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Biochar interventions enriched with alginate-producing bacteria support the growth of maize in degraded soils

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Abstract. Biochar enriched with alginate-producing bacteria increases the ability of the soil to retain air so that it is available for the growth and production of maize in dry land. Pot experiments with mixed clay media with three types of biochar from oil palm shells, oil palm empty fruit bunches and corncob were conducted to study the interaction of biochar and alginate-producing bacteria (alginate production, phosphate solvent and nitrogen fixation) water (100%, 80% and 60%) on the growth of corn plants. Experimental results prove biochar factors, alginate-producing bacterial isolates and field capacity differ significantly from the vegetative phase of corn. Biochar interaction of corncobs with water at 100% lands capacity produces the best crops, but produces leaf area at 80% field capacity. While the interaction of corncob biochar with N-binding bacterial isolates produced the highest number of leaves. Correlation between variables that use role, biochar on the efficiency of water use for maize growth, where plant height, leaf number, leaf number and dry weight are negatively correlated with KAKL. This study provides the latest synthesis to discuss the use of biochar and bacteria as a strategy to increase support for food production of dry land that increases degradation.

1. Introduction

Projections of food need such as rice, corn and wheat in 2050 reach 3.3 billion tons, approved from agriculture. Where degradation and drought have affected one third of agricultural land [1] causing 50% of the soil to lose fertility and the ability to support plant growth [2], reduce production by 9-10% [3], hence increased the level of use of chemical fertilizers every planting season. In Indonesia, 79% of the corn planting area is on dry land [4] and accounts for 2% of the world's corn needs [1]. Therefore, maintaining the availability of sufficient water for the growth of corn is a strategic effort to support the development of food sustainability.



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Biochar applied in the field can improve soil quality and plant growth [5], because it increases the capacity of the soil to retain water and has a significant effect on increasing corn production [6], significantly increases total N by 7%, organic C by up to 69% [7], increase dissolved P which is thought to be due to mineralization by soil microbes [8]. Thereby reducing the demand for fertilizer N [9] and improve the efficiency of fertilizer used [10]. The mechanisms involved are increased soil aeration and water holding capacity, increased microbial activity and nutrient status in the soil, and changes from some important soil chemical properties [11].

The ability of soils to retain water in drought conditions and other extreme hydrological events are crucial to the vulnerability of food production systems and the preservation of soil ecosystem services [12]. The use of PGPR bacteria are a new strategy to increase plant growth of a sustainable manner [13]. The effect of bacterial prim by increasing drought stresses tolerance, increasing plant biomass up to 78% greater and survival five times higher under drought [14]. Alginate is a crude production of extracellular polymeric substances that help bacteria overcome water pressure, as well as strategies used by soil bacteria to survive and adapt to drought [15]. Oligosaccharide alginate is believed to be one of the important components for increasing growth and yields. With significant effects on root and seedling growth [16] such as encouraging root formation and growth of wheat [17] and rice [18], as well as increasing tolerance of wheat to drought stress [17]. The ability of soil bacteria to synthesize alginates must be an attribute of inoculum in drought-affected agricultural land. The ability of soil bacteria to synthesize alginates must be an attribute of inoculum in drought-affected agricultural land. Therefore enriching biochar with alginate-producing bacteria provides the latest synthesis to obtain strategies to maintain the availability of water and nutrients in degraded dry land to support sustainable food production.

2. Materials and methods

The planting media used was clay soil with soil properties as presented in Table 1. Soil was mixed with biochar from oil palm shells, oil palm empty fruit bunches and corncobs (300°C-400°C pyrolysis results) with a ratio of 94% : 6% : 5% with a total media weight of 16.5 kg. The corn seed used was BISI 18 and fertilization was carried out in accordance with the manufacturer's recommendations. The pot experiment was carried out in a plastic house using a split split plot design method of a treatment consisting of three factors, the main plot were the field capacity water content consisting of three levels of 100% (A1), 80% (A2) and 60% (A3), subplots are alginate-producing bacterial isolates consisting of three types based on the ability to produce alginate (B1), dissolve P (B2) and fix N (B3), sub-subplot was biochar types of oil palm shells (C1), oil palm empty fruit bunches (C2), and corncobs (C3). The parameters observed were plant height (cm), number of leaves (strands), leaf area (cm²), and root dry weight (g).

