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THE INFLUENCE OF GENOTYPE, SOIL TILLAGE AND FERTILISATION ON CARBON SEQUESTRATION IN MAIZE

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ABSTRACT. The increase in the concentration of greenhouse gases in the Earth's atmosphere, especially that of CO_2, is a major concern because it is the main source leading to global warming, and its impact on climate change is still widely studied. The intensity and frequency of drought and flooding increase due to the change in climate, which has a negative impact on crop productivity and food security. The aim of this paper was to demonstrate the role of corn in carbon sequestration based on plant biomass and soil organic carbon accumulation. We presented the main factors that contribute to carbon sequestration and concrete examples regarding the capacity of corn hybrids created at National Agricultural Research and Development Institute (NARDI) Fundulea, as well as aspects regarding the importance of soil tillage and fertilisation. In contrast to genotypes in which this ratio was lower, maize genotypes that demonstrated a rise in the root:stem ratio under drought stress conditions produced more biomass, suggesting that the roots of these genotypes can grow at water potentials where stem growth is inhibited, which are attributes that also prove their good potential for carbon sequestration under climate change conditions. The organic carbon content in the superficial soil layer decreased with the intensification of the degree of soil mobilisation (9.95% when working the soil with the chisel, 17.91% for ploughing), but fertilisation had a beneficial effect. The biomass was higher than that of the unfertilised soil. This means that a higher carbon input has a positive influence on the carbon stock in the soil.

Keywords: maize; carbon sequestration; hybrids; soil tillage; fertilisation.

INTRODUCTION

Studies on raising soil organic carbon content and the role of plants in carbon sequestration through plant biomass have gained significant importance as a result of worries about climate change (Lal, 2004b).

The specialised literature data have shown that the best species, genotypes, or technological measures, such as soil conservation, application of fertilisers, organic amendments, crop rotation, and residue management, can be achieved to increase carbon sequestration in agricultural soils and transform the soil into a reservoir for atmospheric carbon (Lal, 2004a; Gillion *et al.*, 2006; Zahra *et al.*, 2016). Presently, soil is thought to be capable of storing between 0.4 and 1.2 Gt of carbon annually.

Maize occupies third place in the world concerning cultivation area (after wheat and rice) and first place in terms of production, being used in human nutrition (canned food, baby food, breast milk, porridge, puddings, tamales and other foods intended for human consumption), animal feed, raw material in industry (filling for plastics, packaging materials, insulating materials, adhesives, chemical products, explosives, paint, paste, abrasives, dyes, insecticides, pharmaceutical products, organic acids, solvents, artificial silk, antifreeze, soaps), the production of fuels to replace petrol and diesel, and more recently indirectly for the potential of carbon sequestration to reduce greenhouse gas emissions.

Romania was the second producer of corn in the European Union (EU) in 2014 and ranked twelfth on the world list. With a production of almost 11 million harvested tonnes, Romania produced over 8% of the total amount of corn harvested in the EU. However, although we have the largest cultivated area of corn in the EU, the yield per hectare is half that of France and Belgium. Information related to the importance of maize crops regarding their ability to contribute to carbon storage in the soil is scarce, both in the world and in Romania.

The goal of the current research was to examine the dynamics of above- and belowground biomass, organic carbon content, nitrogen content, and C/N ratio in relation to maize hybrid, soil tillage, and fertilization to determine the importance of corn cultivation in carbon sequestration.

MATERIALS AND METHODS

To achieve the proposed objective, soil and plant analyses were carried out as part of experiments in controlled environmental conditions and in experimental fields.

To determine the importance of the choice of genotypes in increasing carbon sequestration in agricultural soils, 18 maize hybrids released by the corn breeding department from the NARDI Fundulea were studied under optimal and hydric stress conditions. After being sterilized with 2% sodium hypochlorite and washed with distilled water, the seeds were placed on filter paper in water and were kept at a constant temperature of 25±1°C and in light conditions 14 hours a day to germinate.

Half of the rolls had water stress produced eight days after sowing using a 10% polyethylene glycol 10,000 solution (creating an osmotic potential of about -0.30 Mpa) for ten days. Half of the remaining rolls were produced using tap water (control plants).

Leaf area. Estimates of leaf area were obtained using the following equation: leaf area (mm^2) = L × l × 0.8, where L is the leaf length, l is leaf width and 0.8 is the correction coefficient for maize.

Biomass accumulation. Gravimetric weighing was used to determine biomass accumulation.

