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# Tensile strength of mollisols of contrasting texture under influence of plant growth and crop residues addition



**GEODERM** 

M.E. Carrizo<sup>a,b,1</sup>, C.A. Alesso<sup>a,1</sup>, H.H. Soares Franco<sup>c</sup>, C.J. Bernabé Ferreira<sup>c</sup>, S. Imhoff<sup>a,b,\*</sup>

<sup>a</sup> Facultad de Ciencias Agrarias, Universidad Nacional del Litoral, Kreder 2805, S3080HOF, Esperanza, Argentina

<sup>b</sup> CONICET, Kreder 2805, S3080HOF, Esperanza, Argentina

<sup>c</sup> Departamento de Agronomia, Universidade Estadual de Maringá, Av. Colombo 5790, CEP, 87020-900, Maringá, Brazil

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## ABSTRACT

Tensile strength (TS) of soil aggregates is considered a sensitive and important indicator of the effects of the management practices on soil structure quality, which affects the seed germination and the initial crop growth. However, the influence of plant growth, crop residues addition and the produced aggregating agents on TS has not been widely studied. The objectives of this study were: i) to determine the specific effects of plant growth and different types, rates and location of crop residues in the aggregates tensile strength, and ii) to assess the relationship between the aggregating agents produced by plant growth and crop residues addition and the aggregate tensile strength of soils of contrasting texture. A greenhouse experiment was carried out in pots with a loamy soil (Typic Hapludoll, Santa Isabel series) and a silty-loamy soil (Typic Argiudoll, Esperanza series) under controlled conditions for 112 days. For each soil the following treatments were set up in triplicate: (i) soil type, (ii) with or without plant growth of wheat (Triticum aestivum L.), (iii) with or without residues addition, (iv) location of residues (surface vs. incorporated), (v) wheat vs. soybean (Glycine max L.) residues, and (vi) residue rates (6.3 and 15.7 g of dry matter per pot for wheat, and 6.3 and 18.8 g of dry matter per pot for soybean). Pots were exposed to wetting and drying (W/D) cycles. TS values and aggregating agent's content, i.e., total organic carbon (TOC), particulate organic carbon (POC), hot water extractable carbohydrates (HWEC), dilute acid extractable carbohydrates (DAC), total carbohydrates (TC), total glomalin-related soil protein (T-GRSP), and easily extractable glomalin-related soil protein (EE-GRSP) were measured. TS were significantly lower in the Typic Hapludoll (TS = 39.9 kPa) than in the Typic Argiudoll (TS = 61.6 kPa). TS values were significantly higher in the treatments with plants of wheat than in treatments without plants (49.5 vs. 30.3 kPa in the Hapludoll and 71.2 vs. 50.9 kPa in the Argiudoll). Plant growth increased TS through physical mechanisms, i.e. a greater number of drying-wetting cycles. Also, plant growth increased TS by producing aggregating agents. TS values were not directly affected by the addition of different types, rates and locations of crop residues. However, they increased the content of aggregating agents. Multiple regression analysis showed that TS was significantly related to soil type, TC and T-GRSP. TS increased with increasing TC and T-GRSP. These two variables explained 87% of the model variation. The obtained model provides a basis for understanding which are the most important aggregating agents and, consequently, which are the better management systems to improve o recover the structure quality of soils with different textures.

# 1. Introduction

The intensification of the production systems in the Argentinean Pampas had decreased organic carbon and essential nutrients contents as well as the microbiological activity of the soils. Also the intensification had contributed to soil structure degradation (Ferreras et al., 2009). Soil structure degradation changes soil porosity, which controls water and air transmission and the space in which roots can grow (Oades, 1984). Thus, it causes crop production to be affected (Bronick and Lal, 2005; Whalley et al., 2006; Alvarez and Steinbach, 2009). Soil structure is dependent on the size, stability, distribution and strength of the aggregates as well as on the pore space between and within aggregates (Maa et al., 2015). The study of individual aggregates characteristics, such as water-stable aggregates, mean weight diameter and tensile strength were long used to assess the structural quality of the soils (Kay et al., 1994; Kay and Angers, 1999; Dexter and Watts,

\* Corresponding author at: Facultad de Ciencias Agrarias, Universidad Nacional del Litoral, Kreder 2805, S3080HOF, Esperanza, Argentina.

E-mail address: simhoff@fca.unl.edu.ar (S. Imhoff).

<sup>1</sup> Joint first authors.

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2000; Imhoff et al., 2002; Blanco-Canqui et al., 2005; Zhang et al., 2012). Kay et al. (1994) mentioned that tensile strength is related to the aggregate size distribution after a given energy is applied. Thus, tensile strength yields information on the condition of the quality of a seedbed that is created by tillage (Kay et al., 1994). Also, they indicated that tensile strength is related to the mean weight diameter because both measurements are indicators of the resistance to aggregate fragmentation. According to this author and Dexter (1988a) aggregates tensile strength also affects indirectly the activity of soil organisms and organic matter decomposition because it depends on the microcracks existent inside the aggregates.

