (Edmeades, 2003; Zavattaro et al., 2017) through the addition of nutrients and organic material that improve physical, chemical and biological properties related to carbon accumulation (He et al., 2015). Manure affects soil quality directly by the input of carbon from manure (Xie et al., 2014) or indirectly by the input of carbon from plant residues (shoots and roots), due to an increase in crop productivity (Courtier-Murias et al., 2013). However, most of these studies are in temperate climates with solid manure incorporated into the soil in short-term experiments. Comparatively little is known about the impacts of liquid manure on soil carbon and nitrogen stocks and fractions, and on soil physical attributes when such manure is applied in the long term in subtropical no-till systems as a surface mulch. The few studies on this matter suggest that the minimal soil disturbance combined with long-term manure application has shown some improvement in soil structure (Mellek et al., 2010) and incremental increases in soil organic matter status (Maillard et al., 2015), with reduced runoff and water pollution (Gilley & Risse, 2000; Tomer, Moorman, Kovar, Cole, & Nichols, 2016). On the other hand, liquid manure application on the soil surface can cause surface sealing due to the clogging of soil pores and, thus, increase surface runoff and consequent loss of nutrients and sediments that are potential sources of water pollution (Cherobim,

Favaretto, Melo, & Barth, 2018; Mori, Favaretto, Pauletti, Dieckow, & Santos, 2009).

The effect of long-term liquid manure application on the carbon and nitrogen in soil physical fractions in no-till subtropical soils is not well understood. The physical granulometric fractionation of soil organic matter is a useful technique for the quantification of the particulate organic matter (POM) present in sand-size fractions and the mineral-associated organic matter (MOM) present in silt- and clay-size fractions (Christensen, 1996). The POM fraction acts as an important source of nutrients and energy for soil microorganisms, which, during the decomposition process, excrete organic compounds that act as binding agents and directly influence soil aggregation, (He et al., 2015; Puget, Chenu, & Balesdent, 2000; Yu, Ding, Luo, Geng, & Cai, 2012). The MOM fraction is considered to be stabilized by organo-mineral interactions and physically protected in the interior of the microaggregates (<250  $\mu\text{m}$ ) (Bayer, Martin-Neto, Mielniczuk, & Pavinato, 2004; Cotrufo et al., 2015; Pinheiro, Campos, Balieiro, Anjos, & Pereira, 2015).

The hypothesis of our study is that long-term application of dairy liquid manure (DLM) increases total organic carbon and total nitrogen stocks in no-till soil due to carbon input via manure, with a major effect on the POM fraction and beneficial effects on soil physical-structural attributes. Thus, the objective of this study was to quantify the improvement provided by long-term DLM application to the soil surface of a clayey subtropical Ferralsol in a no-till system for total organic carbon and nitrogen concentrations and stocks, and their physical fractions, with assessment of the effects on soil structure.

## 2 | MATERIAL AND METHODS

### 2.1 | Experimental area

The experiment was conducted in a research station of the ABC Foundation for Agricultural Assistance and Technical Divulgarion, in Castro-PR, southern Brazil (24° 51' 50" S, 49° 56' 25" W; 1027 m altitude). The soil is a

clayey Ferralsol (WRB) or Latossolo Bruno Distrófico típico (Brazilian system), with a 10% slope. The climate is subtropical, humid mesothermal (Cfb, Köppen), with a mean annual precipitation of 1,554 mm (IAPAR, 2000).

The experiment was installed in May 2006 in a cropland area that had been managed under no-till for over 15 years. Some soil physical attributes in the 0–20-cm layer at the beginning of the experiment were: 701 g kg<sup>-1</sup> of clay, 111 g kg<sup>-1</sup> of silt, 188 g kg<sup>-1</sup> of sand, mean weight diameter of water-stable aggregates (MWDw) of 2.92 mm, bulk density of 1.03 kg dm<sup>-3</sup>, microporosity of 0.44 m<sup>3</sup> m<sup>-3</sup>, macroporosity of 0.17 m<sup>3</sup> m<sup>-3</sup>, and saturated hydraulic conductivity of 5.2 mm h<sup>-1</sup> (Abboud, Favaretto, Motta, Barth, & Goulart, 2018). The cropping system includes black oats (*Avena strigosa* Schreb.) as a cover crop or wheat (*Triticum aestivum* L.) as a cash crop in the winter, and soybean (*Glycine max* (L.) Merr.) or maize (*Zea mays* L.) as cash crops in the summer.

The treatments were four rates of dairy liquid manure (DLM; 0, 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>) arranged in a randomized block design with four replicates, making 16 plots of 29.75 m<sup>2</sup> each (8.5 m × 3.5 m). The annual rate of DLM was split into two applications: half in winter, at the establishment of black oats or wheat, and half in summer, at the establishment of soybean or maize. The DLM, obtained from a dairy farm in the region, was applied with a watering can without a sieve, directly over the soil surface as a mulch. On average, the dry matter of the DLM was 78.4 g L<sup>-1</sup> (average of 19 applications; Abboud et al., 2018). Mineral fertilizers were applied to each crop according to specific regional recommendations at the same rate to all treatments. The amounts of nitrogen, phosphorus and potassium applied via mineral fertilizers and DLM are presented in Table 1 (data from Abboud et al., 2018).