For evidence of the importance of soil tillage, mineral composition and organic fertilisation on carbon sequestration, soil samples were collected from two sites from the experimental field of NARDI located at 44° 26’ 33” N, 26° 30’ 40’’ E: a long-term site (since 1977) in a continuous maize cropping system and a site with different soil tillage, cover crops and fertilisation practices. Experimental factors were as follows: factor A – tillage with five gradations (no till; autumn ploughing and disk; spring ploughing and disk; chisel and disk; disk) and factor B – fertilisers applied in four gradations (nonfertilised; manure applied at 4 years, 20 t/ha; nitrogen 100 kg a.i /ha+ phosphorus 80 kg a.i P_2O_5/ha; nitrogen 100 kg a.i /ha + phosphorus 80 kg a.i P2O5/ha + cover crop). The soil samples were collected in 2020 from the 0–5 cm soil layer before corn sowing.

Organic matter was determined volumetrically following the wet oxidation method according to Walkley Black, modified by Gogoașa - STAS 7184/21-82. The samples were collected from the 0–5 cm soil layer.

The organic carbon content of the soil was estimated indirectly based on the conversion coefficient of organic matter of soil from Fundulea (1.724).

The soil organic carbon stock (SOC) was calculated using the following formula (Datta et al., 2018): SOC (t/ha) = organic carbon (%) × bulk density (g/cm^3) × thickness of soil sample (cm).

The data were statistically analysed by analysis of variance (ANOVA).

RESULTS AND DISCUSSION

Genetic basis

The carbon sequestration potential of any crop is determined by biomass and grain production. The production potential of corn hybrids, considering the period from silking to physiological maturity, depends on the development and efficiency of the leaf and root system on the storage capacity in the reserve organs and on the speed of transfer and accumulation of the nutrients obtained in the photosynthesis process.

Biomass components in corn show great variability and are determined by the length of the vegetation period of the hybrids, climatic conditions and cultivation technology, of which soil tillage and fertilisers have the greatest influence.

From the total aerial biomass, the grains represented 40–50%, and the stems with leaves represented 37–43%. The pans and rachis occupied 12–13%. As the vegetation period of the hybrids increased, the percentage of grains decreased and the percentage of vegetative parts in the total aerial biomass increased. Late hybrids stored one-third of the total amount of aerial biomass in grains, while early hybrids stored almost half. New corn hybrids compared to older hybrids had higher yields under different biotic and abiotic stress conditions, such as heat, drought, moisture and cold. This improvement in production over the years is due not only to the improvement of tolerance to abiotic and biotic stress but also to the increase in sowing density. Recent studies carried out at Fundulea have highlighted the great variability of maize genotypes at the leaf surface, as well as the ratio of root biomass/aerial part biomass in conditions of water stress (Petcu *et al.*, 2018).

ANOVA for leaf area revealed the highly significant effect of water stress and genotype and the significant interaction between the two factors on leaf area (*Table 1*). The average leaf surface of the corn hybrids studied under optimal conditions was 2131 mm^2 compared to the leaf surface under water stress conditions, which was 1120 mm^2, on average, indicating a difference of 52% (*Table 1*). A series of hybrids with high leaf area were highlighted in both conditions: HSF 1191-14, HSF 1158-14, HSF 1128-14 and P 9357 (*Figure 1*, upper right quadrant), but also two genotypes with high leaf area in water stress conditions: HSF 3425-16 and HSF 4040-15 (*Figure 1*, upper left quadrant).

Table 1 – Summary of the analysis of variance depicting the sum of squares, the degrees of freedom (DF) and mean square for leaf area evaluated in 18 maize hybrids under water stress and no water stress

Source of variation	Sum of squares	Degrees of freedom	Mean square	F values and significance
Factor A: treatment (control, water stress)	27,574,019	1	27,574,019	166.71***
Factor B: genotype	8,306,788	17	488,634.6	2.95***
Interaction A*B	5,129,984	17	301,763.7	1.82*
Error	11,908,328	72	165,393.4	-