Tensile strength of the aggregates is defined as the force per unit of area that is required to cause the disruption of aggregates (Dexter and Kroesbergen, 1985; Dexter and Watts, 2000), and can be determined by a simple test on aggregates of different size (Dexter and Kroesbergen, 1985; Watts and Dexter, 1998). This indicator is manly influenced by intrinsic soil properties, such as soil water content, soil organic carbon, texture and clay mineralogy (Barzegar et al., 1994; Imhoff et al., 2002; Reis et al., 2014). Also, it is influenced by soil management practices, which determine the degree of disturbance caused on the aggregates (Macks et al., 1996; Munkholm et al., 2001; Blanco-Canqui et al., 2005; Blanco-Canqui and Lal, 2008; Tormena et al., 2008). According to Blanco-Canqui and Lal (2006) any mechanical disturbance of the soil is portrayed in the tensile strength of individual aggregates. Thus, this property is considered a sensitive indicator of the management practices effects on soil structure quality (Dexter and Watts, 2000).

The changes with time of soil strength of individual aggregates are mainly caused by external factors, such as climate conditions, or by internal factors, such as the activity of microorganisms and plant roots. Czarnes et al. (2000) demonstrated that plant growth increased the strength of the soil bonded to the roots compared to the strength of the bulk soil counterpart.

Roots affect soil aggregates strength through abiotic and biotic mechanisms. They generate cycles of drying-wetting, create soil pores and channels, and produce physical enmeshment of soil particles (Six et al., 2004). Live roots produce mucilage that acts as agent of soil aggregation. Besides, that substance stimulate the microbial activity because they are essential carbon sources for the microorganisms (Six et al., 2004; Rillig et al., 2015; Erktan et al., 2016). Dead roots and plant residues also stimulate the microbial activity because they are carbon sources as well (Golchin et al., 1994; Rillig et al., 2006; Linsler et al., 2016). Microorganisms produce many extracellular compounds as part of their metabolisms that are considered important agents of soil aggregation (Rillig and Mummey, 2006; Bronick and Lal, 2005). Between them, polysaccharides have long been associated with the stability of soil aggregates (Tisdall and Oades, 1982). More recently, other compounds produced by fungi, such as glomalin, the glomalin-related soil protein (GRSP) and hydrophobins have received attention as agents of soil aggregation (Rillig and Mummey, 2006; Spohn and Giani, 2011). The functions of these binding agents seem to depend on the type of fungi and plant species (Piotrowski et al., 2004). Furthermore, roots residues and microbial debris increase soil total and particulate organic matter that produces binding agents of soil aggregates when decomposed (Six et al., 2004; Bronick and Lal, 2005).

Some researchers have indicated that the production and functions of the total and particulate organic carbon, carbohydrates, and GRSP are strongly conditioned by the interactions between types of soil microorganisms, type, rates and location of added crop residues and the roots activity (Abiven et al., 2007; Guimarães et al., 2009; Reis et al., 2014).

The effect of root activity and crop residue addition on the size aggregate distribution and aggregates stability of silty-loam soils are well studied (Sonnleitner et al., 2003; Denef and Six, 2005; Cosentino et al., 2006; Abiven et al., 2007; Carrizo et al., 2015). Some studies show the individual influence of the aforementioned factors in the aggregates tensile strength. Kay et al. (1994) indicated that soil wetting/

drying cycles induced changes in the aggregates tensile strength. Materechera et al. (1992) and Munkholm et al. (2001) found plant growth and the microbial activity increased the aggregates tensile strength. Hadas et al. (1994) and Blanco-Canqui and Lal (2007) reported that crop residue addition increased aggregates tensile strength. Despite these reports, few have gone in to details with specific effect of plant growth, residue addition and the produced aggregating agents in the tensile strength of freshly formed soil aggregates. Thus, understanding these effects on soil tensile strength is still a challenge. We hypothesized that plant growth and crop residues addition increase tensile strength by increasing particulate organic carbon, carbohydrates and GRSP production, and that the magnitude of the increase depends on soil texture. Hence, the objectives of this study were: i) to determine the specific effects of plant growth and different types, rates and location of crop residues in the aggregates tensile strength, and ii) to assess the relationship between the aggregating agents, produced by plant growth and crop residues, and the aggregate tensile strength of soils of contrasting texture.

#### 2. Materials and methods

#### 2.1. Experimental design and treatments

A greenhouse experiment was carried out with soils of contrasting texture and total carbon organic (TOC) under controlled temperature (15-25 °C) and humidity (50-70%) conditions. As described in the research of Carrizo et al. (2015), the soils used in this experiment were collected from two fields that were managed under long-term no-till (last ten years) with agricultural rotations located in Santa Fe province (Argentina). The soil of one field is classified as Typic Hapludoll, Santa Isabel series (33°93'S, 61°57'W) with loamy texture (16% clay, 43% silt, and 41% sand) and SOC content of 21.1 g kg<sup>-1</sup>. The other is classified as Typic Argiudoll, Esperanza series (31°26'S, 60°56'W) with silty clay loam texture (24% clay, 71% silt, and 5% sand) and SOC content of 15.3 g kg<sup>-1</sup>. Each field was split in 3 sectors. In each sector soil samples (N = 20) was collected in fall of 2012 at the depth of 0–20 cm. Briefly, soil samples were collected using a shovel and then gently crumbled by the natural planes of weakness. After sampling, crop residues and coarse roots were removed, and the soil was air-dried and sieved through a 2 mm sieve. The material smaller than 2 mm from the 20 samples of each sector was bulked to obtain a composite sample (about 100 kg each) that was used to fill in 5-1 pots up to a bulk density of about  $1.3 \,\mathrm{g}\,\mathrm{cm}^{-3}$ . All treatments were applied on each of the 3 replications of each soil type.

