

## Article

# Cropping System Stability Drives SOC Sequestration and Increases Saturation Deficit in Hot Arid Durum Wheat Cropping Systems

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**Abstract:** Stability analysis has been extensively employed in evaluating the genotype x environment interaction, but it can also be conveniently applied to cropping systems. This study applied stability analysis to a long-term experiment (1998–2016) conducted in an experimental farm in southern Italy. The experiment involved two standard semi-arid cropping systems (WW—durum wheat monocropping and WP—durum wheat–field pea rotation), with two nitrogen fertilization levels (60 kg ha<sup>-1</sup>—N60 and 120 kg ha<sup>-1</sup>—N120), along with an unfertilized control (N0). Soil organic carbon (SOC) was estimated in all treatments at the end of the experiment to investigate the potential for SOC sequestration and soil C saturation by analyzing the stability of all investigated cropping conditions in relation to SOC sequestration. The study also investigated the relationship between C input and SOC sequestration. SOC sequestration per year was higher in WW than in WP, confirming that including legumes in the rotation did not enhance the SOC content. Similarly, the positive impact of nitrogen application on biomass yield did not affect SOC accumulation. The stability analysis revealed that over the long term, WW exhibited greater stability than WP, suggesting that, in semi-arid environments, a more stable cropping system can enhance soil carbon sequestration, with nitrogen fertilization exerting a scarce influence. Segmented regression analysis identified two distinct patterns of SOC accumulation based on C input, separated by a definite threshold of C input (breakpoint) quantified at 14.7 Mg ha<sup>-1</sup>. It was found that the steady-state C level, achieved after the accumulation of this C input, was reached much faster in WW than in WP, in which it could only be inferred at 23 years. In conclusion, this study underscores the role of cropping system stability in driving SOC sequestration. Furthermore, it emphasizes the potential of stability analysis in assisting stakeholders in refining their strategies for different cropping conditions in dry and rainy environments.

**Keywords:** SOC saturation; cropping system stability; SOC sequestration rate



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## 1. Introduction

Despite being a relatively small proportion of soil mass, soil organic carbon (SOC) and soil organic matter (SOM) are pivotal determinants of soil quality [1–3]. Qualitative and quantitative alterations in SOM influence physical, chemical, and biological soil properties [4,5]. Significantly, SOM plays a crucial role in SOC dynamics, simultaneously serving as a carbon source for soil processes and as a sink for carbon sequestration [6].

### 1.1. Crop Rotation Influence on SOC

The content of SOC is governed by the quantity and quality of carbon in crop residue [7–9]. Consequently, crop rotation affects the availability of carbon substrates and decomposition rates by controlling the quantity, quality, timing, and annual rate of C inputs [10].

The quality of residues, in terms of nitrogen (N) and carbon (C) content, can influence the rate of biomass decomposition, thereby affecting the transition of carbon into the SOM fraction [11]. Johnson et al. [12] indicated that the combined effect of the total N and C:N ratio of residue significantly influenced the decomposition rate. Since legumes possess residues with a lower C:N ratio than cereals [13], the former exhibit faster residue decomposition than the latter [10].

Roots also contribute to SOC, along with aboveground plant material. Differences in root mass, quality, and rhizodeposition could lead to disparities in SOC. In fact, cereals return more root biomass to the soil than legumes [14].

### 1.2. Effect of N Fertilizer Application on SOC

Nitrogen (N) fertilization is widely acknowledged as a critical factor in determining crop yields [2,15]. Fertilized fields typically exhibit greater crop yields and produce more residues returned to the soil, contributing to increased SOC [16]. Alvarez [17] observed that carbon sequestration increased with higher nitrogen application across various climates. However, excessive nitrogen input can reduce carbon input in dry and tropical climates due to insufficient soil water. In contrast, Khan et al. [18] reported that SOC concentrations decreased with increasing applications of inorganic nitrogen fertilizer. Despite higher residue input in NPK-fertilized plots, the SOC concentration declined over time, especially in plots with the highest fertilizer rates. Hence, the increased heterotrophic decomposition of SOC due to nitrogen additions led to declining SOC concentration.

Contrary to the apparent positive response to nitrogen addition in aboveground biomass, the response of belowground plant tissues is less straightforward. The addition of nitrogen can change biomass partitioning in plants [19], as an external N addition alleviating nutrient deficiency may induce plants to allocate more fixed carbon to aboveground parts compared to belowground parts [20]. Several studies have supported the optimal partitioning theory by demonstrating altered biomass allocation in plants due to N fertilization; while the production of leaves, wood, and coarse roots increases, fine roots decrease [21,22].

### 1.3. Effect of Soil Aggregates Stability on SOC

Soil aggregates are pivotal in SOC accumulation and dynamics [3]. Organic matter is stored within soil macroaggregates, contributing to their binding and stabilizing effect [3]. These macroaggregates, in turn, can delay organic matter decomposition [23]. Consequently, practices and conditions that enhance the stability of macroaggregates can lead to increased SOC storage, possibly accounting for the lower carbon amounts found in disturbed soils. Coppens et al. [23] observed an increased proportion of soil macroaggregates and higher SOC stocks in plots subjected to rotary tillage and no-tillage compared to those using a moldboard plow. Similarly, Ardenti et al. [24] linked higher SOC accumulation in no-tillage conditions to increased macroaggregate formation and associated carbon and nitrogen content within macroaggregates.

The size distribution of soil aggregates emerges as a crucial factor determining SOC accumulation, as it sets the maximum level of carbon that can be stored in a given soil at a particular time (the carbon saturation point) [25].

### 1.4. SOC Dynamics and Carbon Inputs Stability

Enhancing carbon inputs ( $C_i$ ) to the soil is widely accepted as a strategy for sequestering SOC [26]. However,  $C_i$  alters a series of turnover processes that induce changes in SOC dynamics [27]. The decomposition and transformation processes respond differently to new carbon inputs, influenced by soil properties and the existing carbon input level, quantity, and quality.

Given these complexities, the long-term dynamics of SOC and the carbon input requirements need careful consideration in SOC sequestration targets like the “4 per 1000” initiative [28]. Identifying the required carbon input to achieve SOC saturation targets is crucial in determining site-specific, suitable carbon input management strategies.

In this context, although SOC sequestration in response to the quality and quantity of carbon input has been extensively studied [29,30], the effects of carbon input stability remain largely unexplored.

The primary objective of this study was to:

(i) ascertain the potential for SOC sequestration and soil carbon saturation of two typical semi-arid cropping systems in relation to carbon input stability.