**TABLE 1** Amount of nitrogen (N), phosphorus (P) and potassium (K) applied via mineral fertilizer (Fert) and via dairy liquid manure (DLM) at rates of 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> (split into winter and summer applications) in a clayey Ferralsol under no-till, over 9.5 years (19 cropping seasons). Castro-PR, Brazil

Season	Crop	N				P				K			
		Fert	DLM			Fert	DLM			Fert	DLM		
kg ha <sup>-1</sup>													
		Fert	60	120	180	Fert	60	120	180	Fert	60	120	180
Winter 2006	Black oats	0	55	109	164	0	23	46	69	0	104	208	312
Summer 2006/07	Maize	175	52	104	157	37	18	37	55	93	71	141	211
Winter 2007	Black oats	0	29	57	86	0	12	25	37	0	44	89	133
Summer 2007/08	Soybean	0	61	121	181	26	22	43	64	50	91	183	274
Winter 2008	Wheat	114	53	105	158	39	18	37	55	59	86	171	256
Summer 2008/09	Soybean	0	33	66	99	22	10	19	29	42	47	94	141
Winter 2009	Black oats	0	28	56	85	0	17	34	51	0	36	72	108
Summer 2009/10	Maize	175	78	156	234	41	28	55	83	63	57	115	172
Winter 2010	Wheat	134	47	94	141	30	21	42	63	63	97	193	290
Summer 2010/11	Soybean	0	52	105	157	26	25	51	76	50	124	249	373
Winter 2011	Black oats	0	69	138	207	0	25	50	75	0	126	252	377
Summer 2011/12	Maize	165	56	111	167	39	21	41	62	50	111	222	334
Winter 2012	Wheat	120	67	134	200	26	20	39	59	50	123	246	369
Summer 2012/13	Soybean	0	47	94	141	26	16	32	48	50	4	8	12
Winter 2013	Black oats	0	50	100	150	0	18	37	55	0	117	235	352
Summer 2013/14	Maize	183	43	87	130	41	13	26	39	75	98	196	294
Winter 2014	Wheat	120	156	312	468	26	26	52	79	50	82	164	247
Summer 2014/15	Soybean	0	126	252	378	26	21	42	63	50	105	209	314
Winter 2015	Black oats	0	46	92	138	0	19	38	57	0	85	171	256
Season mean		62	60	121	181	21	20	39	59	39	85	169	254
Annual mean		125	121	241	362	43	39	79	118	78	169	339	508

Note: Adapted from Abboud et al. (2018).

## 2.2 | Soil sampling

Soil samples were collected in September 2015 when the soil was covered with black oats residue from the previous winter, and when the experiment was 9.5 years old. In two pits per plot, three sets of soil samples were collected from the 0–5, 5–10, 10–20 and 20–30-cm layers as follows: (i) core samples of 5.6 cm in diameter and 3.1 cm high, collected with volumetric rings introduced vertically to the centre of each layer (two rings per layer and per pit) to determine the soil porosity and bulk density; (ii) undisturbed blocks of 10 cm × 10 cm wide and thickness equivalent to that of the layer, collected with a spatula (one block per layer and per pit) to determine the aggregate stability; and (iii) disturbed samples collected with a spatula to determine the concentration and stock of total organic carbon (TOC) and total nitrogen (TN), and to perform the physical fractionation of organic matter (but only in the 0–5-cm layer). For the determination of TOC and TN, additional samples were collected in deeper layers (30–45, 45–60, 60–80 and 80–100 cm), using an auger of 21 cm diameter, and bulk density was determined by the excavation method (samples weighted in the field and corrected to dry mass at 105 °C).

## 2.3 | Soil physical-structural attributes

The core samples in volumetric rings were trimmed, water-saturated and put under 6 kPa tension for microporosity determination. Then, they were dried at 105 °C to determine bulk density. Total porosity was calculated based on the bulk density, assuming a particle density of 2.65 Mg m<sup>-3</sup>. Macroporosity was assumed as the difference between total porosity and microporosity.

The undisturbed block samples were used to determine the mean weight diameter of dry-stable aggregates (MWDd), mean weight diameter of water-stable aggregates (MWDw) and the aggregate stability index (ASI) (Kemper & Rosenau, 1986). Blocks were manually and carefully broken along the fracture planes into aggregates passing 8 mm mesh. Those aggregates were air-dried and stratified in a set of sieves of 4.00, 2.00, 1.00, 0.50 and 0.25 mm meshes to determine the MWDd. For MWDw, 50 g of soil, proportionally composed from the dry aggregates size classes, were submitted to wet sieving (4.00, 2.00, 1.00, 0.50 and 0.25 mm meshes) using a Yoder apparatus (15 minutes, 42 cycles per minute). The ASI was obtained by the ratio between the MWDw and MWDd.

## 2.4 | Total organic carbon, total nitrogen and physical fractionation

The third set of soil samples were air-dried, ground and passed through a 2.00-mm mesh. An aliquot was further ground until the sample passed through a 0.50-mm mesh, and approximately 40 mg of this was used to determine the total organic carbon (TOC) and total nitrogen (TN) concentrations, using the dry combustion method (Vario EL III - Elementar Analysensysteme, Germany). To quantify the stocks of TOC and TN, the concentration of these elements in soil and the bulk density were considered. The stock values were then corrected by the soil equivalent mass (Sisti et al., 2004), the soil mass of the control treatment (no DLM) being the reference.

The TOC and TN accumulation rates were calculated according to the difference of TOC or TN stocks relative to the control treatment (no DLM), divided by the age of the experiment (9.5 years).

The physical granulometric fractionation of the soil organic matter of the 0–5-cm layer was based on the method proposed by Christensen (1996). Approximately 20 g of soil was shaken for 16 hr in 80 mL of water, in a 300-mL flask containing seven 11-mm-diameter poly-acetal beads. This procedure was adopted (after previous tests) to break up the >53- $\mu$ m aggregates without fragmenting the POM. The suspension was passed through a 53- $\mu$ m sieve to separate the sand fraction and POM (sand-POM). This fraction was dried at 45 °C.