For each soil the following treatments were set up in triplicate: (i) soil type, (ii) with or without plant growth of wheat (*Triticum aestivum* L.), (iii) with or without residues addition, (iv) location of residues (surface or incorporated), (v) wheat *vs.* soybean (*Glycine* max L.) residues, (vi) residue rates (6.3 and 15.7 g of dry matter per pot that is equivalent to 0.2 and 0.5 kg of dry matter  $m^{-2}$  for wheat, and 6.3 and 18.8 g of dry matter per pot that is equivalent to 0.2 and 0.6 kg of dry matter  $m^{-2}$  for soybean; where 0.2 kg was considered low rate and 0.5 kg and 0.6 kg were considered high rate) (Fig. 1). The two location of residue were used to simulate the tillage system used in the region studied; *i.e.* no-till and conventional tillage.

Residues were cut down into 1 cm pieces and applied before seeding. In the incorporated crop residues treatments, residues were hand-mixed within the upper 10 cm of the soil. Immediately, in the treatments with plants, pre-germinated seeds were planted and four plants of wheat were allowed to grow per pot (127 plants m<sup>-2</sup>). All necessary nutrients were added through Hoagland solution. The salts used to make up the solution were KNO<sub>3</sub>, Ca(NO<sub>3</sub>)<sub>2</sub>, NH<sub>4</sub>H<sub>2</sub>PO<sub>4</sub>, MgSO<sub>4</sub>, and micronutrient (H<sub>3</sub>BO<sub>3</sub>, MnCl<sub>2</sub>, ZnSO<sub>4</sub>, CuSO<sub>4</sub>, H<sub>2</sub>MoO<sub>4</sub>, iron tartrate) (Hoagland and Arnon, 1950). For each soil, all pots had the same water content at the beginning of the experiment. Then, pots were exposed to wetting and drying (W/D) cycles. Each time that soil



Fig. 1. Summary of treatments applied in each soil.

water content varied from 10% aeration to two-thirds of field capacity, the occurrence of one cycle of W/D was recorded. The duration of each cycle, *i.e.* the number of days elapsed between watering, was also recorded for each pot. According to Imhoff et al. (2016), the values of volumetric water content at saturation, water content at 10% of airfilled porosity and at field capacity were 0.48, 0.38 and 0.36 cm<sup>3</sup> cm<sup>-3</sup> (thus two-thirds of field capacity correspond to 0.24 cm<sup>3</sup> cm<sup>-3</sup>) for the Typic Argiudoll, and 0.46, 0.36 and 0.30 cm<sup>3</sup> cm<sup>-3</sup> (thus two-thirds of field capacity correspond to 0.20 cm<sup>3</sup> cm<sup>-3</sup>) for the Typic Hapludoll, respectively. Pots were weighed every day and watered with deionized water or Hoagland solution until the set value were reached. The amount of required water was calculated from the pot weight at each day, the initial pot weight filled with oven dried-soil, the soil volume and the density of the water.

The experiment was finished at senescence stage to avoid significant root decomposition, which happened at day 112 of the cycle. The aerial biomass was harvested allowing the soil to dry until soil water content was about 20% of water content at saturation. The upper 10 cm layer of each pot was sampled and the soil was gently crumbled by the natural planes of weakness and then, the aggregates were air-dried. Great care was taken to avoid damaging the natural aggregates. One part of the aggregates was used to carry out the tensile strength (TS) tests, and other part was ground and passed through a 2 mm sieve to determine the aggregating agents.

#### 2.2. Tensile strength of aggregates

The TS of aggregates was determined on air-dried aggregates by the indirect tension test (Dexter and Kroesbergen, 1985; Dexter and Watts, 2000). Each test was performed with an electronically controlled loading device at a constant strain rate of  $0.03 \text{ mm s}^{-1}$  until the formation of a continuous tensile crack running approximately between the polar diameters. Each soil aggregate was placed in the most stable position and loaded progressively between a fixed lower plate and an upper parallel mobile plate that was assembled to an electronic load cell

of 20 kg capacity. The electrical output was recorded and transformed to force units by a data acquisition system. A set of soil aggregates (n = 45) sized between 12.5 and 19.0 mm was obtained per pot and subjected to the test; thus, a total of 4860 TS tests (2 soils  $\times$  18 treatments  $\times$  3 replications  $\times$  45 aggregates) were carried out. This size range was chosen partly because the aggregates are easy to handle and measure, and partly because these aggregates must be fragmented by tillage to form an ideal seedbed which is typically composed of aggregates from 1 to 5 mm diameter (Dexter, 1988a; Dexter, 1988b). Once tested, each set of 45 aggregates was oven dried at 105 °C to calculate soil water content.

TS was calculated according to Dexter and Kroesbergen (1985):

$$TS = 0.576* \frac{P}{D^2}$$
 (1)

where: the parameter 0.576 is the proportionality constant, P is the applied force at failure (N) and  $D^2$  is the effective diameter (mm). On the assumption that aggregate density is constant, the effective diameter (*D*) of each aggregate was calculated as in Watts and Dexter (1998):

$$D = D_m \left(\frac{M}{M_0}\right)^{\frac{1}{3}} \tag{2}$$

where: *Dm* is the average diameter of aggregate (mm), which was considered equal to the average of sieves size [(19.0 + 12.5)/2] used for the aggregate selection. *M* is the mass of the individual aggregate (g) and *Mo* the average mass of aggregates in each treatment (g).