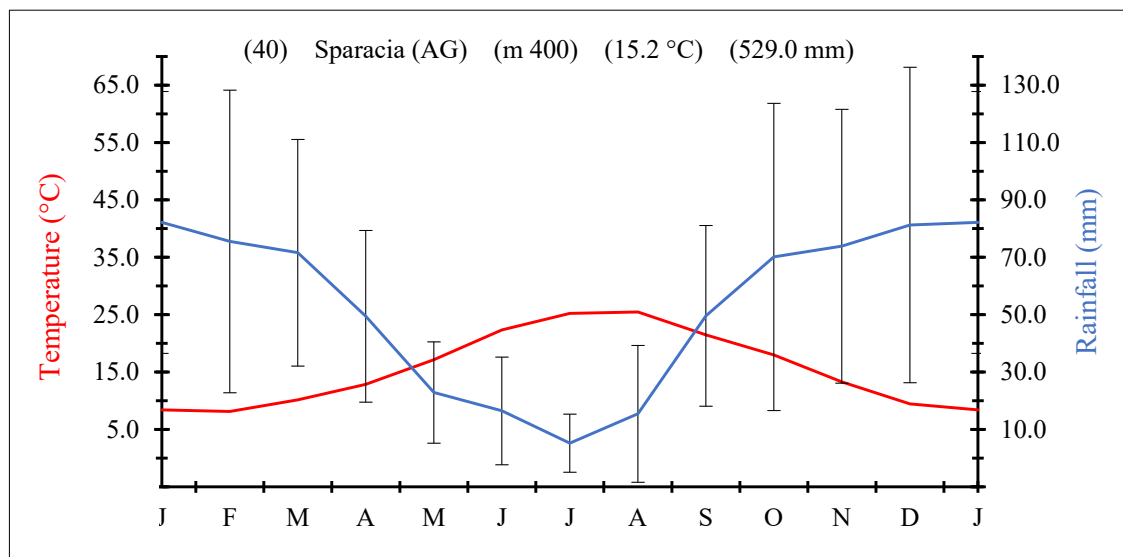
A secondary objective was to:

(ii) quantify the relationship between carbon input and SOC sequestration in bulk soil and SOM fractions concerning different cropping systems and N fertilization rates.

## 2. Materials and Methods

### 2.1. Study Area and Experimental Design

A long-term experiment spanning from November 1998 to June 2016 (18 consecutive cropping seasons) was established at the “Sparacia” experimental farm (Cammarata, AG, Italy; 37°37'74" N, 13°42'53" E; 400 m a.s.l.). The soil is a sub-alkaline Eutric Vertisol [31] with a 6% slope. The soil's textural class is clayey (57.7% clay, 16.2% silt, 26.1% sand). The mean annual precipitation is 529 mm, and the annual maximum and minimum mean air temperatures are 21.4 °C and 9.0 °C, respectively (Figure 1).



**Figure 1.** Ombrothermic diagram according to Bagnouls and Gaussen. Average 40-year data (1967–2006). Vertical bars are standard deviations for rainfall data.

The trial was carried out using a split-plot design with three replications. The main plots featured two cropping systems: durum wheat monocropping (WW) and durum wheat–field pea rotation (WP). Sub-plots involved two nitrogen fertilization levels ( $60 \text{ kg ha}^{-1}$ —N60, and  $120 \text{ kg ha}^{-1}$ —N120), plus an unfertilized control (N0). The main plot size for the cropping systems was  $75 \text{ m} \times 5 \text{ m} = 375 \text{ m}^2$ , while each sub-plot for the N treatment was  $5 \text{ m} \times 5 \text{ m} = 25 \text{ m}^2$ . For data collection and soil sampling, a  $16 \text{ m}^2$  sample area ( $4 \text{ m} \times 4 \text{ m}$ ) was used, excluding a 0.5 m wide area on each elementary plot's side. Wheat and pea were sown in rows 20 cm apart for wheat and 30 cm for pea. The details of the crop management are provided in Table 1.

**Table 1.** Major information on crop management throughout the 18-year trial.

	Durum Wheat (W)			Field Pea (P)
Genotype	Cv Simeto			Cv Belinda
Soil tillage	Summer tillage (30 cm)—two light tillage pre-sowing			
Fertilization pre-sowing	100 kg ha <sup>-1</sup> of P <sub>2</sub> O <sub>5</sub>			
Fertilization (top-dressing; N kg ha <sup>-1</sup> )	N0	N60	N120	---
Fertilization time	3–5 leaves stage (Zadock's scale Z13–Z15)			
Sowing time	From the last decade of November to the first decade of December			
Seed density	350 germinable seeds m <sup>-2</sup>		50 germinable seeds m <sup>-2</sup>	
Harvest time	End of June			
Weed control	Manual			
Elementary plot size	25 m <sup>-2</sup>			

## 2.2. Soil Sampling, Fractionation, and Soil Analysis

At the beginning of the trial (November 1998), three soil samples were randomly collected from the selected experimental area at 0–30 cm depth before sowing. These samples were used for determination of initial soil organic content (SOC<sub>0</sub>) by means of the Walkley–Black titration method [32].

In the last trial year, after crop harvesting (June 2016), three soil samples were collected from each plot at 0–30 cm depth. These samples were air-dried, mixed, and sieved at 1 mm. Mechanical shaking of 50 g air-dried fine soil, using a Shaker AS 200 Sieve (RETSCH analytical-203 mm diameter sieves; an amplitude of 2 cm, frequency of 1.6 Hz, and water flux of 2 L min<sup>-1</sup>) on a column with sieves of >75 µm, 75–25 µm, and <25 µm, was used to isolate wet aggregate-size fractions. After fractionation, three main fractions were separated: >75 (macroaggregates), 25–75 µm (microaggregates), and <25 µm (silt and clay fraction). The weight distribution of the soil fractions and carbon content was measured. The soil organic content at the end of the trial (SOC<sub>t</sub>) was assessed through the previously used Walkley–Black method [32]. Furthermore, the bulk density (g cm<sup>-3</sup>) was measured using the tube core method [33], based on the ratio between the volume of the collected sample (5 × 15 cm cylinder) and the dry weight (g) of the soil sample.

## 2.3. C Input Estimation

In each trial year, the weight of the aboveground biomass harvested from one square meter was used to estimate the total aerial biomass in each elementary plot. Thus, 18 elementary samples (3 N levels × 2 Cropping Systems × 3 replicates) were obtained in every year of experimentation, and used for the subsequent determinations.

Wheat and pea straws were removed according to local practices, and only the root biomass and a portion of the stem (10% of the total aboveground biomass) left after harvesting were considered as carbon input to the soil [34,35].

Assuming, for both species, a 40% carbon content on dry matter, a root-to-shoot ratio (Krs) of 0.24 and 0.18 was used for wheat and pea, respectively, based on average estimates used in similar works for legumes and cereals [36].