The suspension passed through the 53- $\mu$ m mesh was collected and treated with a few drops of 0.5 mol L<sup>-1</sup> CaCl<sub>2</sub> and left for 12 hr rest to flocculate the solids. The supernatant was removed and the flocculate centrifuged to concentrate the solids, which were subsequently dispersed by sonication in an ultrasound apparatus, with dispersion energy of 900 J mL<sup>-1</sup> (determined by previous tests to completely disperse aggregates >2  $\mu$ m). The sonicated suspension was subjected to 10 daily cycles of gravitational sedimentation in 1-L cylinders of 30 cm height, based on Stokes's Law, to separate the silt fraction (53–2  $\mu$ m) from the clay fraction (<2  $\mu$ m). The supernatant containing the clay was transferred to another recipient and, at the end of the sedimentation cycles, it was treated with 0.5 mol L<sup>-1</sup> CaCl<sub>2</sub> to concentrate the clay, which was subsequently dried at 45 °C. The silt fraction, which accumulated at the bottom of the sedimentation cylinder, was also dried at 45 °C. It was assumed that the silt and clay fractions contained the organic matter associated with minerals (silt-MOM and clay-MOM).

After drying, all fractions were ground to 0.20 mm using a mortar for the determination of carbon and

nitrogen concentrations, following the dry combustion method previously described.

## 2.5 | Carbon lability index and carbon management index

The carbon lability index (CLI) and the carbon management index (CMI) were calculated based on Blair, Lefroy, and Lise (1995), considering the sand-POM of the 0–5-cm layer as the labile fraction in the CLI (Dieckow et al., 2005). The control treatment (no DLM) was the reference, with CMI = 100 and CLI = 1.0.

## 2.6 | Carbon input into the soil by plants and DLM

First, aboveground biomass input was estimated using data of grain yield of soybean, maize and wheat, and data of aboveground dry matter yield of black oats. These data were collected during the 9.5 years of the experiment (19 cropping seasons). We considered harvest indexes of 0.52 for soybean, 0.49 for maize and 0.53 for wheat, which were obtained in the same region (Pierri et al., 2016). The root biomass input was estimated by considering the aboveground inputs and the shoot-to-root ratios of 4.4 for black oats (Pietola & Alakukku, 2005), 5.2 for soybean, 5.6 for maize and 6.5 for wheat (Bolinder, Janzen, Gregorich, Angers, & VandenBygaart, 2007). Additionally, the extra-root addition (exudates and other matters, such as fine roots and root hairs) was estimated as being equivalent to 0.65 times the amount of root biomass (Bolinder et al., 2007). The overall plant biomass input was the sum of the aboveground (shoot) and root inputs. To estimate the amount of carbon input by plants, we considered a concentration of 400 g C kg<sup>-1</sup> in the dry biomass (Bayer, Martin-Neto, Mielniczuk, & Ceretta, 2000).

The carbon input by the DLM was estimated by considering the manure dry matter (DM) in each of the 19 applications (data from Abboud et al., 2018) and the carbon concentration of 407 g C kg<sup>-1</sup> DM, which was the average of four measurements: 425.1, 433.1, 366.1 and 403.7 g C kg<sup>-1</sup> DM for summer 2012, summer 2013, winter 2014 and summer 2014, respectively.

## 2.7 | Statistical analyses

The results were submitted to analysis of variance (ANOVA), considering a randomized block design, and means were compared by using the Tukey's test ( $p < .05$ ). The analyses were performed in the R environment

(RStudio Team, 2016) with the help of the ExpDes package (Ferreira, Cavalcanti, & Nogueira, 2013).

## 3 | RESULTS

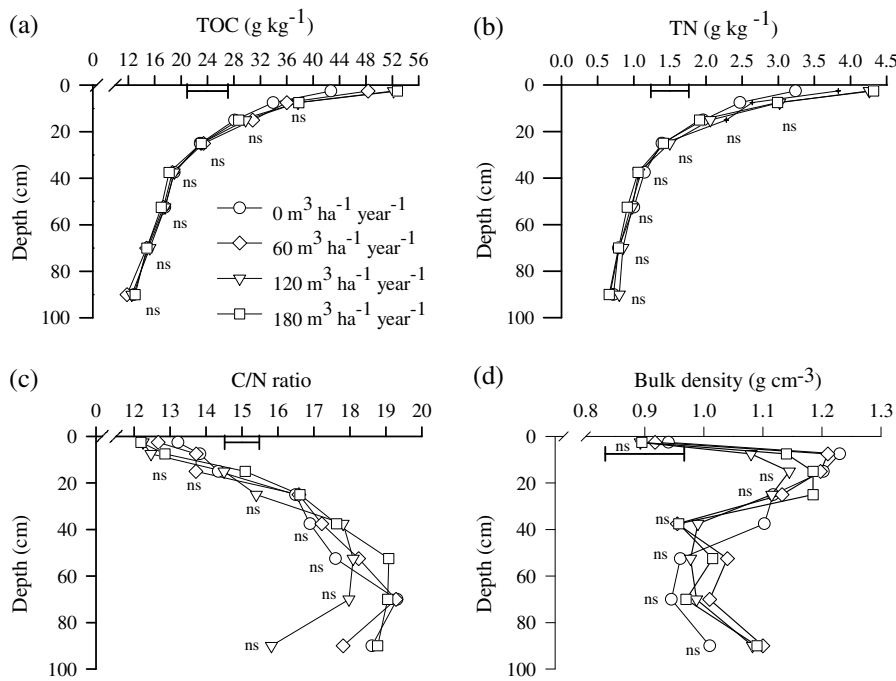
### 3.1 | Total organic carbon and total nitrogen

The long-term application of DLM increased significantly the TOC and TN concentrations in the topsoil layer (0–5 cm) and tended to increase them in the subsequent 5–10-cm layer (Figure 1a,b). Rates of DLM application of 120 or 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> increased TOC and TN concentration by around 22% and 32%, respectively, compared to the control treatment (unmanured) in the 0–5-cm layer. The C/N ratio at this depth decreased by 4%, 7% and 8% with DLM application of 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> (Figure 1c).