*** significant at P < 0.01%, * significant at P < 0.5%.

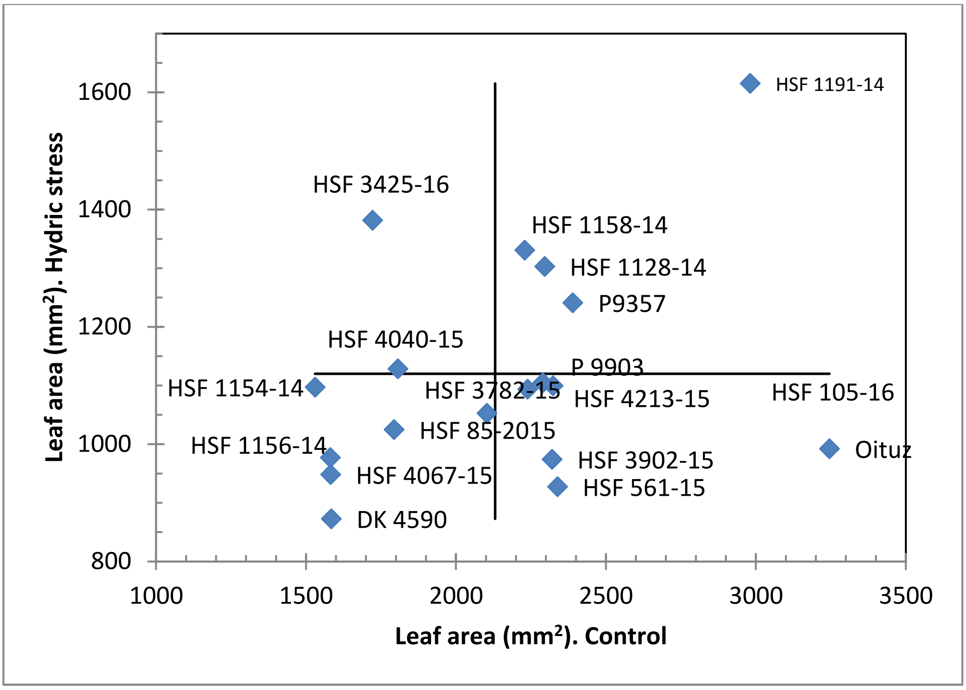

Figure 1 – Leaf area of the maize genotypes studied under optimal and water stress conditions. The horizontal line represents the average leaf area for the control and the vertical line represents water stress

Six hybrids had reduced leaf area under water stress conditions but high leaf area under optimal conditions, suggesting that these hybrids may have a mechanism to avoid drought by conserving water (HSF 105-16, Oituz, HSF 3902, HSF 561-15, HSF 4213-15 and P 9903). Data from the literature have shown that this is beneficial, especially when drought continues into the grain filling phase.

Under water stress conditions, even if the growth of the leaf surface is inhibited, carbon assimilation is maintained close to normal limits, and excess carbon is stored and used either for osmotic regulation or for root growth. The root:stem biomass ratio in the hybrids created at Fundulea was higher under conditions of water stress (*Table 2*).

The increase in the root:stem biomass ratio indicates the development of the density of the lateral roots compared to the leaf surface, which translates into a better capacity to support the water regime in the plant under water stress conditions. The following genotypes showed an increase in this ratio under conditions of water stress: Oituz, F 376, HSF 160-11, HSF 31-11 and HSF 170-11.

Table 2 – The effect of water stress on biomass accumulation in the stems and roots of studied maize hybrids

Genotype	**Stem (g/pl)**		**Root (g/pl)**		**Root/stem**	
	Control	**Water stress**	**Control**	**Water stress**	**Control**	**Water stress**
Oituz	0.81	0.43	0.93	0.75	1.15	1.75
P9357	0.75	0.37	1.05	0.89	1.41	2.40
P 9903	0.70	0.41	0.75	0.70	1.08	1.71
DK 4590	0.59	0.34	0.73	0.60	1.24	1.77
HSF 1154-14	0.56	0.38	0.74	0.78	1.32	2.08
HSF 1158-14	0.65	0.43	0.86	0.80	1.32	1.86
HSF 1191-14	0.83	0.45	1.05	0.82	1.26	1.83
HSF 1128-14	0.68	0.35	0.89	0.72	1.31	2.06
HSF 1156-14	0.59	0.27	0.91	0.44	1.55	1.64
HSF 4040-15	0.82	0.50	1.04	0.86	1.26	1.70
HSF 3782-15	0.57	0.32	0.90	0.74	1.57	2.33
HSF 561-15	0.59	0.31	0.75	0.70	1.28	2.22
HSF 4067-15	0.52	0.33	0.58	0.56	1.12	1.72
HSF 3902-15	0.76	0.37	0.80	0.70	1.05	1.89
HSF 4213-15	0.69	0.38	0.85	0.67	1.23	1.77
HSF 85-2015	0.43	0.28	0.55	0.47	1.26	1.66
HSF 105-16	0.75	0.41	0.81	0.56	1.08	1.36
HSF 3425-16	0.56	0.41	0.76	0.71	1.36	1.73
Average	*0.66*	*0.37*	*0.83*	*0.69*	*1.27*	*1.86*
*LSD 5%**	*0.3*	*0.18*	*0.16*	*0.19*	*0.22*	*0.21*