## 2.4. Data Calculation

C input (C<sub>i</sub>) to the soil, based only on root biomass, was calculated according to Kong et al. [34] and Chung et al. [37] as follows:

$$C_i \left( \text{Mg ha}^{-1} \text{y}^{-1} \right) = Krs \times \text{Aboveground biomass} \left( \text{Mg ha}^{-1} \right) \times \text{Root carbon content}(\%) \times 1.1 \quad (1)$$

where  $K_{rs}$  is the Belowground residue/Aboveground biomass ratio, whereas the coefficient 1.1 takes into account the 10% of the aboveground residue left in the soil.

Cumulative carbon input (CCi) was calculated for each cropping system by summing the Ci values calculated for all the trial years.

SOC stock ( $\text{Mg ha}^{-1}$ ) in the 0–30 cm layer was calculated using the equation:

$$\text{SOC (Mg ha}^{-1}\text{)} = \text{SOC (g kg}^{-1}\text{)} \times \text{BD (Mg m}^{-3}\text{)} \times \text{d (m)}/10 \quad (2)$$

where SOC is soil organic C content, BD is the bulk density, and d is the thickness of the soil layer.

C sequestration efficiency (Cse %) was calculated according to the following equation [34]:

$$\text{Cse} = \text{Ssr}/\text{Ci} \times 100 \quad (3)$$

where Ssr (SOC sequestration rate;  $\text{Mg}^{-1} \text{ha}^{-1} \text{yr}^{-1}$ ) was estimated for the topsoil according to the equation [34]

$$\text{Ssr} = (\text{SOC}_t - \text{SOC}_0)/y \quad (4)$$

In Equation (4),  $\text{SOC}_t$  and  $\text{SOC}_0$  are the stocks of SOC at time t (2016) and at the beginning of the experiment (1998), respectively, and y is the duration of the experiment (years). Positive and negative Ssr values were considered SOC gains or losses for the cropping systems, respectively [38,39].

## 2.5. SOC Saturation Estimation

The potential C saturation deficit ( $\text{SOC}_{\text{sat}}$ ) was calculated as follows:

$$\text{SOC}_{\text{sat}} = \text{SOC}_{\text{sat}<25 \mu\text{m}} + \text{SOC}_{>25 \mu\text{m}} \quad (5)$$

where  $\text{SOC}_{\text{sat}>25 \mu\text{m}}$  is the C content ( $\text{g kg}^{-1}$ ) of coarser soil particles  $> 25 \mu\text{m}$  (%), and  $\text{SOC}_{\text{sat}<25 \mu\text{m}}$  is the potential C saturation ( $\text{g kg}^{-1}$ ) of fine soil particles  $< 25 \mu\text{m}$  (%). This index was calculated according to Hassink's equation [40]:

$$\text{SOC}_{\text{sat}<25 \mu\text{m}} = 1 - \text{SOC}_0/\text{Pc} \quad (6)$$

where  $\text{SOC}_0$  is the SOC content at the beginning of the trial, and Pc is the soil C protective capacity of  $< 25 \mu\text{m}$  particles, obtained as follows:

$$\text{Pc} = 4.09 + 0.37 \times \text{particles} < 25 \mu\text{m} (\%) \quad (7)$$

## 2.6. Stability Analysis

Although yield stability analysis has primarily been used in genotype-environment interaction trials, applying stability analysis methods to cropping systems is not new [41–43]. It has been adopted in long-term experiments to evaluate fertilization treatments [44] and compare different agronomic treatments [45].

The regression method according to Finlay and Wilkinson [46] was used to calculate the stability analysis parameters of the experimental treatments, estimating the angular coefficient of the linear regression of treatment yields on the environment mean yield, i.e., the average yield of all comparable treatments each year. When “year”  $\times$  “treatment” interactions are detected in a conventional ANOVA model, stability analysis can provide a simple method of determining whether this interaction is a function of the environment. According to Finlay and Wilkinson [46], the slope regression (b) is the response of the cropping system to the environmental index derived from the average performance of all cropping systems in each environment. If b does not significantly differ from 1, then the cropping system is adapted to all environments. A  $b > 1$  indicates cropping systems with a higher sensitivity to environmental change and a greater specificity of adaptability to high-yielding environments, while a  $b < 1$  describes a greater resilience to environmental

change. The adaptability to low-yielding environments must be approached as a more constant, albeit low, carbon input into the soil cropping system capacity.

### 2.7. Breakdown Regression

Many studies have stressed critical thresholds associated with ecological studies [47,48]. In general, thresholds occur when the response of an ecological process is not linear but changes at a particular level (breakdown) that can be considered the threshold. Soil organic matter sequestration follows these basic principles, with the threshold after the breakdown considered the soil C steady-state condition [49]. Knowledge of the time over which the SOC steady state occurs could be helpful for SOC prediction and designing environmental policy based on C accounts.

In this study, the regression between soil organic carbon (SOC) and the explanatory variable (cumulative carbon input—CCi) can be divided into two linear relationships at different ranges of CCi when a single linear model is inadequate. The value of the breakpoint (Bp) was estimated as follows:

$$\begin{cases} \text{SOC}_1 = a_1 + b_1\text{CCi} & \text{for } \text{CCi} \leq \text{Bp} \\ \text{SOC}_2 = a_2 + b_2\text{CCi} & \text{for } \text{CCi} \geq \text{Bp} \end{cases} \quad (8)$$

The two equations for SOC need to be equal at the breakpoint (when CCi = Bp):

$$a_1 + b_1\text{Bp} = a_2 + b_2\text{Bp} \quad (9)$$

Solving for  $a_2$ :

$$a_2 = a_1 + \text{Bp}(b_1 - b_2) \quad (10)$$

Then, by replacing  $a_2$  with the equation above:

$$\text{SOC} = a_1 + b_1\text{CCi} \quad (11)$$

for  $\text{CCi} \leq \text{Bp}$ , and

$$\text{SOC} = (a_1 + \text{Bp}(b_1 - b_2)) + b_2\text{CCi} \quad (12)$$

for  $\text{CCi} > \text{Bp}$ .

### 2.8. Statistical Analyses

Data for SOC content, aggregate size, and CCi underwent analysis of variance (ANOVA) based on the experimental layout using the SPSS statistical software version 22 (IBM Corp., Armonk, NY, USA, 2013) [50]. Before the statistical analysis, all data was preliminarily checked for the assumptions of ANOVA [51]. Specifically, variance homogeneity was assessed using Levene's test, and normality was checked through the Shapiro–Wilk test, both implemented in the same statistical package.

Differences between means were further evaluated using Tukey's HSD test at  $p < 0.05$ .

## 3. Results

### 3.1. Carbon Input Influence on SOC Stock

The between-subjects effects ANOVA on the annual carbon input (Aci) showed a high statistical significance only for the "Cropping System" (CS) factor, while the "Nitrogen level" (N) and its interaction with CS showed a lower ( $0.01 < p < 0.05$ ) significance level (Table 2). On the other hand, the within-subjects effects ANOVA (over years analysis) indicated a high significance for all factors, both individually and in interaction. The "year" (Y) factor showed the highest F value (Table 3).