The TOC and TN stocks in the top 0–10 cm of soil amended with DLM 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> were significantly larger than those in the unmanured soil: TOC was increased by 17% (average of 47.6 vs. 40.8 Mg C ha<sup>-1</sup>) and TN was increased by 27% (average of 3.84 vs. 3.02 Mg N ha<sup>-1</sup>) (Table 2). Accumulation rates of 0.72 Mg C ha<sup>-1</sup> year<sup>-1</sup> and of 86 kg N ha<sup>-1</sup> year<sup>-1</sup> in the top 10 cm soil were calculated after application of 120 or 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> of DLM (Table 2). The 60 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> DLM application rate had an intermediate effect on TOC and TN stocks, not differing significantly either from control or DLM rates of 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> (Table 2). No significant changes in TOC or TN stocks by DLM application were observed below 10-cm depth (Table 2).

### 3.2 | Carbon and nitrogen in physical fractions

According to the results of physical fractionation in the top 5 cm of soil, much of the carbon and nitrogen accruals after application of DLM rates of 120 or 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> occurred in the sand-POM and silt-MOM fractions (Table 3). Considering the distribution of the carbon and nitrogen accumulation rates in the three evaluated fractions (Table 3), approximately 47 and 62% of the C and N stock increments with DLM rates of 120 or 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> took place in the sand-POM, whereas silt-MOM accounted for approximately 28 and 43%, respectively. In the clay-MOM fraction, the increments of carbon were not significant, but those of nitrogen were, so that 16% of the nitrogen accrual occurred in this fraction (Table 3).



**FIGURE 1** (a) Concentration of total organic carbon (TOC), (b) concentration of total nitrogen (TN), (c) C/N ratio and (d) soil bulk density of a clayey Ferralsol under no-till and subjected to application rates of dairy liquid manure of 0, 60, 120 and 180  $\text{m}^3 \text{ha}^{-1} \text{year}^{-1}$  over 9.5 years. Castro-PR, Brazil. Horizontal bars represent the least significant difference according to Tukey's test ( $p < .05$ ); <sup>ns</sup> not significant

The carbon management index (CMI) was influenced by the DLM, with carbon increment from 45 to 66, compared to the control treatment (Table 3).

### 3.3 | Soil physical-structural attributes

In general, with the application of DLM rates of 120 or 180  $\text{m}^3 \text{ha}^{-1} \text{year}^{-1}$ , macroporosity and aggregate MWD tended to increase, whereas bulk density and microporosity tended to decrease (Figure 1d and Figure 2). However, few of these changes were significantly different in comparison to the control treatment (Figure 1d and Figure 2). Soil bulk density ranged from 0.89 to 1.23  $\text{g cm}^{-3}$  for all treatments and depths (Figure 1d), but DLM application was significant in reducing bulk density only in the 5–10-cm layer (1.23 to 1.08  $\text{g cm}^{-3}$ ). Soil aggregation also was significantly improved by DLM applications but only in the 0–5-cm layer (Figure 2d,e). MWD<sub>d</sub> and MWD<sub>w</sub> increased on average by 26% compared to the control treatment. The ASI ranged from 0.92 to 0.98 (Figure 2f), showing high aggregate stability even in the treatment without DLM application and indicating the effectiveness of the no-till system in improving soil structure in a clayey soil.

## 4 | DISCUSSION

The long-term application of DLM on the soil surface improved the soil organic matter status mostly in the

0–10-cm topsoil (Table 2). The TOC and TN accruals are attributed mainly to plants plus manure leading to an increase in overall carbon inputs, which were approximately twice as high with DLM rates of 120 or 180  $\text{m}^3 \text{ha}^{-1} \text{year}^{-1}$  compared to the control (9.6–11.6 vs. 5.2  $\text{Mg C ha}^{-1} \text{year}^{-1}$ , Table 4). Regarding plant input, the DLM application of 120 and 180  $\text{m}^3 \text{ha}^{-1} \text{year}^{-1}$  increased the C input by around 12% (from 5.2  $\text{Mg C ha}^{-1} \text{year}^{-1}$ , in the control treatment, to 5.8  $\text{Mg C ha}^{-1} \text{year}^{-1}$ ; Table 4). This increase was possibly caused by the addition of nitrogen, phosphorus, potassium and micronutrients to soil by manure (Table 1), improving net primary production of crops as well as residue return (Courtier-Murias et al., 2013; Leonardo, Caviglia, & Pinero, 2017; Wenhai et al., 2016). However, the increase of carbon input via manure was much greater than that via plants; whereas the DLM rate of 180  $\text{m}^3 \text{ha}^{-1} \text{year}^{-1}$  increased plant carbon input by 0.6  $\text{Mg C ha}^{-1} \text{year}^{-1}$ , it increased the manure carbon input by 5.7  $\text{Mg C ha}^{-1} \text{year}^{-1}$ , compared to the control treatment (Table 4). However, that does not necessarily mean that the manure applied on the soil surface (not incorporated) was the main contributor to the soil carbon accrual, because there is evidence that roots may contribute more than residue deposited on the soil surface (Rasse, Rumpel, & Dignac, 2005). We suggest that both the direct manure input and the indirect increase of plant input (aboveground and roots) are both important processes contributing to the soil carbon accrual after manure application on the surface of no-till soil (Gai et al., 2018; Wei et al., 2017).