# 2.3. Aggregating agents

Total organic carbon (TOC) and particulate organic carbon (POC) contents were measured in triplicate by wet oxidation method as described by IRAM-SAGPYA 29571-2 (2007). First soil samples were sieved through a 2 mm sieve to separate the coarse material. A portion of the material < 2 mm was used to determine TOC. Other portion was

subjected to physical fractionation as described Irizar et al. (2010); it was sieved through a 0.053 mm sieve and the material retained on this sieve was used to determine POC.

Carbohydrate content was determined in triplicate on samples of 1 g according to Puget et al. (1999). Hot water extractable carbohydrates (HWEC) were digested with 10 mL distilled water at 80 °C for 24 h. Dilute acid extractable carbohydrates (DAC) were obtained from a dilute acid hydrolysis, which consisted in treating the soil with 10 mL of 0.5 M H<sub>2</sub>SO<sub>4</sub> at 80 °C for 24 h. Total carbohydrates (TC) involved the processing of soil with 2 mL 12 M H<sub>2</sub>SO<sub>4</sub> for 16 h at room temperature. Subsequently, this mixture was diluted to obtain a concentration of 1 M H<sub>2</sub>SO<sub>4</sub> and then placed at 100 °C for 5 h (Puget et al., 1999). The obtained extracts were used to quantify each fraction of carbohydrate by colorimetry with glucose solution as standards using the sulfuric acidphenol procedure (Dubois et al., 1956). Glomalin is a rich mixture of proteinaceous, humic, lipid and inorganic substances (Gillespie et al., 2011; Wright and Upadhyaya, 1996). As the glomalin fraction gained by high temperature extraction is not completely pure, it is addressed as glomalin-related soil protein (GRSP). This designation is used since the Bradford method is not a specific test for a particular protein (Rillig, 2004). Total GRSP (T-GRSP) and easily extractable fraction (EE-GRSG) were determined by the method described Wright and Upadhyaya (1996). The extraction of T-GRPS was performed on 1 g samples by using 50 mM sodium citrate pH 8.0. This mixture was autoclaved at 121 °C for 60 min several times until obtaining a clear color extract. Finally, the extract was centrifuged at  $10,000 \times g$  for 20 min. The extraction of EE-GRSP was performed on 1 g samples by using 20 mM sodium citrate pH 7.0. This mixture was autoclaved at 121 °C for 30 min only one time. Quantification of both fractions were analyzed for protein by the Bradford protein assay using bovine serum albumin as standard (Bradford, 1976).

#### 2.4. Statistical analysis

Tensile strength data were analyzed as split-plot design with soil types as the main plot and pots as subplots. A mixed model was fitted with treatments as fixed effects and replications as random effect. An additional classification factor was added to include the contrast "crop residue vs. control" because the levels of the crop residue factor are an augmented factorial with combinations of type, location and rate of crop residues plus a control following the approach proposed by Piepho et al. (2006). The main effects and interactions of soil type, plant growth and crop residues addition on TS were assessed at a significance level of 5% by ANOVA with Satterthwaite approximation of denominator degree of freedom. The normality and homoscedasticity of the residuals were checked by Shapiro-Wilks and Levene tests. Differences of aggregating agents between soils were determined by independent ttest. The relationship between TS and aggregation agents was modeled by stepwise multiple regression analysis. Multicollinearity was assessed using the variance inflation factor (VIF) according to Neter et al. (1996) to avoid including highly correlated independent variables into the model. The relationship between TS and mean weight diameter, measured in the same condition by Carrizo et al. (2015), was assessed by regression analysis. Details are given in Carrizo et al. (2015). All statistical analysis were performed using the statistical package R (R Development Core Team, 2015).

# 3. Results and discussion

# 3.1. Descriptive statistics and analyses of variance of tensile strength of aggregates

The mean values of TS per treatment for each soil are shown in Fig. 2.

The results of ANOVA showed that differences of TS between treatments were significantly accounted (p < 0.001) by the main

effects of plant growth and soil type, whereas the main effect of crop residue addition was not significant. All interactions between the factors studied were not significant.

The TS mean value of all treatments with plants was significantly higher than the mean value of all treatments without plants in both soils. Plants can increase the TS of aggregates that are adjacent to the roots through abiotic mechanisms associated with the intensity and duration of the drying cycles.

The number of drying–wetting cycles and the mean duration of each cycle are shown in Figs. 3 and 4. The number of drying–wetting cycles and the mean duration of each cycle in treatments with plants was higher than in treatments without plants. The number of cycles and their duration was similar between soil types, indicating that the presence of plant growth was more important than soil texture. Soil drying increases the cohesion between soil particles by increasing the contact points and capillary forces, which in turn increase TS in the failure zones (Kemper and Rosenau, 1984; Horn and Dexter, 1989; Kay and Angers, 1999; Czarnes et al., 2000). Materechera et al. (1992) and Munkholm et al. (2001) found that the increase of soil strength was caused by the combined effect of drying–wetting cycles, root exudates and microorganisms influence, which is in agreement with our results.

The treatments with addition of different types, rates and location of crop residues (without plant growth) had no-significant direct effect on TS. Hadas et al. (1994) in a laboratory experiment found that the addition of cotton residues to a disaggregated silty-loam soil had indirectly increased TS by increasing the amount of fungal mycelia. In field experiments, Blanco-Canqui and Lal (2007) verified that the annual application of different rates of wheat straw for 10-years on a siltyloam soil produced higher TS values. The increase of TS was attributed to the release of organic compounds, humic acids and polysaccharides from residues. Blanco-Canqui and Lal (2008) observed that corn (Zea mays L.) residues removal produced lower TS values by reducing residues-derived organic substances. According to Puget et al. (1999), exudates produced during plant residues decomposition contribute to aggregate soil particles more slowly than root exudates. Therefore, the lack of direct effects of the residues on TS could have been caused by the short period of residues decomposition, i.e. a crop cycle, which could have been insufficient to increase TS.