**Table 2.** Between-subjects effects analysis of variance on annual carbon input (ACi).

	SS	d.f.	MS	F	p
N	5.150	2	2.575	4.738	0.030
CS	19.788	1	19.788	36.403	<0.001
N × CS	6.522	2	3.261	5.999	0.016
Residuals	6.523	12	0.544		

N: Nitrogen fertilization; CS: cropping system.

**Table 3.** Within-subjects effects analysis of variance on annual carbon input (ACi).

	SS	d.f.	MS	F	p
Y	35.336	17	2.079	39.175	<0.001
Y × N	10.609	34	0.312	5.881	<0.001
Y × CS	7.587	17	0.446	8.411	<0.001
Y × N × CS	5.125	34	0.151	2.841	<0.001
Residuals	10.824	204	0.053		

Y: year; N: Nitrogen fertilization; CS: cropping system.

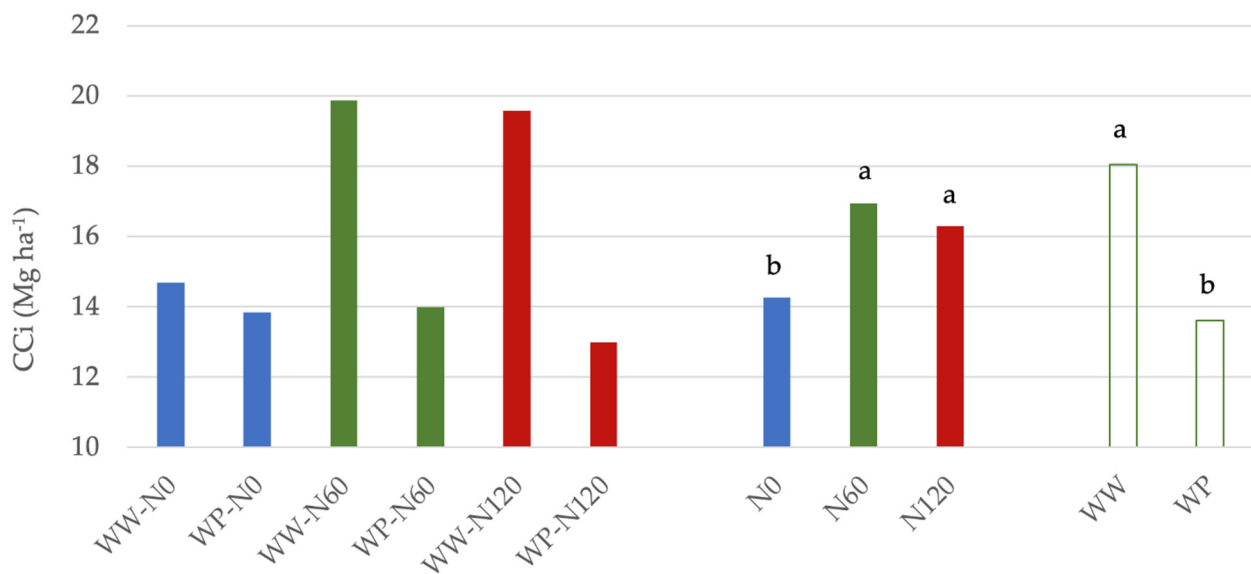
Table 4 summarizes the annual carbon input (ACi) results, showing a relatively high standard deviation and an increment with nitrogen fertilization levels up to 60 kg ha<sup>-1</sup>, with no significant difference with the highest fertilization rate (120 kg ha<sup>-1</sup>). Statistically significant differences were observed based on the cropping system (1.0 Mg ha<sup>-1</sup> y<sup>-1</sup> in WW and 0.76 Mg ha<sup>-1</sup> y<sup>-1</sup> in WP crop rotation). No substantial interaction effect was noted between the N and CS factors.

**Table 4.** Statistics of the annual carbon input (ACi) during the trial period.

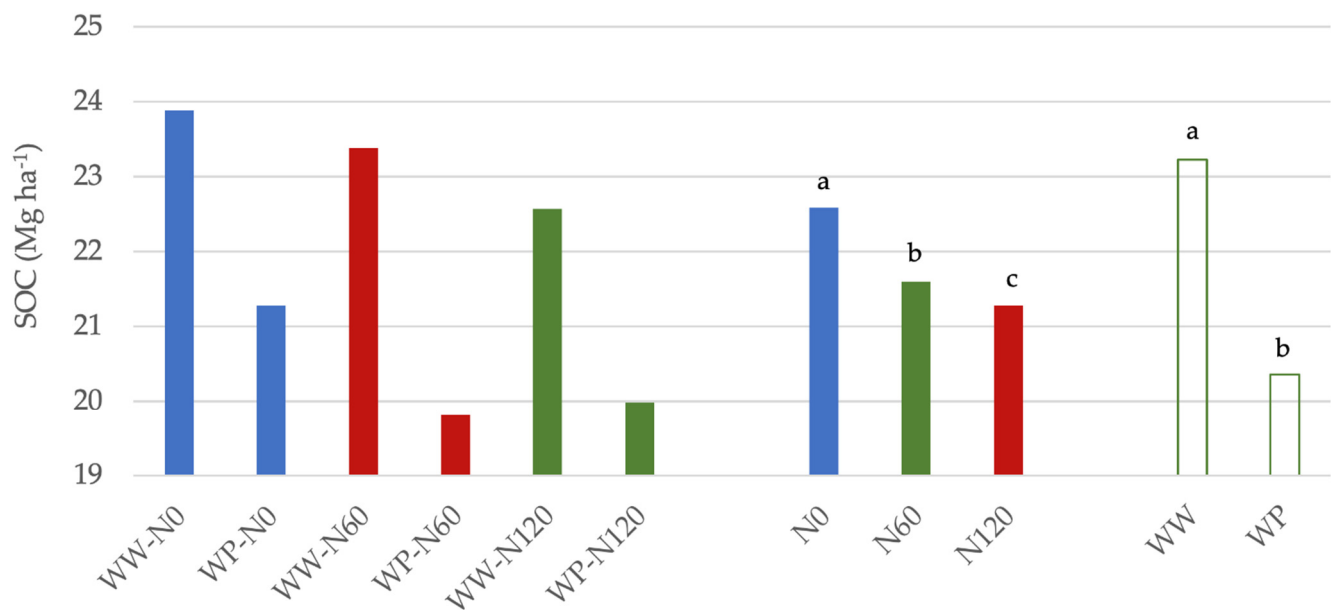
	ACi (Mg ha <sup>-1</sup> y <sup>-1</sup> )	St.dev.	Max	Min	Range
<b>N × CS</b>					
WW-N0	0.82	0.17	0.92	0.33	0.59
WP-N0	0.77	0.12	0.77	0.35	0.43
WW-N60	1.10	0.22	1.27	0.44	0.83
WP-N60	0.78	0.28	1.33	0.43	0.89
WW-N120	1.09	0.32	1.44	0.32	1.11
WP-N120	0.72	0.39	1.55	0.28	1.27
<b>N</b>					
N0	0.79 b	0.14	0.85	0.34	0.51
N60	0.94 a	0.25	1.30	0.44	0.86
N120	0.91 a	0.36	1.49	0.30	1.19
<b>CS</b>					
WW	1.00 a	0.24	1.21	0.36	0.84
WP	0.76 b	0.27	1.22	0.35	0.86

For ACi values, means followed by the same letter are significantly non diverse at  $p \leq 0.05$  (Tukey's HSD test). CS: cropping system (WW: durum wheat monocropping; WP: durum wheat–field pea rotation); N: N fertilization level (N0: unfertilized plots; N60: 60 kg ha<sup>-1</sup> N; 120: 120 kg ha<sup>-1</sup> N).