**TABLE 2** Total organic carbon (TOC) and total nitrogen (TN) stocks and their accumulation rates ( $\Delta$ ) in a clayey Ferralsol under no-till and application of dairy liquid manure (DLM) at rates of 0, 60, 120 and 180  $\text{m}^3 \text{ha}^{-1} \text{year}^{-1}$ , over 9.5 years. Castro-PR, Brazil

Layer (cm)	DLM				<i>p</i> value	CV (%)
	0	60	120	180		
<i>TOC stock (mg ha<sup>-1</sup>)</i>						
0–5	19.9 b	22.7 a	24.5 a	24.7 a	.003	6
0–10	40.8 b	44.7 ab	47.5 a	47.7 a	.02	6
0–20	74.5 <sup>ns</sup>	81.6 <sup>ns</sup>	82.3 <sup>ns</sup>	83.1 <sup>ns</sup>	.09	6
0–30	100.1 <sup>ns</sup>	107.7 <sup>ns</sup>	107.1 <sup>ns</sup>	109.6 <sup>ns</sup>	.21	6
0–45	131.4 <sup>ns</sup>	138.6 <sup>ns</sup>	137.8 <sup>ns</sup>	140.1 <sup>ns</sup>	.36	5
0–60	156.7 <sup>ns</sup>	163.1 <sup>ns</sup>	162.6 <sup>ns</sup>	164.6 <sup>ns</sup>	.62	5
0–80	184.9 <sup>ns</sup>	190.8 <sup>ns</sup>	190.7 <sup>ns</sup>	192.4 <sup>ns</sup>	.81	6
0–100	210.8 <sup>ns</sup>	214.7 <sup>ns</sup>	215.4 <sup>ns</sup>	217.6 <sup>ns</sup>	.91	6
<i><math>\Delta C</math> (Mg ha<sup>-1</sup> year<sup>-1</sup>)</i>						
0–5	-	0.29 *	0.48 *	0.50 *	.003	46
0–10	-	0.41 <sup>ns</sup>	0.71 *	0.73 *	.02	65
0–20	-	0.74 <sup>ns</sup>	0.82 <sup>ns</sup>	0.91 <sup>ns</sup>	.10	80
0–30	-	0.80 <sup>ns</sup>	0.74 <sup>ns</sup>	1.01 <sup>ns</sup>	.21	102
0–45	-	0.76 <sup>ns</sup>	0.67 <sup>ns</sup>	0.91 <sup>ns</sup>	.36	125
0–60	-	0.67 <sup>ns</sup>	0.62 <sup>ns</sup>	0.82 <sup>ns</sup>	.63	176
0–80	-	0.62 <sup>ns</sup>	0.61 <sup>ns</sup>	0.79 <sup>ns</sup>	.98	243
0–100	-	0.41 <sup>ns</sup>	0.49 <sup>ns</sup>	0.71 <sup>ns</sup>	.96	299
<i>TN stock (mg ha<sup>-1</sup>)</i>						
0–5	1.51 c	1.80 b	2.00 ab	2.03 a	0.001	6
0–10	3.02 b	3.40 ab	3.84 a	3.83 a	0.008	8
0–20	5.38 <sup>ns</sup>	6.14 <sup>ns</sup>	6.20 <sup>ns</sup>	6.27 <sup>ns</sup>	0.20	10
0–30	6.94 <sup>ns</sup>	7.71 <sup>ns</sup>	7.81 <sup>ns</sup>	7.92 <sup>ns</sup>	0.32	10
0–45	8.84 <sup>ns</sup>	9.54 <sup>ns</sup>	9.51 <sup>ns</sup>	9.71 <sup>ns</sup>	0.61	10
0–60	10.29 <sup>ns</sup>	10.86 <sup>ns</sup>	10.90 <sup>ns</sup>	11.01 <sup>ns</sup>	0.82	11
0–80	11.76 <sup>ns</sup>	12.35 <sup>ns</sup>	12.46 <sup>ns</sup>	12.51 <sup>ns</sup>	0.85	11
0–100	13.22 <sup>ns</sup>	13.70 <sup>ns</sup>	14.08 <sup>ns</sup>	13.87 <sup>ns</sup>	0.84	10
<i><math>\Delta N</math> (kg ha<sup>-1</sup> year<sup>-1</sup>)</i>						
0–5	-	30 *	51 *	54 *	0.0005	36
0–10	-	39 <sup>ns</sup>	85 *	86 *	0.008	58
0–20	-	79 <sup>ns</sup>	87 <sup>ns</sup>	93 <sup>ns</sup>	0.20	97
0–30	-	81 <sup>ns</sup>	91 <sup>ns</sup>	104 <sup>ns</sup>	0.30	118
0–45	-	73 <sup>ns</sup>	71 <sup>ns</sup>	91 <sup>ns</sup>	0.60	172
0–60	-	60 <sup>ns</sup>	63 <sup>ns</sup>	76 <sup>ns</sup>	0.82	247
0–80	-	62 <sup>ns</sup>	74 <sup>ns</sup>	79 <sup>ns</sup>	0.85	265
0–100	-	50 <sup>ns</sup>	91 <sup>ns</sup>	68 <sup>ns</sup>	0.84	281

Note: Means followed by the same letter within a row are not significantly different (Tukey's test,  $p < .05$ ). For accumulation rates ( $\Delta\text{TOC}$  or  $\Delta\text{TN}$ ), \* denotes a significant accumulation rate relative to control (no DLM). <sup>ns</sup> not significant. Abbreviation: CV, coefficient of variation.