*Least significant differences of 5%

Compared to genotypes where this ratio was lower (HSF 2809-11, HSF 474-11, HSF 465-11, HSF 21-11), production was higher. The explanation for the increase in the root: stem biomass ratio under water stress conditions is also given by the fact that in corn and other species, roots can grow at water potentials at which stem growth is inhibited, which also demonstrates the potential of corn for carbon sequestration under climate change conditions. For simulations of SOC dynamics, knowledge of maize's underground biomass build-up is crucial in the context of new green deal requirements regarding the maintenance of soil health. Measurement of belowground C inputs in the field, however, may be challenging, and rhizodeposition is especially difficult to quantify (Amos and Walters, 2006). At silking or harvest spontaneous sampling at varying depths is used to estimate the net cumulative belowground biomass of maize (Komainda *et al.*, 2018). Our approach quantified root biomass by direct measurements for maize in the early vegetation stage. In this context, based on the premise that the genotypes will maintain these characteristics even in the maturity stages, they will contribute more to carbon sequestration in the soil through the intake of higher biomass.

The root:shoot biomass ratio (R:S) of existing crops has been the subject of numerous studies, but less research has been done on the variation within a crop species, which is crucial information for choosing genotypes to boost soil carbon stocks for mitigating climate change and ensuring food security.

Our research agrees with current developments (Mathew *et al.*, 2019) in selecting potential genotypes for breeding including evaluating the distribution of biomass between the roots and shoots in response to different soil water levels.

Soil tillage and fertilisation

Data from specialised literature show that disturbing the soil by conventional systems not only affects the stability of the soil aggregates but exposes them to a higher oxygen concentration, leading to high gas exchange, which leads to greater soil carbon loss (Hipps, 1991; Liebman and Staver, 2001; Zaharia and Cociu, 2009). Thus, negative conditions are created due to carbon sequestration in the soil, the activity of microorganisms and the loss of water from the soil through evaporation. The results obtained at Fundulea showed a reduction in the organic carbon content because of soil tillage and the carbon:nitrogen ratio in the case of the no till variant (*Table 3*).

Table 3 – Effect of tillage on soil organic matter, organic carbon content and C:N ratio. Maize monoculture (42 years) (average values)

Soil tillage	Organic carbon (%)	Total nitrogen (%)	C:N	SOC t/ha
No till	2.01	0.15	13.42	15.18
Worked with a chisel	1.81	0.125	14.48	11.31
Ploughing	1.65	0.113	14.60	10.40
LSD 5%*	0.52	0.029	2.49	2.75

*Least significant differences of 5%

Soil tillage with a chisel or plough had the effect of reducing the carbon stock in the soil by 25% (11.31 t/ha) and 31% (11.31 t/ha), respectively, compared to the untilled soil (15.18 t/ha) (*Table 3*). Similar results were obtained by Krauss *et al.* (2022), who highlighted that in temperate Europe, reduced tillage has an impact on soil organic carbon stores. When compared to ploughing, reduced tillage raised SOC stocks in the top layer (0–5 cm) by 20.8% or 3.8 Mg ha^{-1}.

In the long term, the effect of a single tillage operation on soil carbon stored in soil in which tillage has not been carried out for a long period appears to differ between soils with different properties.

The sampling depth is also very important. A detailed study carried out by Petcu (1996) highlighted that, in the superficial soil layer (0–5 cm), the total carbon content was higher than in the soil worked by ploughing and disc, as explained by the maintenance and accumulation of large amounts of plant residues. The situation was not the same in the deeper soil layers (0–40 cm), where the carbon total content decreased. According to the average values of the total carbon content in the 0–40 cm layer, the following ranking in descending order of tillage variants resulted: work with disk, ploughed in autumn, ploughed in spring and not tilled.

Soil tillage indirectly leads to the intensification of mineralisation processes in the soil, which explains the results presented above.

The analyses carried out by Domnariu *et al.* (2022) at the same long-term site with corn confirmed the data from the specialised literature regarding the effect of soil tillage on the activity of soil microorganisms, highlighting that the microbial and fungal mass of the soil is lower in soil tilled with a chisel or plough compared to mechanically uncultivated soil.

The long-term use of the conventional tillage system, based on intensive tillage by ploughing with turning the furrow and removing plant residues, followed by numerous secondary works, can affect soil fertility over time (Lal, 1991), which requires crop fertilisation.