The cumulative carbon input (CCi) results (Figure 2) revealed that WW–N60 had the highest value (around 20 Mg ha<sup>-1</sup>). Among N fertilization rates, 60 kg ha<sup>-1</sup> of nitrogen led to the highest biomass productivity, as expressed by the highest CCi average. Concerning the CS mean values, this parameter was greater in WW (18.0 Mg ha<sup>-1</sup>) compared to WP (less than 14.0 Mg ha<sup>-1</sup>), resulting in final SOC contents of 23.2 and 20.4 Mg ha<sup>-1</sup> of carbon for WW and WP, respectively (Figure 3).



**Figure 2.** Cumulative carbon input (CCi) (Mg ha<sup>-1</sup>) in two cropping systems (WW—durum wheat monocropping and WP—durum wheat–field pea rotation), with two N fertilization levels (60 kg ha<sup>-1</sup>—N60 and 120—N120), plus the unfertilized control (N0). Means marked by the same letter indicate no statistically significant differences at  $p < 0.05$  (Tukey’s HSD test).



**Figure 3.** Soil organic carbon (SOC; Mg ha<sup>-1</sup>) at the end of an 18-year trial in two cropping systems (WW—durum wheat monocropping and WP—durum wheat–field pea rotation), with two N fertilization levels (60 kg ha<sup>-1</sup>—N60 and 120—N120), plus the unfertilized control (N0). Means marked by the same letter indicate no statistically significant differences at  $p < 0.05$  (Tukey’s HSD test).

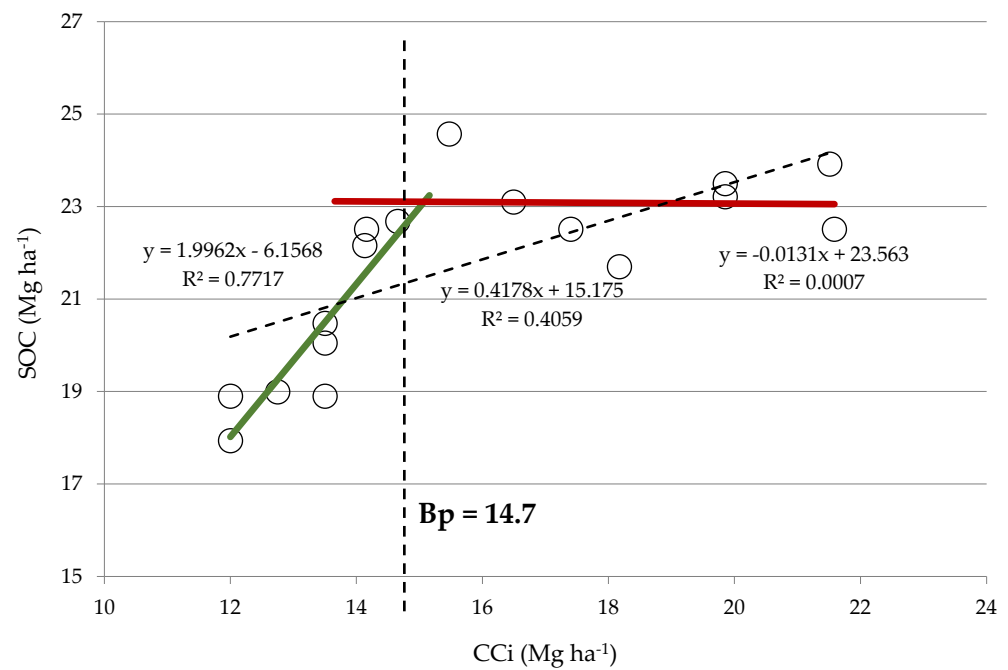
Consequently, a significant difference of more than 3 Mg ha<sup>-1</sup> in SOC sequestration was found between the two cropping systems after 18 years (Figure 3). Significant differences were also found among the fertilization treatments, with the unfertilized plots (N0) displaying a definite SOC increment.

All interaction effects between the “cropping system” and “fertilization” factors demonstrated the effectiveness of the durum wheat monocropping system in SOC sequestration and the negative impact of N fertilization mainly when crop rotation was adopted (Figure 3).



### 3.2. Breakdown Regression

Segmented regression analysis identified a breakpoint at 14.7 Mg ha<sup>-1</sup> of CCI, corresponding to 23.5 Mg ha<sup>-1</sup> of SOC (Figure 4). The ANOVA results for the regressions (Tables 5 and 6) indicated high significance levels for the segmented and the single regression models. However, the segmented regression model (with 18% unexplained variance) fitted the data better than the single regression, where more than 45% of the total variance remained unexplained. Regression below the breakpoint demonstrated a high positive and direct association ( $R^2 = 0.777$ ) between CCI and SOC, indicating a high C sequestration rate. Beyond the breakpoint, the regression slope was not significantly different from 0 ( $p = 0.06$ ) (Figure 4).



**Figure 4.** Regression lines between cumulative carbon input (CCI) and soil organic carbon (SOC), with (red and green lines) and without (black dotted line) the breakpoint (Bp).

**Table 5.** ANOVA table for the linear regression without and with the breakpoint.

		SS	d.f.	MS	F	P
Linear regression	Explained	35.6	1	35.6	12.4	0.009
	Unexplained	45.8	16	2.7		
Breakdown regression	Explained	63.2	3	21.1	16.2	0.001
	Unexplained	18.2	14	1.3		

**Table 6.** Results of regression of SOC (Mg ha<sup>-1</sup>) against CCI (Mg ha<sup>-1</sup>) with the optimal breakpoint (Bp).