Zhang et al. (2015), in a long-term experiment with manure and plant residue (*Zea mays* L.), verified changes in the soil organic carbon stocks at 0–20-cm depth. They also emphasized that manure and plant

production was an important strategy for soil organic carbon accumulation.

Regardless of the underlying processes, the higher rates of DLM increased the TOC pool in the 0–10-cm



Fraction	DLM				<i>p</i> value	CV (%)
	0	60	120	180		
<i>Mass ratio (kg fraction kg<sup>-1</sup> soil)</i>						
Sand-POM	0.27 <sup>ns</sup>	0.29 <sup>ns</sup>	0.29 <sup>ns</sup>	0.30 <sup>ns</sup>	.40	7
Silt-MOM	0.29 <sup>ns</sup>	0.28 <sup>ns</sup>	0.29 <sup>ns</sup>	0.29 <sup>ns</sup>	.40	2
Clay-MOM	0.44 <sup>ns</sup>	0.43 <sup>ns</sup>	0.43 <sup>ns</sup>	0.41 <sup>ns</sup>	.22	4
<i>C concentration (g C kg<sup>-1</sup> soil)</i>						
Sand-POM	7.83 b	11.37 ab	12.76 a	12.41 ab	.06	22
Silt-MOM	12.34 b	15.79 ab	15.86 ab	17.07a	.06	14
Clay-MOM	23.28 <sup>ns</sup>	21.58 <sup>ns</sup>	23.89 <sup>ns</sup>	23.77 <sup>ns</sup>	.63	12
<i>N concentration (g N kg<sup>-1</sup> soil)</i>						
Sand-POM	0.47 b	0.78 ab	0.89 a	0.86 a	.04	25
Silt-MOM	0.71 b	0.99 ab	1.06 a	1.17 a	.01	16
Clay-MOM	2.24 <sup>ns</sup>	2.13 <sup>ns</sup>	2.39 <sup>ns</sup>	2.42 <sup>ns</sup>	.47	12
<i>C/N ratio</i>						
Sand-POM	16.6 a	14.6 b	14.4 b	14.5 b	.014	5
Silt-MOM	17.7 a	15.9 b	14.9 b	14.7 b	.001	5
Clay-MOM	10.4 <sup>ns</sup>	10.1 <sup>ns</sup>	10.0 <sup>ns</sup>	9.8 <sup>ns</sup>	.53	5
<i>C stock (mg C ha<sup>-1</sup>)</i>						
Sand-POM	4.40 b	5.94 ab	6.40 a	6.58 a	.02	15
Silt-MOM	5.77 b	6.87 ab	7.26 a	7.52 a	.02	10
Clay-MOM	9.75 <sup>ns</sup>	9.87 <sup>ns</sup>	10.80 <sup>ns</sup>	10.60 <sup>ns</sup>	.19	7
<i>ΔC stock (mg C ha<sup>-1</sup> year<sup>-1</sup>)</i>						
Sand-POM	-	0.16 <sup>ns</sup>	0.21 *	0.23 *	.02	59
Silt-MOM	-	0.11 <sup>ns</sup>	0.15 *	0.18 *	.03	66
Clay-MOM	-	0.01 <sup>ns</sup>	0.11 <sup>ns</sup>	0.09 <sup>ns</sup>	.18	151
<i>N stock (mg N ha<sup>-1</sup>)</i>						
Sand-POM	0.30 b	0.44 a	0.48 a	0.49 a	.01	16
Silt-MOM	0.38 b	0.48 ab	0.53 a	0.56 a	.01	11
Clay-MOM	0.83 b	0.87 ab	0.97 a	0.96 a	.06	8
<i>ΔN stock (kg N ha<sup>-1</sup> year<sup>-1</sup>)</i>						
Sand-POM	-	15 *	20 *	20 *	.004	44
Silt-MOM	-	10 <sup>ns</sup>	16 *	20 *	.03	68
Clay-MOM	-	5 <sup>ns</sup>	15 *	15 *	.11	106
CLI	1.0 <sup>ns</sup>	1.3 <sup>ns</sup>	1.2 <sup>ns</sup>	1.3 <sup>ns</sup>	.24	18
CMI	100 b	145 ab	153 ab	166 a	.06	22

Note: Means followed by the same letter within a row are not significantly different (Tukey's test,  $p < .05$ , except for C concentration of sand and silt,  $p < .10$ ). For accumulation rates ( $\Delta C$  or  $\Delta N$ ), \* denotes a significant accumulation rate relative to control (no DLM). <sup>ns</sup> not significant. Abbreviation: CV, coefficient of variation.