Biomass accumulation was influenced by tillage and fertilisation. The best results were obtained in the variants where the soil was tilled in autumn by ploughing and fertilised with $N_{100}P_{80}$ + cover crop, followed by variant tillage with the chisel and fertilisation with $N_{100}P_{80}$ + cover crop. The lowest values were recorded for the soil tillage option through spring ploughing associated with non-fertilisation of the crop (*Table 4*).

The results suggest that the evolution of corn was decisively influenced by the system of tillage and applied fertilisation.

Thennarasu *et al.* (2014) studied the influence of fertilisation on the carbon sequestration potential of forage maize and showed that the application of improved manure and vermicompost sequestered more carbon from the atmosphere (4.06 t/h; 3.47 t/ha) compared to mineral fertilisation (2.64 t/ha). In our country, research carried out by Cociu and Cizmas (2011) recommended the use of a soil conservation work system (conservative agriculture) with the maintenance of plant residues to reduce soil degradation and increase the amount of water stored in the soil.

Hera and Eliade (1978) found that although the incorporation of wheat straw or corn stover enriches the soil in organic carbon, only one-quarter of this carbon is found in the soil carbon in dilute alkaline solutions and practically does not appear in the carbon of humic acids. However, the situation changes if 60 kg of mineral N/ha is added to the plant residues.

The data in *Table 5* show the effect of tillage and fertilisers on SOC content in the superficial soil layer (0–5 cm).

The application of organic and mineral fertilisation changes SOC. The highest amount of SOC was observed under manure treatment and disk (14.2 t/ha) and autumn ploughing + disk (13.2 t/ha; *Table 5*).

Fertilization with minerals and with organics improves SOC in two separate ways:

- because organic manure has a high concentration of C, the soil gets C directly from the manure;
- applying organic manure and inorganic fertilizer to the soil improves its physical properties, resulting in increased crop biomass, which then enhances the subsequent organic carbon addition through veg residues in the soil (He *et al.*, 2015; Kukal *et al.*, 2009; Kundu *et al.*, 2007; Mustafa *et al.*, 2021). However, there are contrasting findings regarding the beneficial effect of fertilisation for carbon sequestration, depending on the water regime and temperatures in the region being studied.

In this regard, Hijbeek *et al.* (2018) showed that although the tropics have relatively higher soil fertility and yield benefits, the faster decomposition of SOM caused by higher temperatures reduces the potential for carbon storage and constricts the use of this win-win scenario.

Table 4 – Effect of tillage and fertilisation on biomass accumulation (aerial part) in maize in the silking phase (t/ha)

Soil tillage	Unfertilised	Manure 20 t/ha (applied at 4 years)	Fertilised $N_{100}P_{80}$	$N_{100}P_{80}$ + cover crop (mustard + rapeseed)
No till	6.5	6.5	6.5	6.5
Autumn ploughing + disk	8.9	9.3	9.2	9.4
Spring ploughing + disk	7.6	7.8	8.0	8.1
Chisel + disk	8.3	8.6	8.5	8.8
Disk	7.9	8.2	8.2	8.4
LSD 5%* for soil tillage		2.30		
LSD 5% for fertilisation		2.04		

*Least significant differences of 5%

Table 5 – Effect of tillage and fertilisation on the soil organic carbon stocks

Soil tillage	Unfertilised	Manure 20 t/ha (applied at 4 years)	Fertilised $N_{100}P_{80}$	$N_{100}P_{80}$ + cover crop (mustard + rapeseed)
No till	17.4	17.4	17.4	17.4
Autumn ploughing + disk	10.6	13.2	12.0	12.6
Spring ploughing + disk	11.2	12.7	11.7	12.4
Chisel + disk	10.4	11.6	11.6	11.6
Disk	12.2	14.2	12.3	12.8
LSD 5%*	0.66	0.82	0.64	0.62

*Least significant differences of 5%

CONCLUSIONS

Maize biomass accumulation is related to water availability, genotype and technology measures.

The traits of the maize genotypes that shown an increase in the root: stem ratio under water stress settings demonstrate their potential for carbon sequestration. The roots of these genotypes can develop at water potentials at which stem growth is inhibited.

As soil mobilization increased in intensity, the amount of organic carbon in the soil dropped.

Biomass accumulation was influenced by tillage and fertilisation. The best results were obtained when the soil underwent autumn ploughing or chisel and mineral fertilisation combined with a cover crop. The obtained results show that the addition of organic manure to mineral fertilizers increased soil carbon sequestration while simultaneously improving plant biomass production.

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All authors declare that they have read and approved the publication of the manuscript in this present form.

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