	Bp	d.f.	SOC	s.d.	CCI	s.d.
Linear regression	12.0	18	21.8	2.2	15.8	1.6
x < Bp	14.8	10	20.7	2.1	13.5	1.0
x > Bp	14.8	8	23.3	1.1	18.8	1.1

Under the assumption that the constant in the >Bp regression is the SOC steady-state level, the constant in the <Bp regression represents the theoretical SOC stock at zero C

input. The slope regression under the breakpoint can be used to estimate the years of soil C sequestration time (CsT) up to the SOC saturation for a cropping system's C input according to:

$$\text{CsT} = (\text{constant in the } >\text{Bp regression} - \text{constant in the } <\text{Bp regression}) / \text{ACi}$$

As the breakpoint was assessed at  $14.7 \text{ Mg ha}^{-1}$ , in this long-term experiment, the C steady-state level was not reached with the annual C input of the WP cropping system ( $0.76 \text{ Mg ha}^{-1}$ ), and, in general, it was not achieved in N0 either. For the annual C input, the SOC steady state was attained after 17 years in WW, while an estimated span of 23 years was inferred for WP.

### 3.3. Aggregate Size Distribution and Associated Carbon

The distribution of soil aggregate fraction-size classes in the topsoil significantly differed among the treatments. The  $<0.25 \text{ mm}$  fractions (aggregate) accounted for 60% in WW and 64% in WP cropping systems. N fertilization decreased the  $<0.25 \text{ mm}$  fractions by 63–59% from 0 to  $120 \text{ N}$  fertilization ( $p < 0.05$ ). A substantial increase in the  $0.25\text{--}0.75 \text{ mm}$  fraction (from 19 to 27%) was noted with N fertilization (Table 7).

**Table 7.** Distribution of soil aggregates into size classes and calculated C amounts according to cropping system (WW and WP) and N fertilization level (0, 60, and  $120 \text{ kg N ha}^{-1}$ ).

	Aggregate Size ( $\text{g kg}^{-1}$ )			SOC Bulk ( $\text{g kg}^{-1}$ )	g C $\text{kg}^{-1}$ Bulk Soil		
	$>75 \mu\text{m}$	$25\text{--}75 \mu\text{m}$	$<25 \mu\text{m}$		$>75 \mu\text{m}$	$25\text{--}75 \mu\text{m}$	$<25 \mu\text{m}$
<b>N × CS</b>							
WW-N0	190	197	613	8.0	0.80	0.73	5.80
WP-N0	162	181	657	7.1	1.23	1.10	4.03
WW-N60	170	213	617	7.5	1.97	2.03	2.93
WP-N60	154	206	640	6.7	1.70	1.74	2.55
WW-N120	136	297	567	7.3	1.47	2.10	3.45
WP-N120	154	234	612	6.6	1.10	1.73	3.11
<b>N</b>							
N0	176 a	189 b	635 a	7.5 a	1.02 b	0.91 c	4.92 a
N60	162 a	210 a	628 a	7.1 b	1.83 a	1.88 b	2.74 c
N120	145 b	266 a	589 b	7.0 b	1.28 b	1.92 a	3.28 b
<b>CS</b>							
WW	165	236 a	599 b	7.6 a	1.41	1.62	4.06 a
WP	157	207 b	636 a	6.8 b	1.34	1.53	3.23 b

Means followed by the same letter indicate no statistically significant differences at  $p \leq 0.05$  (Tukey's HSD test). CS: cropping system (WW: durum wheat monocropping; WP: durum wheat–field pea rotation); N: N fertilization level (N0: unfertilized plots; N60:  $60 \text{ kg ha}^{-1} \text{ N}$ ; N120:  $120 \text{ kg ha}^{-1} \text{ N}$ ).

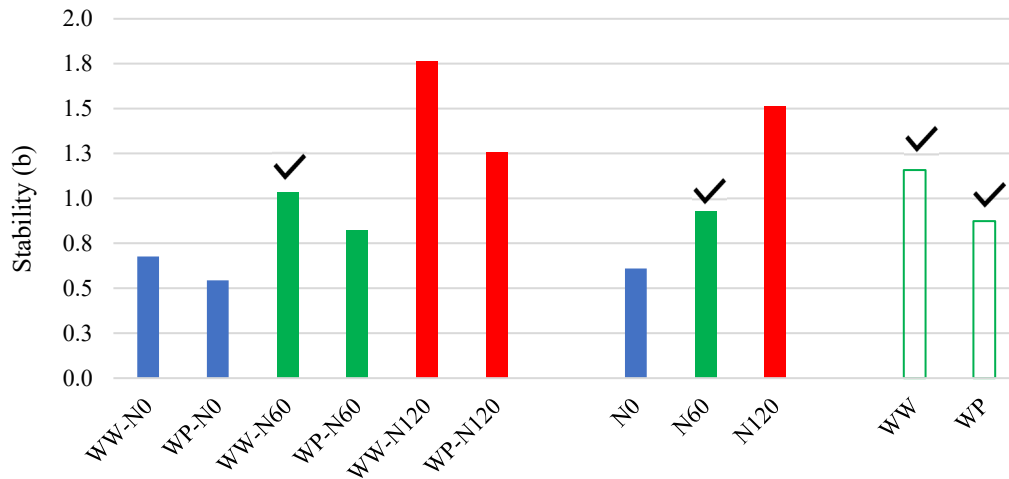
The cropping system influenced the SOC allocation among different aggregate size classes, with the associated C concentrated mainly in the micro-sized fraction (Table 7). Nitrogen fertilization increased C stocks in the  $25\text{--}75 \text{ mm}$ -sized fractions from  $0.91$  to  $1.92 \text{ g C kg}^{-1}$  bulk soil in N0 and N120, respectively. The wheat–pea rotation reduced SOC levels in the silt + clay-sized fractions compared to wheat monocropping.

### 3.4. Carbon Input Stability and SOC Sequestration

Combined ANOVA over the years (Tables 2 and 3) highlighted the relative importance of year, rotation, and fertilization in assessing carbon dynamics in soil, also showing a highly significant “year” × “treatment” interaction.