#### 3.2. Plant growth effect on the aggregating agents

Plant roots release exudates that act as aggregating agents of soil particles. They also stimulate the microbial activity. Microorganisms produces substances that also act as aggregating agents (Puget et al., 1999). According to the MANOVA analysis plants affected significantly and positively the aggregating agents contents (POC, DAC, TC, GRSP, EE-GRSP) (Wilks' lambda = 0.16; F(6,47) = 41.01; p < 0.0001 for the Hapludoll and Wilks' lambda = 0.26, F(6,47) = 22.09; p < 0.0001 for the Argiudoll) with the exception of HWEC (Table 1).

These results suggest that the aggregating agents produced by roots and microorganisms were responsible for the increase of TS (biotic mechanism). Angers et al. (1993) also found that proportional changes in DAC were greater than changes in HWEC.

#### 3.3. Crop residues effect on the aggregating agents

Residues are decomposed by microorganisms and fungi that release organic compounds, humic acids and polysaccharides, which in turn increase the strength of the soil aggregates.

The MANOVA analysis showed that the contents of all aggregating agents were significantly higher in the treatments with residues and without plant growth than in the treatment without residues and without plant growth (POC, HWEC, DAC, TC, GRSP, EE-GRSP) (Wilks' lambda = 0.03; F(12,38) = 15.5; p < 0.0001 for the Hapludoll; and Wilks' lambda = 0.06, F(12,38) = 10.02; p < 0.0001 for the Argiudoll) (Table 2).



Fig. 2. Mean values of tensile strength of the aggregates (TS, kPa) for the different treatments with plant growth and crop residues addition for a Typic Hapludoll and a Typic Argiudoll. Bars represent the standard errors.

The increase of the rates of residues increased the contents of all aggregating agents in both soils. The comparison between residues types (soybean vs. wheat) showed that treatments with soybean residues had greater content of DAC and HWEC in the Hapludoll and greater content of DAC in the Argiudoll (Wilks' lambda = 0.007; F (12,38) = 34.76; p < 0.0001 for the Hapludoll, and Wilks' lambda = 0.005, F(12,38) = 39.97; p < 0.0001 for the Argiudoll). These results may be related to the different chemical composition of the residues. It is well known that soybean residues have lower carbon:nitrogen relationship than wheat residues, i.e. soybean residues have greater proportion of easily degradable carbohydrates than wheat residues, which corroborates the results obtained. In both soils, the comparison between residues location showed that the contents of some aggregation agents (DAC, TC and GRSP) were significantly higher when soybean and wheat residues were incorporated into of soils (Wilks' lambda = 0.16; F(6,47) = 41.01; p < 0.0001 for the Hapludoll, and Wilks' lambda = 0.05, F(6,47) = 11.33; p < 0.0001 for the Argiudoll). One possible explanation for these results is that microorganisms can decompose the residues more easily when residues are in close contact with the soil.

According to Blanco-Canqui and Lal (2007), residues decompose

and reload slowly the soil with organic agents, which increase gradually TS over time. Our results are someway in agreement with the findings of these authors. Residues addition in treatments without plant growth induced an increase in the content of soil aggregating agents even though a direct effect of them on TS could not be verified. These suggest that the duration of the experiment or the rates of residues were not enough for the microorganisms release aggregating agents in abundant quantity to verify a direct independent effect of the residues on TS.

# 3.4. Relation between aggregates tensile strength and the aggregating agents grouping all treatments

## 3.4.1. Descriptive statistics of aggregating agents grouping all treatments

Previous results show that both plant growth and residue addition had individually induced an increase in the content of aggregating agents. Therefore, statistical analyzes were performed grouping all treatments. Descriptive statistics of aggregating agents grouping all treatments for each soil are shown in Table 3.

In general, aggregating agents' contents in the Typic Hapludoll were higher than in the Typic Argiudoll, except the concentration of EE-GRSP. We attributed this result to both, the features of the experiment



Fig. 3. The number of drying-wetting cycles and the mean duration of each cycle (days/cycle) for the different treatments with plant growth and crop residues addition for a Typic Hapludoll and a Typic Argiudoll. Bars represent the standard errors.

and the extraction method. Wu et al. (2014) mentioned that the T-GRSP was recently divided into two fractions. One fraction (EE-GRSP) is more labile and corresponds to material of recent deposition. The other fraction is more difficultly-to-extract and older. The lack of differences in the content of EE-GRSP between soils may have been caused because in both soils the same amounts of residues were applied, independently of the fact that they differed in the content of colloids and therefore, in the possible contact points between them and EE-GRSP. Also, the Bradford method and ELISA assay may be useful in measuring glomalin pools, when soil organic matter concentrations are low or in controlled experimental conditions (Wright and Upadhyaya, 1999; Lovelock et al., 2004; Rosier et al., 2006). Otherwise, these authors found that under conditions where significant extraneous protein additions occur, such as manure, sewage or litter addition, results may not reflect the correct values of the glomalin pools in soils.