Table 1. Physical and chemical properties of soil used as a growing medium before biochar and alginate-producing bacteria were added.

Physical Properties	Value	Unit	Chemical Properties	Value	Unit
Clay texture	60	%	pH (H ₂ O)	5.8	
Texture Class	clay		Organic matter		
COLE	0.146	%	- C (Walkey & Black)	1.96	%
Bulk Density	1.24	g/cm ³	- N (Kjeldah)	0.15	%
PD	2.58	g/cm ³	- C / N	13	
Porosity	52	%	P ₂ O ₅ (Olsen)	9.5	ppm
Permeability	0.41	cm/hour	CEC	32.58	cmol(+)kg ⁻¹
Water content	9	%			
Field Capacity	51	%			

Source: Data after analysis at the Soil Fertility Laboratory, Hasanuddin University, 2019.

3. Results and discussion

3.1. Plant height

Table 2 presents the average height of corn plants produced in various biochar treatments for / of various moisture levels in the field capacity. The results show a significant influence of the interaction of biochar with water content of the field capacity for / of the height of corn plants. The average height of the best corn plants are produced by the interaction of biochar from corncobs (C3) with 100% field capacity (A1) moisture content. This is significantly different when there is an interaction with the field content of 80% (A2) and 60% (A3). This result was also not significantly different from the average height of maize produced by the interaction of oil palm empty fruit bunch (C2) biochar with a field moisture content of 80% (A2).

Biochar significantly influences the average height of maize at all levels of field capacity. The average height of the best plants are produced by biochar from corncobs (C3) at 100% field capacity (238.67 cm) water content and is generally better at all levels of field capacity (Figure 1 and 2). However, oil palm empty fruit bunches biochar (C2) at 80% field capacity (A2) moisture content is capable of producing plant height that is not significantly different (233.89 cm). The two treatments were able to match the average plant height in accordance with the standard height of the maize variety used (230 cm) [19]. Though this research took place in degraded soil conditions (table 1). Plant height is a plant size that is often observed as an indicator of growth and plant height as the main parameter of the plant ecology strategy [20].

Table 2. Average plant height (cm) of corn in various biochar and bacterial isolate treatments as well as moisture content in different field capacities

Moisture content of field capacity	Biochar (C)			Duncan _{0.05}
	Palm kernel shell (C1)	Oil palm empty fruit bunches (C2)	Corn cob (C3)	
100% (A1)	221.22 ^z	231.28 ^y	238.67 ^x	3.68
80% (A2)	199.33 ^z	233.89 ^x	227.00 ^y	3.87
60% (A3)	159.56 ^z	182.56 ^y	192.44 ^x	

Numbers followed by the same letters in columns (a, b, c) and rows (x, y, z) mean that they are not significantly different in Duncan's DMRT α 0.05 further test.

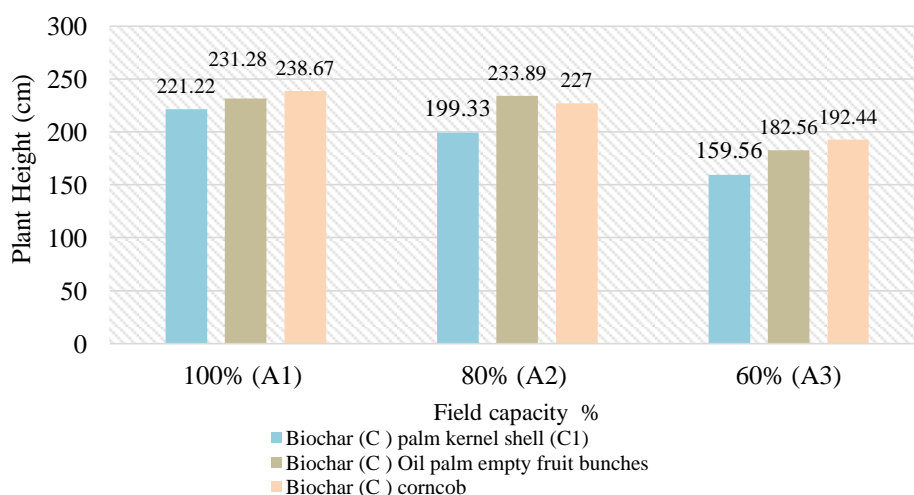


Figure 1. Average height of corn (cm) in the biochar treatment.