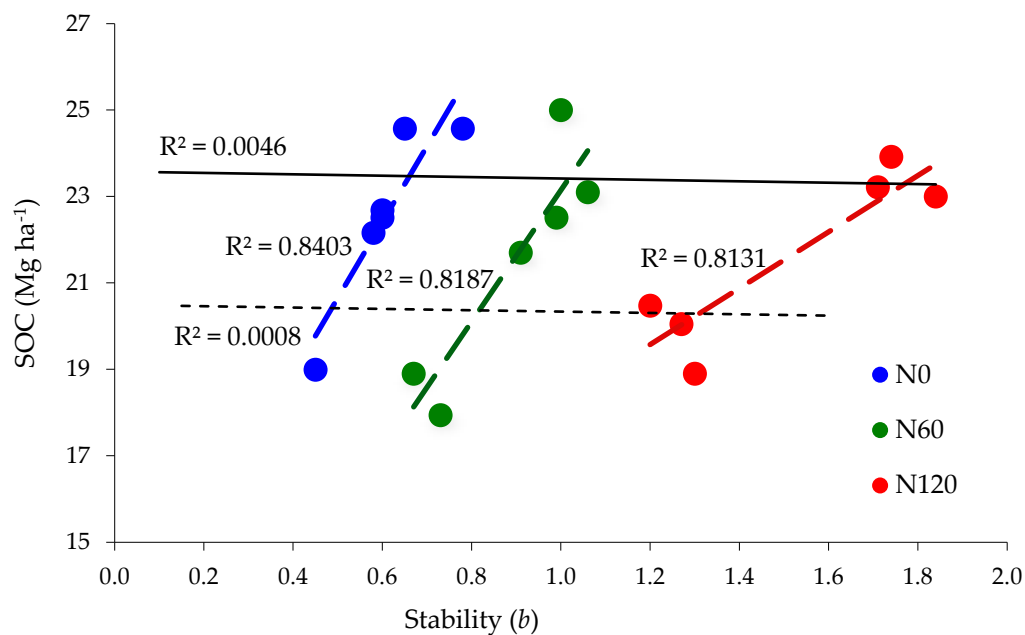
Stability analysis on the annual carbon input (ACi) based on slope regression (b) indicated that WW had higher stability ( $b = 1.14$ ) compared to WP ( $b = 0.84$ ). Nitrogen fertilization showed an increasing b coefficient with higher nitrogen rates ( $0.61$ ,  $0.89$ , and

1.46 for the three rates, respectively), and only N60 had a b coefficient of 1. Among the CS × N interactions, only WW–N60 exhibited a b value significantly different from 1 (Figure 5).



**Figure 5.** Stability in terms of “b” coefficient in two cropping systems (WW—durum wheat monocropping and WP—durum wheat–field pea rotation), with two N fertilization levels (60 kg ha<sup>−1</sup>—N60 and 120—N120), plus an unfertilized control (N0). Means marked with a tick are significantly non different from 1.

Nitrogen fertilization had a significant effect on the cropping system carbon input stability values (Figure 6). The lowest b coefficients were observed in N0 fertilization, while N120 exhibited values between 1.25 and 1.8. Notably, the highest SOC values were obtained by N0 and N60 at stability values <1, and the same SOC amount obtained with increasing N levels corresponded to increased b values. In other words, as the stability levels were rising with the N fertilization rate, an increase in productivity (carbon input) through additional N supply never affected SOC sequestration.



**Figure 6.** SOC sequestration according to the interaction between stability (b coefficient) and N fertilization. N0, blue line; N60, green line; N120, red line; continuous horizontal line, WW; dotted horizontal line, WP.

### 3.5. Cropping System Stability and Carbon Saturation Deficit

After 18 years, as shown in Figure 7, in the WP cropping system  $SOC_{sat}$  remained relatively constant at higher stability levels ( $b = 1$  to  $b = 1.8$ ), while from  $b = 0.5$  to  $b = 1$ , the SOC saturation deficit quickly decreased from 0.4 to 0.1  $g\ kg^{-1}$ . In contrast, the WW cropping system showed distinct behavior based on  $b$  values, with a recognizable breakpoint at about  $b = 1$ .

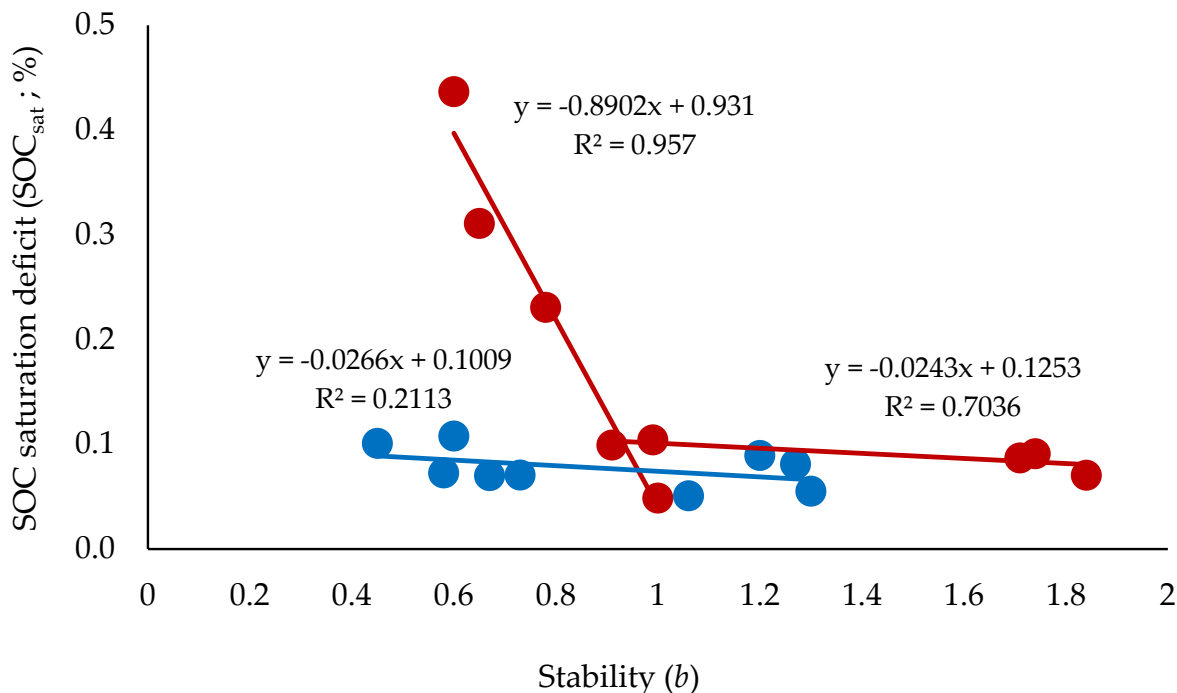


Figure 7. Relationship between stability and  $SOC_{sat}$  (red for WW; blue for WP).

This suggests that the WW cropping system has greater potential for SOC sequestration, mainly when low stability in carbon input occurs. This outcome is especially evident in the regression plot between stability values and Cse (Figure 8), where a definite, non-linear, inverse association appears.