The values of TOC and POC were similar to those found by Ferreras et al. (2009) in soils similar to those of this study. Carbohydrates and T-GRSP contents were similar to those found by Puget et al. (1999) and Wright et al. (2007).

3.4.2. Correlation analyses between aggregating agent contents and tensile strength

Correlation analyses were performed to explore the existence of a direct influence of the aggregating agent's contents on TS. Linear correlations coefficients for each soil are shown in Table 4.

In both soils almost all aggregating agents' contents were significantly and positively correlated with TS. Several studies show that TS increases with increasing TOC due to this agent acts as a binding between soil mineral particles (Ekwue, 1990; Blanco-Canqui and Lal, 2007; Guimarães et al., 2009; Reis et al., 2014). Conversely, Dexter (1985) and Zhang (1994) mentioned that TOC can affect TS in other way, especially in coarser textured soils: it may cause an effect of dilution that reduces soil bulk density and increases the aggregate porosity, thus TS decreases. This mechanism seems to be responsible for the relationships found in this study in the Typic Hapludoll, which had higher content of aggregating agents and lower TS.

Tensile strength was significantly and positively correlated with POC only in the Typic Argiudoll. Plante et al. (2006) found that for a given level of carbon input the relationship between mineral surface area and soil organic matter varies with texture. Additionally, different organic carbon pools can saturate soils at different rates. This behavior



Fig. 4. The mean duration of each cycle (days/cycle) for the different treatments with plant growth and crop residues addition for a Typic Hapludoll and a Typic Argiudoll. Bars represent the standard errors.

was associated with the size of the aggregates and the silt + clay content (Gulde et al., 2008). In this study, differences in silt + clay content between the soils could have determined a different carbon saturation behavior and thus, different relationships between POC and TS. The influence of TOC and POC on TS can be very variable due to these agents are conditioned by several factors, such as degree of

humification (Ekwue, 1990; Zhang, 1994), physical and chemical TOC and POC stage (Zhang, 1994; Reis et al., 2014), soil management (Blanco-Canqui et al., 2005), soil texture and mineralogy (Imhoff et al., 2002). In these studies significant relationships between TS and soil physical and chemical properties, such as organic matter and texture, were found (Materechera et al., 1992; Barzegar et al., 1994; Imhoff

#### Table 1

Mean values and standard deviation of particulate organic carbon (POC), hot water extractable carbohydrates (HWEC), dilute acid carbohydrates (DAC), total carbohydrates (TC), total glomalin-related soil protein (T-GRSP) and easily extractable fraction (EE-GRSP) for the treatments with and without plant growth for a Typic Hapludoll and a Typic Argiudoll.

Variables	With plant growth	Without plant growth	With plant growth	Without plant growth
	Typic Hapludoll		Typic Argiudoll	
POC (g kg <sup>-1</sup> ) HWEC (mg C kg <sup>-1</sup> ) DAC (mg C kg <sup>-1</sup> ) TC (mg C kg <sup>-1</sup> ) T-GRSP (mg g <sup>-1</sup> ) EE-GRSP (mg g <sup>-1</sup> )	$\begin{array}{l} 4.65  \pm  0.48  \mathrm{a} \\ 45.83  \pm  9.60  \mathrm{a} \\ 1189  \pm  108.46  \mathrm{a} \\ 1398  \pm  86.79  \mathrm{a} \\ 3.71  \pm  0.14  \mathrm{a} \\ 1.22  \pm  0.05  \mathrm{a} \end{array}$	$\begin{array}{l} 4.32  \pm  0.48 \ \mathrm{b} \\ 42.95  \pm  9.23 \ \mathrm{a} \\ 940  \pm  221.67 \ \mathrm{b} \\ 1242  \pm  112.50 \ \mathrm{b} \\ 3.43  \pm  0.15 \ \mathrm{b} \\ 1.01  \pm  0.07 \ \mathrm{b} \end{array}$	$3.73 \pm 0.28$ a $30.43 \pm 7.75$ a $919.9 \pm 101.60$ a $1111 \pm 105.73$ a $1.93 \pm 0.29$ a $1.18 \pm 0.06$ a	$\begin{array}{l} 3.21 \ \pm \ 0.26 \ \mathrm{b} \\ 27.40 \ \pm \ 6.04 \ \mathrm{a} \\ 730.33 \ \pm \ 91.20 \ \mathrm{b} \\ 985 \ \pm \ 58.58 \ \mathrm{b} \\ 1.63 \ \pm \ 0.25 \ \mathrm{b} \\ 1.01 \ \pm \ 0.07 \ \mathrm{a} \end{array}$

Different letters indicate differences between treatments in each soils (t-test,  $\alpha < 0.05$ ).

#### Table 2

Mean values and standard deviation of particulate organic carbon (POC), hot water extractable carbohydrates (HWEC), dilute acid carbohydrates (DAC), total carbohydrates (TC), total glomalin-related soil protein (T-GRSP) and easily extractable fraction (EE-GRSP) for the treatments without addition of crop residues and with addition of soybean and wheat residues (without plants) for a Typic Hapludoll and a Typic Argiudoll.