Figure 2. Average height of corn (cm) in the field capacity treatment (100%, 80%, 60%) and biochar type: a. Biochar from oil palm empty fruit bunches, b. biochar from oil palm shells, c. biochar from corncobs

3.2. Number of leaves

Statistical analysis of the number of leaves (strands) of corn plants is presented in table 2. The interaction of biochar treatment and bacterial isolates significantly affected the number of corn leaves. The Duncan test shows that the highest average number of corn leaves (15 strands) is produced by biochar interactions from corn cobs (C3) added with nitrogen fixation bacteria (B3) isolates. These results were significantly different from the interaction of corncob biochar with alginate-producing bacterium isolates (B1) and phosphate solubilizing bacteria (B2) isolates. Likewise, the interaction of biochar from empty fruit bunches and oil palm shells with nitrogen fixing bacteria isolates was significantly different from these results. The increase in the number of leaves of corn plants is significantly influenced by nutrient uptake, especially nitrogen from the soil. Increasing the number of leaves and leaf area is influenced by nitrogen levels that can be absorbed by plants from the soil. Biochar stimulates total leaf and total biomass, increases total amount of nitrogen and phosphorus soils, and total carbon, and increases the associated microbial community [21], so the addition of nitrogen fixing bacteria isolates will help the efficiency of nitrogen uptake by corn plants.

Table 3. Average number of corn leaves (strands) in various biochar and bacterial isolate treatments as well as moisture content in different field capacities.

Bacterial isolate (B)	Biochar (C)			Duncan $\alpha 0.05$
	Palm kernel shell (C1)	Oil palm empty fruit bunches (C2)	Corn cob (C3)	
Alginate-producing (B1)	14.00 _b ^y	14.44 _b ^x	14.11 _c ^y	0.22
Phosphate solvent (B2)	14.78 _a ^x	14.83 _a ^x	14.33 _b ^y	0.23
Fixation N (B3)	13.89 _c ^z	14.44 _b ^y	15.00 _a ^x	

Numbers followed by the same letters in columns (a, b, c) and rows (x, y, z) mean that they are not significantly different in Duncan's DMRT $\alpha 0.05$ further test.

3.3. Leaf area

Statistical analysis showed that biochar type and field capacity had a very significant effect on corn leaf area (table 3). The Duncan test shows that the average leaf area produced at 80% field capacity is 608.57 cm² wider and significantly different at 100% field capacity (593.42 cm²) and 60% (517.18 cm²). The highest average leaf area in various types of biochar was produced in biochar corncobs (592.64 cm²). These results differ significantly from oil palm empty fruit bunch biochar (578.64 cm²) and oil palm shell biochar (547.89 m²). The mechanism of biochar that affects the leaf area of corn plants is to increase pH, cation exchange capacity, C-Organic, P-Total and biomass accumulation water level. The increase in CEC will increase the exchange of cations on the surface of biochar. Thereby contributing to maintain NH₄⁺ which leads to an increase in N nutrients in the soil [22]. Increasing the amount of N fertilizer increases the leaf area and number of leaves so that the leaf area becomes wider and the area covered is getting bigger [23]. Biochar can increase the ability of cation adsorption, which is two to three times colloidal minerals and 30-90% of the absorbing power of soil minerals[24].

Table 4. Average leaf area (cm²) of corn in various biochar and bacterial isolate treatments as well as moisture content in different field capacities.

Moisture content field capacity (A)	BIOCHAR (C)			Average	Duncan $\alpha 0.05$
	Palm kernel shell (C1)	Oil palm empty fruit bunches (C2)	Corn cob (C3)		
100% (A1)	605.16	586.07	589.04	593.42b	8.61
80% (A2)	564.88	616.44	644.39	608.57a	8.79
60% (A3)	473.62	533.42	544.50	517.18c	
Mean	547.89z	578.64y	592.64x		
Duncan $\alpha 0.05$	5.70	6.00			

Numbers followed by the same letters in columns (a, b, c) and rows (x, y, z) mean that they are not significantly different in Duncan's DMRT $\alpha 0.05$ further test.