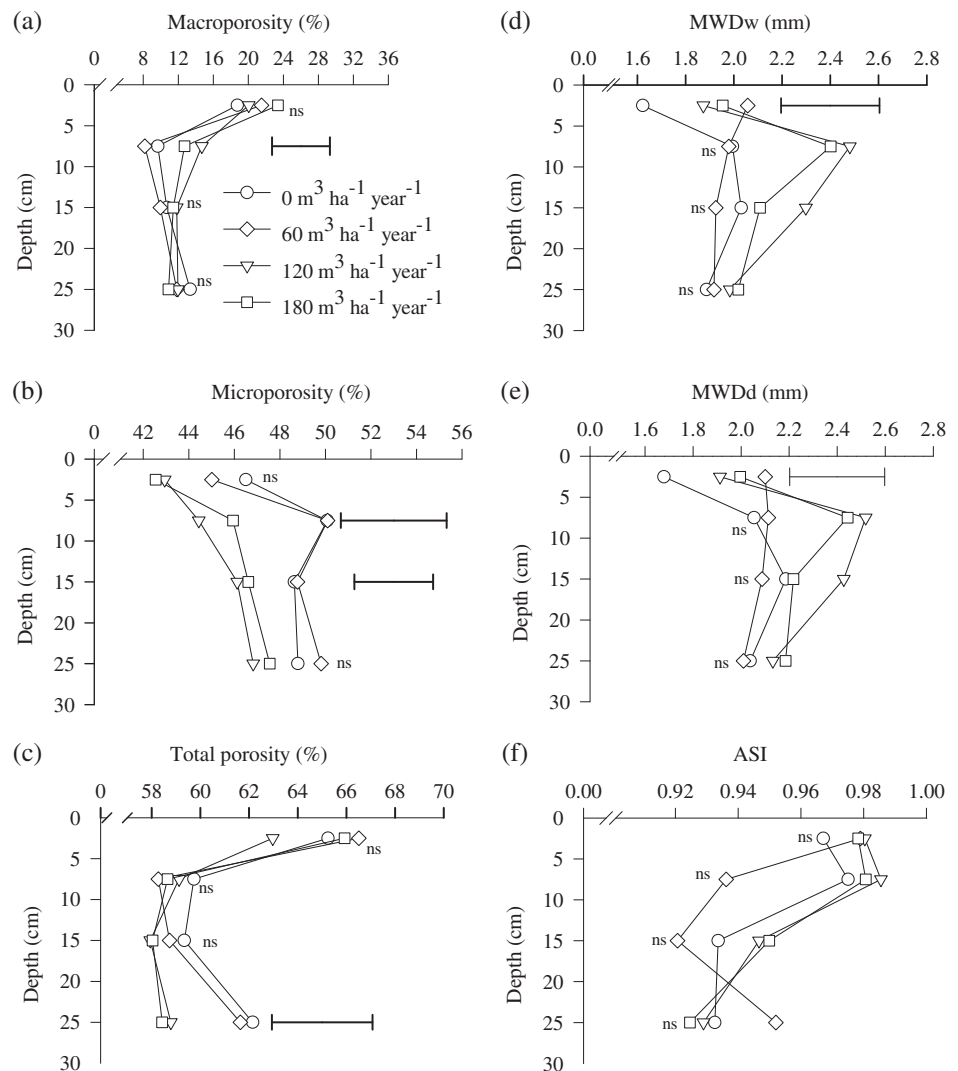
topsoil by 0.72 Mg C ha<sup>-1</sup> year<sup>-1</sup> and the TN pool by 86 kg N ha<sup>-1</sup> year<sup>-1</sup> (Table 2). The beneficial effect of increased soil organic matter is related to all soil quality indicators (Gregorich, Carter, Angers, Monreal, & Ellert, 1994) and food security (Soussana et al., 2019). Moreover, those results suggest that DLM application in no-till soil

is also a good practice to increase soil carbon sequestration in no-till systems, an important factor in the mitigation of climate change (Wei et al., 2017; Wenhai et al., 2016).

Much of the carbon and nitrogen accruals with DLM application occurred in the sand-POM and silt-MOM

**TABLE 3** Carbon (C) and nitrogen (N) in sand-POM, silt-MOM and clay-MOM fractions in the 0–5-cm layer of a clayey Ferralsol under no-till and application of dairy liquid manure (DML) at rates of 0, 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>, over 9.5 years, and the respective carbon lability index (CLI) and carbon management index (CMI). Castro-PR, Brazil

**FIGURE 2** (a) Macroporosity, (b) microporosity, (c) total porosity, (d) mean weight diameter of water-stable aggregates (MWDw), (e) mean weight diameter of dry-stable aggregates (MWDd) and (f) aggregate stability index (ASI) of a clayey Ferralsol under no-till and subjected to application rates of dairy liquid manure of 0, 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> over 9.5 years. Castro-PR, Brazil. Horizontal bars represent the least significant difference according to Tukey's test ( $p < .05$ , except for microporosity at depth 10–20 cm,  $p < .10$ ); ns not significant



fractions (Table 3). As plant and manure inputs into the soil are essentially organic fragments of plant material, it was predictable that the first and largest increase in carbon and nitrogen stocks was observed in the sand-POM fraction. That is desirable because POM plays an important role as an easily available source of food and energy for soil microorganisms and fauna, in nutrient cycling and crop production and, ultimately, in soil quality (Gregorich et al., 1994). The CMI was influenced by the DLM rates (Table 3) and this carbon index could be considered a good indicator of soil quality (Wenhai et al., 2016) because it was able to show soil carbon changes as a result of the management system (Ghosh et al., 2016). For silt-MOM fractions, the significant increment of carbon and nitrogen stocks is possibly explained by the fact that this fraction is also partially composed of particulate organic matter (Dieckow et al., 2005). As the coarse POM undergoes breakdown, possibly its smallest fragments fall into the silt-size class. In the clay-MOM fraction, the significant increment of nitrogen, but not carbon, may be

related to the nature of the stable organic matter in this fraction, which is basically of microbial origin (Cotrufo, Wallenstein, Boot, Deneff, & Paul, 2013) and so more enriched in nitrogen than the original organic matter. However, significant increments of carbon may also occur in this fraction beyond the term of our experiment (10 years).