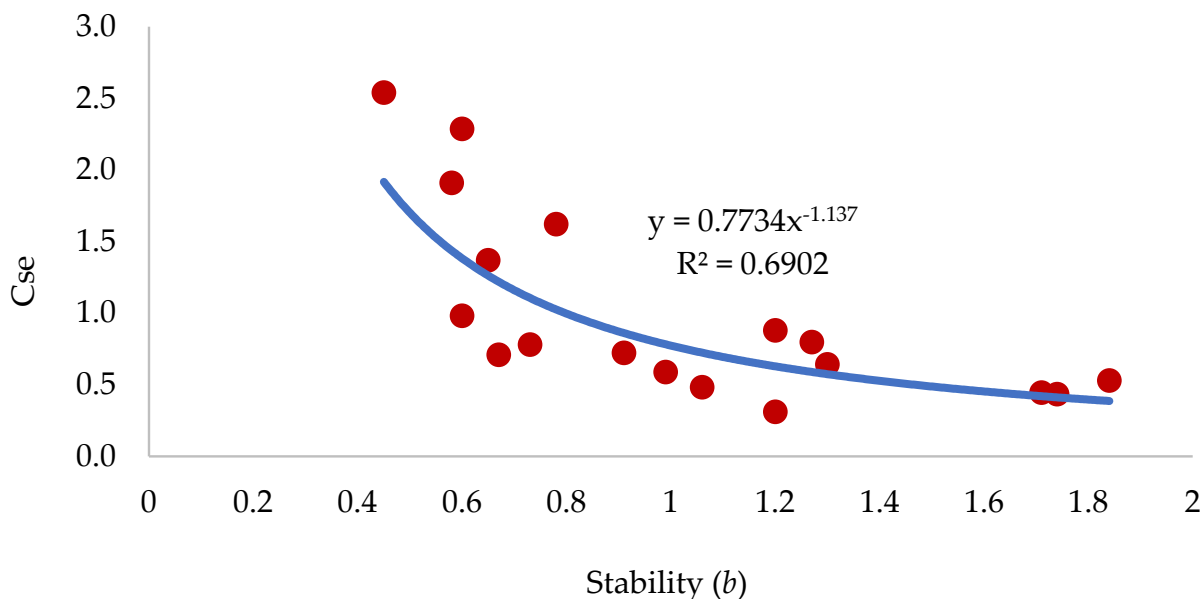


Figure 8. Relationship between stability and Cse (carbon sequestration efficiency).

## 4. Discussion

### 4.1. Role of Carbon Input Stability in SOC Sequestration

These experimental findings demonstrate that field pea residues degrade faster than wheat due to their lower C:N ratio. This suggests that continuous wheat cultivation may sequester more soil carbon than incorporating legumes into the crop rotation. These observations align with evaluations on rotation effects conducted by several researchers in different environments. In Argentina, Studdert and Echeverría [52] reported that soybean-produced  $1.2 \text{ t ha}^{-1}$  residues induced a net loss of soil carbon, whereas corn, with  $3.0 \text{ t ha}^{-1}$  residues, significantly curtailed soil carbon loss from the system. Hence, they concluded that an absence of legumes in a crop rotation leads to higher SOC values. In semi-arid Canada, Curtin et al. [53] reported that while black lentil added carbon between  $1.4$  and  $1.8 \text{ t ha}^{-1}$ , wheat contributed 2–3 times this amount annually. Thus, these authors demonstrated that cereals outperform legumes in achieving maximum carbon sequestration rates in semi-arid environments. Similar results were also found in durum wheat cropping systems in semi-arid areas [25,54,55], as well as in different climatic settings [43].

In fact, crop rotations that include legumes typically generate lower biomass than monocultures of durum wheat, resulting in less crop residue returned to the soil.

Plant residues represent the primary carbon input to the soil; thus, increased crop residue associated with proper management practices can either augment SOC or reduce the rate of SOC loss [56,57]. This assumption was affirmed by Blanco-Canqui and Lal [56] through several instances of positive relationships between the removal of crop residues and the reduction of SOC concentration.

Despite nitrogen fertilization boosting yields, no significant correlation emerged between crop residue quantity and SOC stocks across diverse rotations or years. Similar conclusions were drawn by Poirier et al. [58], who observed that increased crop residue did not substantially impact SOC storage. Thus, while nitrogen application elevates crop and residue yields, its influence on SOC remains inconsistent. Similar findings have been reported in the same Mediterranean climate [25,55,59], confirming that the legumes in the rotation did not significantly increase the residue amount or SOC content.

Furthermore, excessive nitrogen applications beyond the optimal rate for maximum crop yield lead to a higher nitrogen uptake. This, in turn, improves stover quality by lowering the C:N ratio [60], accelerating residue decomposition [61,62] and eventually causing a negative SOC impact. In rainfed semi-arid environments, nitrogen application rates are generally lower compared to more humid regions. This is because elevated nitrogen levels foster high biomass productivity during the winter period (typically without water deficiency) but also lead to excessive water use, limiting productivity during the reproductive stage.

Specifically, only under N0 conditions was the carbon input equivalent between the continuous wheat (WW) and wheat–pea rotation (WP), yet the SOC remained higher solely within the WW cropping system.

Nitrogen input is predominantly determined by mineral fertilization in durum wheat monocropping, whereas in WP rotation, mineral fertilization is administered only in wheat cultivation years. The only scenario in which the WP cropping system receives more nitrogen than WW was when no fertilizer was applied (N0).

### 4.2. Role of Carbon Input Stability in SOC Saturation

While considerable research has been conducted on SOC sequestration in response to the quality and quantity of carbon input [29,30], the influence of carbon input stability remains largely unexplored.

SOC sequestration arises from a balance between carbon input (identified with biomass productivity) and SOC loss, determined by soil organic matter (SOM) decomposition [63]. Many factors play a role in assessing this balance, also including microbial activity [64,65]. Moreover, due to climatic variations, carbon inputs' quality, quantity, and intake frequency into the soil impact the ultimate carbon budget [66–68].

In the semi-arid environment where the trials were conducted, constant carbon input additions into the soil ( $b = 1$ ) seem more effective in promoting SOC sequestration than high interannual variability in carbon input. This effectiveness is influenced by the choice of cropping system (with WP outperforming WW) and nitrogen fertilization, which amplifies the variability of residues incorporated into the soil.

## 5. Conclusions

Yield stability is, generally, a critical parameter for comparing cropping systems in relation to climate variability. The regression method in stability analysis shows large differences among different cropping systems and their contribution to carbon input, but it is also strongly influenced by nitrogen fertilization.

The results show that stability analysis is a suitable method for interpreting the significant environment  $\times$  cropping system interactions revealed by the combined analysis of variance over the years in long-term experiments. It may also provide a more direct method of assessing temporal variability in long-term experiments.

In this work, the hypothesis that a more stable cropping system can improve soil carbon sequestration has been demonstrated, also reconsidering the role of nitrogen fertilization, especially in semi-arid environments, where nitrogen excess can determine biomass yield reduction in a dry season and vice-versa, thereby reducing and increasing the stability of carbon input via biomass production.

Carbon input stability also increases the potential carbon sequestration in the soil through the dimension of carbon sinks and seems to be correlated with soil microbial community and substrate utilization strategies, improving or reducing SOM decomposition.

Strategies and recommendations to stakeholders could be refined using stability analysis in relation to agronomic treatment performances over time. This approach could also stress differences between crop rotations and fertilizer treatments in dry and rainy environments.

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