3.4. Root dry weight

Table 4 shows that the average dry weight of corn root was significantly affected by the interaction of field capacity (A), bacterial isolate (B) and biochar type (C). Duncan's test showed that the highest average dry weight of corn root was 44.33 (g), produced by the interaction of biochar oil palm empty fruit bunches (C2), alginate-producing bacterial isolates (B1) at 100% field capacity (A1). These results are significantly different from all other oil palm empty fruit bunch biochar treatments. However, it did not differ significantly from the average root dry weight produced by the interaction of oil palm shells (C1) with isolates of alginate-producing bacteria (B1) and phosphate solvent bacteria (B2) at 100% field capacity. Likewise with the average dry weight of corn roots produced by the interaction between biochar cobs and bacterial nitrogen fixation isolates at 100% field capacity. The role of the interaction of biochar and alginate-producing bacterial isolates in root formation is seen in water-deficient soil conditions (60%). This can be seen in the interaction of corncob biochar with alginate-producing bacterial isolates (B1) and their interactions with nitrogen fixation bacterium isolates (B3) at a field capacity of 60%, resulting in an average root dry weight of 38.67 g and 33.67 g which is not significantly different from the results at 100% field capacity.

These results confirm that biochar with the addition of alginate-producing bacteria is able to retain water thereby increasing root formation under drought conditions. This result was confirmed by previous studies, where olisaccharide alginate had a significant effect on root and seed growth [16]. Similar to the study of [25], who confirmed the role of biochar in increasing root biomass, root volume and surface area. The results concluded that biochar plays a role in the development of root

morphology to reduce nutrient and plant water shortages. Biochar application affects the microbes associated with the root and significantly increases the number of root nodules. Plants absorb almost all water and nutrients through the root system, root morphology and vitality are a reflection of the capacity of plants to absorb water and nutrients, and are used as indicators of root development [26]. Root length is related to water absorption and nutrition [25].

Table 5. Average dry weight of root (g) of corn in various biochar and bacterial isolate treatments and different moisture content in the field capacity.

Field capacity (A)	Bacterial isolate (B)	Biochar (C)			Np. Duncan
		palm kernel shell (C1)	Oil palm empty fruit bunches (C2)	Corn cob (C3)	
100% (A1)	Alginate-producing (B1)	36,67ab ^x	44,33a ^x	24,67e ^y	0.68
	Phosphate solvents (B2)	42,33a ^x	33,33b ^{xy}	26,67de ^y	0.71
	Nitrogen fixation (B3)	21,67e ^z	31,00bc ^y	40,33a ^x	0.73
80% (A2)	Alginate-producing (B1)	30,33bcd ^x	24,33c ^x	30,33bce ^x	0.75
	Phosphate solvents (B2)	28,33bcd ^{xy}	26,67bc ^y	35,33abc ^x	0.76
	Nitrogen fixation (B3)	35,33ab ^x	26,33bc ^y	36,33abc ^x	0.77
60% (A3)	Alginate-producing (B1)	23,00d ^y	33,67b ^x	38,67ab ^x	0.78
	Phosphate solvents (B2)	26,33cd ^x	27,00bc ^x	29,33ce ^x	0.79
	Nitrogen fixation (B3)	31,33abc ^x	27,67bc ^x	33,67abcd ^x	