	POC (g kg $^{-1}$ )	HWEC (mg C kg $^{-1}$ )	DAC (mg C $kg^{-1}$ )	TC (mg C kg <sup><math>-1</math></sup> )	T-GRSP (mg $g^{-1}$ )	EE-GRSP (mg $g^{-1}$ )
Treatment			Typic Hapludoll			
WCR	$3.33 \pm 0.06b$	$28.13 \pm 0.55b$	636.3 ± 8.08b	1059.66 ± 21.54b	$3.07 \pm 0.06b$	$0.84 \pm 0.03b$
Wheat	$4.45 \pm 0.40a$ $4.31 \pm 0.29a$	$44.91 \pm 8.32a$ $44.69 \pm 8.02a$	$1118 \pm 53.19a$ 839 ± 205.53a	$1290.9 \pm 78.39a$ 1240.3 ± 109.58a	$3.48 \pm 0.08a$ $3.48 \pm 0.06a$	$1.03 \pm 0.04a$ $1.04 \pm 0.03a$
			Typic Argiudoll			
WCR	$2.89 \pm 0.06b$	$24.34 \pm 0.92b$	$629.3 \pm 7.64b$	889.7 ± 14.50b	$1.11 \pm 0.02b$	$0.82 \pm 0.03b$
Soybean	$3.32 \pm 0.30a$	31.87 ± 5.61a	786.7 ± 65.29a	1008 ± 33.51a	$1.62 \pm 0.19a$	$0.03 \pm 0.03a$
Wheat	$3.19~\pm~0.17ab$	$23.76~\pm~3.99b$	$699.2 \pm 89.85b$	986.1 ± 62.39ab	$1.56 \pm 0.14a$	$1.01 \pm 0.03a$

WCR = without crop residues. Different letters indicate differences between treatments in each soils (*t*-test,  $\alpha < 0.05$ ).

#### Table 3

Descriptive statistics of total organic carbon (TOC), particulate organic carbon (POC), hot water extractable carbohydrates (HWEC), dilute acid carbohydrates (DAC), total carbohydrates (TC), total glomalin-related soil protein (T-GRSP) and easily extractable fraction (EE-GRSP) grouping all treatments for the Typic Hapludoll and the Typic Argiudoll.

Variables	Mean	SD	Min.	Max.	CV (%)
Organic carbon (g kg <sup>-1</sup> ) TOC					
Typic Hapludoll	21.8a	7.0	20.1	23.3	3.8
Typic Argiudoll	17.8b	8.0	16.0	19.5	4.4
POC					
Typic Hapludoll	4.5a	0.5	3.3	5.4	11.2
Typic Argiudoll	3.4b	4.0	2.8	4.2	10.7
Carbohydrates (mg C kg $^{-1}$ )					
HWEC					
Typic Hapludoll	44.4a	9.5	27.5	65.1	21.3
Typic Argiudoll	28.9b	7.1	18.5	44.0	24.4
DAC					
Typic Hapludoll	1065.4a	213.6	629.0	1395.0	20.1
Typic Argiudoll	825.1b	135.3	598.0	1060.0	16.4
TC					
Typic Hapludoll	1320.8a	126.9	1043.0	1493.0	9.6
Typic Argiudoll	1048.1b	105.9	875.0	1345.0	10.1
Glomalin (mg $g^{-1}$ )					
T-GRSP					
Typic Hapludoll	3.6a	0.2	3.0	4.0	5.6
Typic Argiudoll	1.8b	0.3	1.1	2.3	17.4
EE-GRSP					
Typic Hapludoll	1.1a	0.1	0.8	1.3	10.8
Typic Argiudoll	1.1a	0.1	0.8	1.2	9.7

SD = Standard deviation, Min = minimum, Max = maximum, CV = coefficient of variation (%). Different letters indicate differences between soils *t*-test.

et al., 2002; Blanco-Canqui et al., 2005; Tormena et al., 2008; Reis et al., 2014). Our findings agree with the results obtained by these authors.

Dexter and Kroesbergen (1985) stated that TS variation may also arise because of the aggregate shape variability, which is caused by variations in texture and organic matter content. Aggregates obtained from the Typic Hapludoll were more spherical (rounded blocks) than those obtained from the Typic Argiudoll (subangular-angular blocks). The Typic Hapludoll has greater proportion of large primary particles (>  $53 \,\mu$ m) and smaller proportion of silt content and, consequently, has fewer number of contact points between the soil particles when compared to the Typic Argiudoll. Taboada et al. (2008) studied soil cracking and clod shrinkage of some silty loam soils of Argentina that have < 10% of coarse-grained particles. They found that these soils have aggregates with low structural porosity and few fine cracks. Both characteristics cause soils to have high TS due to this property is very sensitive to the presence of microcracks within the aggregates (Kay and Dexter, 1992). Thus, ours results are consistent with the findings of the mentioned authors and explain why the Typic Argiudoll have higher TS values than the Typic Hapludoll.

#### 3.4.3. Effects of the aggregating agents and soil type on tensile strength

In this study, a model to predict TS from the aggregating agents was developed for both soils using multivariable regression analysis. Each soil was included as a discrete variable (Hapludoll = 0; Argiudoll = 1). The parameters of the model are shown in Table 5.

The obtained model explained 87% of the data variability (F = 228.85, P < 0.0001). The *a* parameter varied significantly with soil, which means that both soils differ in the initial TS values probably because of the differences in silt + clay and organic carbon content. Tensile strength was positively related to TC and EE-GRSP. Aggregating agents associated with polysaccharide released by plants and fungi showed the highest correlation coefficients whereas the other agents show moderate or weak correlation coefficients (Table 4), explaining the model obtained. Calculated mean value of TS for the Hapludoll was 39.9 kPa whereas for the Argiudoll was 61.6 kPa. The presence of active roots and crop residues increase the microorganisms' activity, which in turn increases the TC and EE-GRSP production. Ours results indicates that these aggregating agents were mainly responsible for the increase of TS in the soils studied.