In general, the application of DLM improved the soil physical-structural attributes (bulk density, porosity and aggregates) mostly in the 10-cm topsoil (Figure 1d and Figure 2) in a direct response to the incremental increases in soil carbon and nitrogen (Table 2), decreasing the risk of water pollution by surface runoff (Gilley & Risse, 2000; Tomer et al., 2016). In a similar study in the same region, but in a silt-clay loam soil, Mellek et al. (2010) also observed that DLM affected those physical attributes. They found that in the top 5 cm of soil with the highest DLM rate (180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>) applied over a 2-year period, the DLM significantly improved the bulk density and macroporosity compared to unamended soil. Manure is a

**TABLE 4** Carbon input via plants (black oats, wheat, maize and soybean) and via dairy liquid manure (DLM) at rates of 0, 60, 120 and 180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup> (split into winter and summer applications) in a clayey Ferralsol under no-till, over 9.5 years (19 cropping seasons). Castro-PR, Brazil

Season	Crop	Plants <sup>a</sup>				DLM				Total (plants + DLM)			
		0	60	120	180	0	60	120	180	0	60	120	180
input (mg C ha <sup>-1</sup> )													
Winter 2006	Black oats	1.85	1.86	2.22	2.01	0.00	1.11	2.21	3.32	1.85	2.97	4.43	5.32
Summer 2006/07	Maize	6.80	7.18	7.39	7.71	0.00	0.92	1.84	2.77	6.80	8.10	9.24	10.48
Winter 2007	Black oats	0.37	0.43	0.42	0.36	0.00	0.53	1.06	1.59	0.37	0.96	1.48	1.95
Summer 2007/08	Soybean	1.54	1.68	1.87	1.80	0.00	1.09	2.19	3.28	1.54	2.78	4.06	5.08
Winter 2008	Wheat	2.40	2.67	2.70	2.61	0.00	1.06	2.13	3.19	2.40	3.73	4.82	5.81
Summer 2008/09	Soybean	1.75	1.81	1.97	1.83	0.00	0.57	1.14	1.71	1.75	2.38	3.11	3.54
Winter 2009	Black oats	0.32	0.47	0.52	0.55	0.00	0.62	1.25	1.87	0.32	1.09	1.77	2.41
Summer 2009/10	Maize	6.03	6.04	6.58	6.87	0.00	0.55	1.10	1.64	6.03	6.59	7.67	8.51
Winter 2010	Wheat	1.91	2.36	2.49	2.22	0.00	1.04	2.09	3.13	1.91	3.41	4.57	5.36
Summer 2010/11	Soybean	1.74	1.80	1.86	1.93	0.00	1.14	2.27	3.41	1.74	2.93	4.13	5.34
Winter 2011	Black oats	1.14	1.59	1.54	1.42	0.00	1.38	2.76	4.14	1.14	2.97	4.30	5.56
Summer 2011/12	Maize	7.22	7.25	7.01	7.09	0.00	1.15	2.30	3.44	7.22	8.39	9.31	10.54
Winter 2012	Wheat	1.33	1.83	1.97	1.96	0.00	1.07	2.13	3.20	1.33	2.90	4.10	5.16
Summer 2012/13	Soybean	2.05	2.31	2.24	2.21	0.00	0.90	1.80	2.70	2.05	3.21	4.04	4.91
Winter 2013	Black oats	0.92	1.09	1.18	1.08	0.00	0.97	1.93	2.90	0.92	2.05	3.11	3.98
Summer 2013/14	Maize	7.16	7.55	7.99	7.89	0.00	0.87	1.74	2.60	7.16	8.42	9.73	10.49
Winter 2014	Wheat	2.01	2.17	2.23	2.30	0.00	1.15	2.31	3.46	2.01	3.32	4.54	5.76
Summer 2014/15	Soybean	2.17	2.28	2.15	2.19	0.00	1.12	2.23	3.35	2.17	3.40	4.38	5.54
Winter 2015	Black oats	0.92	1.09	1.18	1.08	0.00	0.99	1.98	2.98	0.92	2.08	3.16	4.06
Season mean		2.61	2.81	2.92	2.90	0.00	0.96	1.92	2.88	2.61	3.77	4.84	5.78
Annual mean <sup>b</sup>		5.22 c	5.63 b	5.84 a	5.80 a	0.00	1.92	3.84	5.75	5.22 d	7.55 c	9.60 b	11.56 a

<sup>a</sup>Sum of aboveground (shoot) and root biomass and exudate.

<sup>b</sup>Annual means followed by the same letter are not significantly different (Tukey's test,  $p < .05$ ).

direct source of energy and nutrients for the soil microorganisms, and organic compounds released during decomposition act primarily as cementation agents of macroaggregates. Organo-mineral interactions act on the microaggregates (Bayer, Martin-Neto, Mielniczuk, Dieckow, & Amado, 2006) and the effect of manure on this attribute is expected only in the long term. Perhaps the lack of more significant differences in our study with application over an almost 10-year period was because the positive effects of no-till management per se on improving physical-structural attributes in this clayey soil might have surpassed the effects of DLM amendment.

## 5 | CONCLUSIONS

The application of DLM, over a 10-year period, on the soil surface in a no-till clayey Ferralsol, at rates of 120 or

180 m<sup>3</sup> ha<sup>-1</sup> year<sup>-1</sup>, increased the TOC and TN stocks in the top 10 cm of soil at rates that averaged 0.72 Mg C ha<sup>-1</sup> year<sup>-1</sup> and 86 kg N ha<sup>-1</sup> year<sup>-1</sup>. Much of this increase occurred in sand-POM and silt-MOM size fractions and was attributed to increases in carbon input directly via manure and indirectly via plants. Soil physical-structural attributes were improved by DLM application in the 10-cm layer following the soil carbon and nitrogen increments. Overall, the application of DLM on the surface of no-till soil proved to be a strategy to ameliorate soil organic matter, an important factor for agronomic (crop productivity) and environmental (water pollution and climate change) features.

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#### DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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