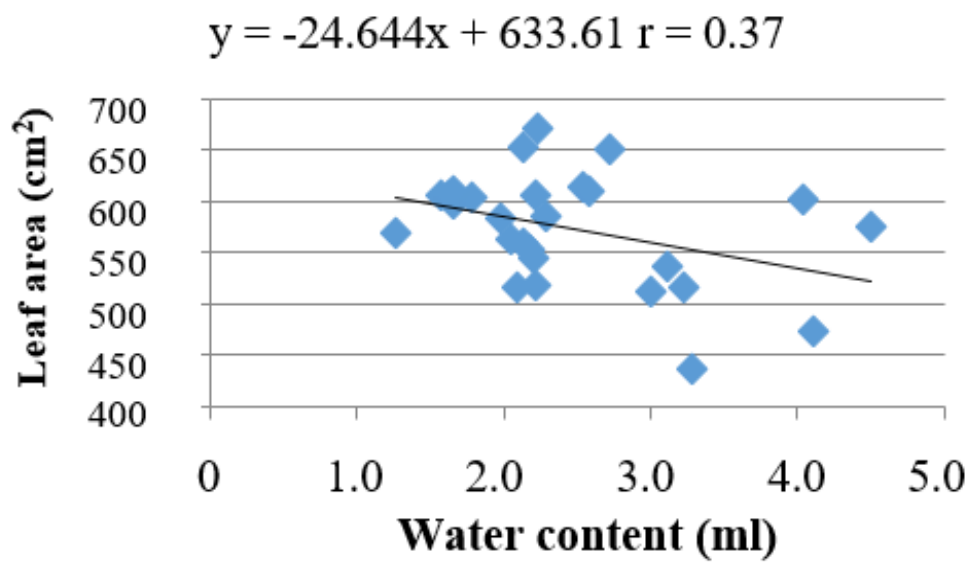
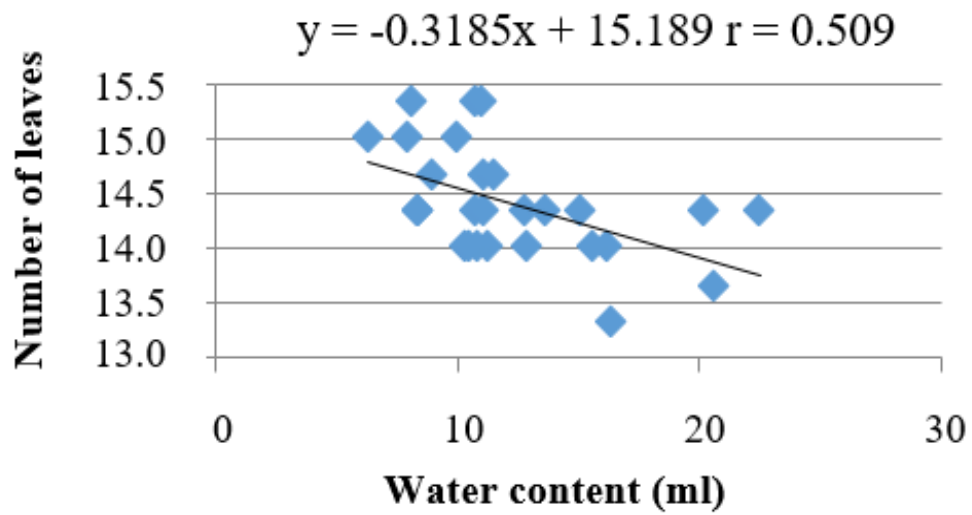
Numbers followed by the same letters in columns (a, b, c) and rows (x, y, z) mean that they are not significantly different in Duncan's DMRT α 0.05 further test.

3.5. Correlation between variables

Table 6 shows the correlation between variables namely water content, plant height, number of leaves, leaf area and root dry weight. Correlation between water content and maize growth variables is illustrated in figure 3 very significant correlation was seen between the number of leaves with plant height, number of leaves with leaf area and plant height with leaf area. This shows that the higher the plant, the more leaves are formed. The correlation between water content and growth variables is very interesting where the correlation occurs inversely. Although the effect is not real, but the growth of corn plants in the study does not require large amounts of water. These results confirm the initial possibility of biochar from oil palm shells, oil palm empty fruit bunches and corncobs able to retain water and nutrients so that their use is more efficient during the growth of maize. And it is possible that alginate-producing bacteria that develop on the surface of biochar play a role in maintaining soil moisture so that the water needed in this experiment is more efficient. The results of investigations of clay loam in alginate beads have a high adsorption ability to absorb pollutants in polluted water [27]. Other research results also confirm that biochar increases the efficiency of using N fertilizer [28]. These results may confirm the initial mechanism that occurred in this study. Nevertheless further research still needs to be a more detailed investigation of some physiological variables of corn plants related to water needs.

Table 6. Correlations between observed variables of corn plants

Variable	Water content	Number of leaves	Plant height	Leaf area	Root dry weight
water content	1				
number of leaves	-0.509	1			
plant height	-0.528	0.552**	1		
leaf area	-0.370 ^{tn}	0.491**	0.817**	1	
root dry weight	-0.197 ^{Tn}	0.347 ^{tn}	0.219 ^{tn}	0.144 ^{tn}	1