According to Chenu and Cosentino (2011) the effects of carbohydrates on the aggregation process are associated with their strong affinity to adsorb to mineral surfaces because of their surface reactivity. As a result, they bridge soil particles and aggregates together. A similar function accomplishes the glomalin-related soil proteins because they contain N-linked oligosaccharides and iron that act as bonds between soil particles (Wright and Upadhyaya, 1996; Chenu and Cosentino,

#### Table 4

Correlation coefficients among tensile strength of the aggregates (TS), total organic carbon (TOC), particulate organic carbon (POC), hot water extractable carbohydrates (HWEC), dilute acid carbohydrates (DAC), total carbohydrates (TC), total glomalin-related soil protein (T-GRSP) and easily extractable fraction (EE-GRSP) for the Typic Hapludoll and the Typic Argiudoll.

	TOC	POC	HWEC	TC	DAC	T-GRSP	EE-GRSP
Typic Hapludoll	0.51***	0.20 <sup>NS</sup>	0.10 <sup>NS</sup>	0.52***	0.43**	0.58***	0.59***
Typic Argiudoll	0.38**	0.35**	0.07 <sup>NS</sup>	0.30*	0.47***	0.21 <sup>NS</sup>	0.60***

NS not significant, \*P < 0.05 probability level, \*\*P < 0.01 probability level, \*\*\*P < 0.001 probability level.

#### Table 5

Estimates, standard errors, t values, probabilities and variance inflation factor (VIF) of the coefficients of the multiple regression analysis for the tensile strength of the aggregates (TS, kPa).

Variable	Parameter estimate	Standard error	T value	$Pr  >  \left  t \right $	VIF
Intercept	- 59.805	6.149	-9.72	< 0.0001	0
Soil	27.820	1.998	13.93	< 0.0001	3.626
TC	0.022	0.007	3.27	0.0015	5.080
EE-GRSP	63.181	6.787	9.31	< 0.0001	2.148

Soil: Hapludoll = 0, Argiudoll = 1, TC = total carbohydrates, EE-GRSP = easily extractable glomalin-related soil proteins.

2011; Carrizo et al., 2015). Therefore, our results confirm the finding of these authors. Also they highlight that the effect of the aggregating agents on soil aggregation may be evaluated through the TS indicator.

Moreover, we have previously reported (Carrizo et al., 2015) that the mean weight diameter (MWD) of aggregates was related to EE-GRSP, HWEC and TC. The EE-GRSP fraction decreased all aggregate breakdown mechanisms whereas HWEC and TC decreased slaking and microcracking of aggregates. We verified the existence of significant correlations between TS and the MWD obtained by Carrizo et al. (2015) for the same soils. We found that TS was correlated with the MWD after applying fast wetting (r = 0.66, p < 0.0001; r = 0.52, p = 0.001) and slow wetting (r = 0.50, p = 0.002; r = 0.50, p < 0.0001) in the Typic Hapludoll and Typic Argiudoll, respectively. These results indicate that EE-GRSP and TC not only increased the aggregates stability of aggregates sized between 2 and 5 mm (Carrizo et al., 2015) but also the TS of the macroaggregates in both soils. EE-GRSP and TC increased the bindings between soil particles leading to the formation of microaggregates and also the bindings between the microaggregates leading to the formation of stable macroaggregates. Besides, values of TS in both soils were within the common range mentioned by several authors, which agrees with a condition of friable soil (Kay et al., 1994; Imhoff et al., 2002). As a result, macroaggregates will better withstand the action of disruptive forces that produce disaggregation, such as tillage operations, maintaining a suitable aggregate size for roots and plants growth in both soils.

Models, such as the one found in this study, can be a useful tool to better understand which are the most important agents involved in the formation of strong aggregates of soils with different texture. This knowledge may allow establishing the better management system to improve soil aggregation. The formation of a good seedbed with one pass tillage or with direct seeding requires that large aggregates or the soil bulk breaks down into aggregates of suitable size for crop establishment. Because of TS is considered a sensitive indicator of the management practices effects on soil structure, differences in aggregates TS have important consequences in determining soil response to tillage. Soils with very high TS require improving soil structure before trying direct seeding to avoid failure in crop establishment (Macks et al., 1996) because of high TS values correspond to a mechanically unstable condition according to the friability classification (Imhoff et al., 2002). In the Typic Argiudolls of the Flat Pampas is very common to observe failure in seed germination and initial growth of the crops. Therefore, the reduction of the TS values should be one of the main purposes of farmers to achieve successful establishment of crops. Our results indicate that management systems that produce large volume of residues and roots and, consequently, large volume of aggregating agents released by roots and microorganisms would create a better soil structure than other systems in loam and silty-loam soils.

## 4. Conclusions

Plant growth increased TS through physical (drying–wetting cycles) and chemical mechanisms (production of aggregating agents). The direct influence of crop residues addition on TS was not significant. Nevertheless, plant growth and residue addition increased the content of all aggregating agents. The values of TS were mainly conditioned by the content of carbohydrates and glomalin-related soil proteins in both soils although the soil with higher silt+clay content (Argiudoll) had higher TS demonstrating the additional influence of these mineral fractions. The knowledge of the aggregating agents that control TS can help to determine the best management system to improve soil aggregation. Further studies should focus on the role that different types, rates and location of crop residues have on TS in the long term.

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