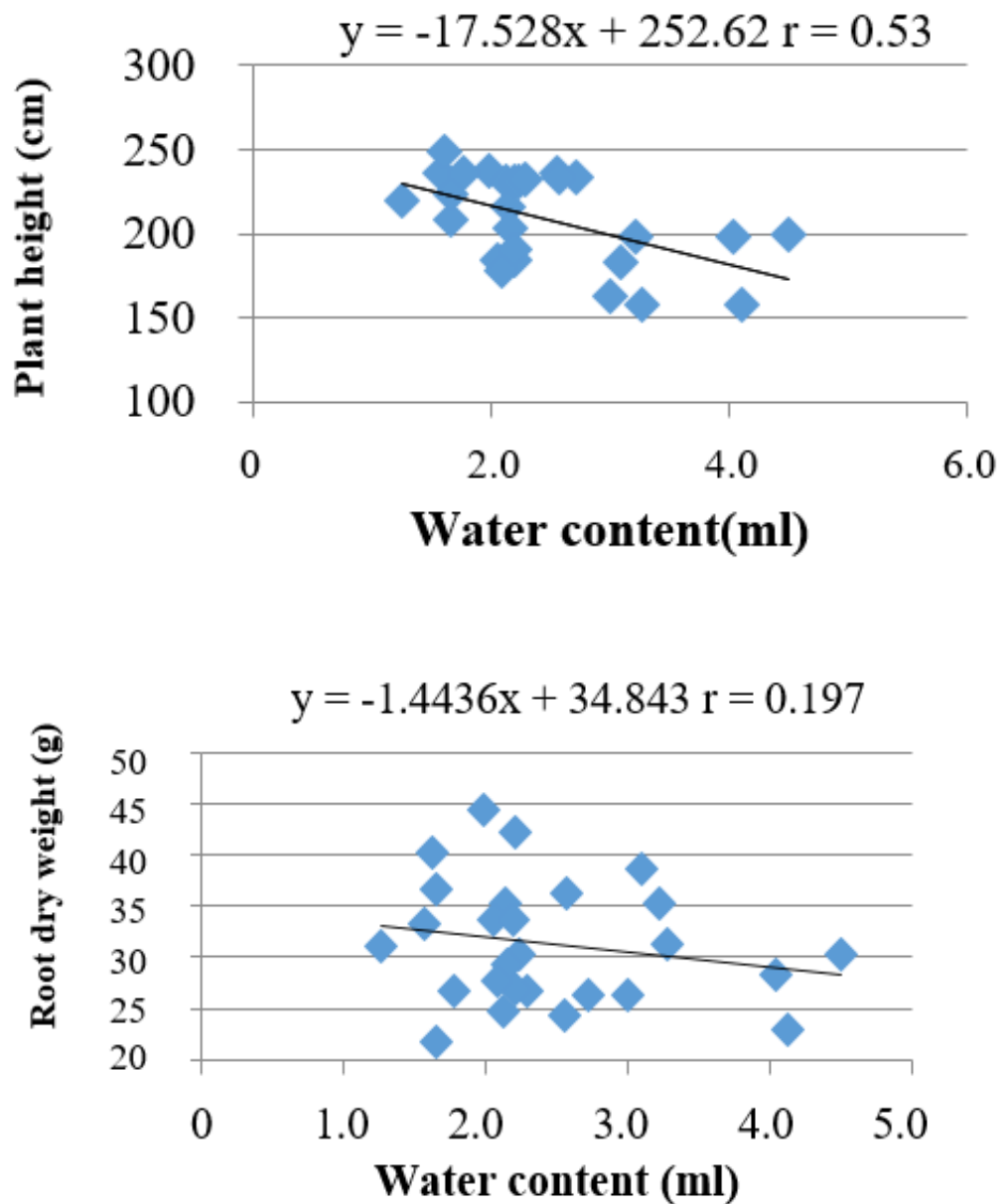


Figure 3. Correlation between water content with various observations of corn plants.

4. Conclusions

- The increase in plant height is significantly affected by the availability of water capable of retention by biochar.
- The increase in the number of leaves is significantly affected by the ability of bacterial isolates capable of fixing nitrogen retained by biochar from corncobs
- The leaf area of corn plants is significantly affected by the availability of ground water and biochar freely.

- d) Root dry weight is significantly affected by interactions between biochar and alginate-producing bacterial isolates in drought-soil conditions.
- e) Correlation between variables shows that there is a role of biochar and alginate-producing bacteria on the efficiency of water and nutrient use during the growth of maize plants grown on clay applied by biochar and alginate-producing bacteria.

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