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Proline Accumulation and Growth of Bean Leaf (*Phaseolus vulgaris* L.) With Biochar Application in The Shallow Water Table Environment

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Abstract

Agronomic constraints for vegetable cultivation in riparian wetlands are low soil quality and shallow water table conditions. This study aims to identify the effect of biochar application and shallow water table on proline accumulation and plant growth of bean leaf (*Phaseolus vulgaris* L.) on the generative stage. This study was carried out from April to August 2018 in factorial randomized block design. The first factor was doses of biochar (0, 1, and 2 $kg.m^{-2}$), and the second was shallow water table condition (20 cm and 10 cm below the soil surface) given in the generative stage. The results showed that biochar application and the shallow water table significantly affected leaf parameters; proline accumulation and increases in the sucrose levels occurred on the leaves. Proline content increased started at 3 days after treatment (DAT) and decreased at 7 DAT.until recovery day (at 7 DAT). The biochar application of 2 $kg.m^{-2}$ provided good aeration in the soil, which enhanced the bean's ability to survive under excess water. Proline accumulation is one of the adaptation mechanisms of beans to abiotic stress in shallow water table conditions.

Keywords: crop adaptability, ameliorant, submerged land, wetlands.

Introduction

The riparian wetland is classified as suboptimal land but has high potential for rice, food crops, and vegetables. The total riparian wetland area in Indonesia is approximately 13.29 million ha and spread across several regions: 2.79 ha in Sumatera, 3.58 ha in Kalimantan, 6.31 ha in Papua, and 0.61 ha in Sulawesi (Alwi et al., 2017). The commonly cultivated vegetables in the riparian wetland are tomato, chili, eggplant, beans, corn, kale, lettuce, and basil (Emile et al., 2012; White, 2013).

The main constraints of plant cultivation in riparian wetlands are unpredictable flooding periods during the rainy season and drought during the dry season due to climate change (Lakitan et al., 2018c). Flooding and drought stress decreased crop yields (Bailey-Serres et al., 2012). The whole riparian wetland area is flooded during the rainy season for several months. During high flooding, plants could be cultivated by utilizing floating cultivation systems (Siaga et al., 2018). However, farmers need additional costs to construct the raft.

In riparian wetlands, rice cultivation starts when the water level has receded or at the end of the flooding season and is harvested at the beginning of the dry season (Lakitan et al., 2018a). At the time, local farmers (1) the crops that were planted after the rice harvest immediately experience water logging in the vegetative stage, and drought stress in the generative stage. This was due to the soil dries gradually; (2) the third crop, planted after second crop harvest, requires additional water supply during for both vegetative and generative stages, but this crop will probably under shallow water table stress in the generative stage if rainy season occurs earlier than expected. Therefore, it is crucial to understand vegetable crops that can adapt to this environment.

Plants have several mechanisms to survive under shallow water table conditions, including anatomical and morphological adaptations and metabolic mechanisms (Ashraf, 2012). The metabolic mechanism known as osmotic adjustments, such as the enhancement of proline concentration in leaf tissue (Singh et al., 2000). Proline ($C_5H_9NO_2$) is a photogenic amino acid compound accumulated both in normal conditions (Kishor et al., 2015) and abiotic stress due to non-optimal environments such as drought (dehydration), salinity, waterlogging, and extreme temperature (Sarker et al., 2005; Gupta et al., 2014; Fichmann et al., 2015). Proline accumulation occurs due to the activation of proline biosynthesis and the inactivation of proline degradation. Proline is an osmoprotectant compound that is synthesized in plants via the glutamine and ornithine pathways (Delauney et al., 1993). In the primary pathway, glutamine is synthesized into proline through two intermediate compounds, glutamine semialdehyde (G.S.A.) and proline-5-carboxylate (P5C); then P5C is reduced to proline by enzyme P5C reductase (P5CR) (Delauney et al., 1993). By accumulating proline in the cell, the osmotic potential of the cell decreases, thereby increasing the cell's capacity to maintain turgidity (Turner, 2018). Cell turgidity is essential for photosynthesis, enzyme activity, and cell enlargement (Claussen, 2005). Previous studies have reported that environmental stress causes proline accumulation in peas (Karatas et al., 2014), tobacco (Borgo et al., 2015), tomatoes (Gharsallah et al., 2016), and lentils (Ankita et al., 2017). Therefore, proline is a crucial indicator for detecting plants' adaptability to shallow water table stress.

After flooding, another crucial issue in the riparian wetland is the low physical, chemical, and biological soil quality. Riparian wetlands are sub-optimal land for crop cultivation because of having low chemical attributes such as low pH (Alwi et al., 2017), deficiency of macronutrients, and containing excessive amounts of toxic metals (Lakitan et al., 2018c). According to Noor et al. (2006), most of the riparian wetlands soils are classified as alluvial that have poor drainage. Poorly drained soil often has poor aeration, so oxygen availability is limited for the aerobic microorganisms' activities in the soil. The low availability of oxygen is also caused by the soil which is still wet after the flood recedes. In wet soil, pores are still filled with water so that oxygen becomes limited for plants and for aerobic microbial activities in the soil. This reduces the physical quality as well as the biological quality of the soil. To increase fertility and enhance the physical properties of soil, ameliorant materials such as agricultural lime, manure and biochar could be applied to soil (Barrow, 2012; Nariratih et al., 2013; Goulding, 2016; Kartika et al., 2018). Therefore, ameliorant materials addition could increase plant growth (Lakitan et al., 2018a). This study focused on applying biochar to improve soil quality and the adaptability of beans to the shallow water table.

Material and Methods

This study used 72 common bean cultivars. Bean seeds were soaked in warm water for 3 hours and sown into 40 cm x 25 cm polybags which contained 14.7 cm^{-3} of a mixture of soil, chicken manure, and biochar (the height of growing media ±30 cm). Soil and chicken manure were mixed in a ratio of 2:1 (v/v). The biochar was applied according to treatment dose. This research used crushed wood charcoal with a size of approximately 1mm. Biochar was applied to the soil one week prior to planting time and mixed evenly on the surface of growing media. Plants were irrigated twice daily and supplied 6 g N.P.K. fertilizer per plant at transplanting. Pest and diseases were controlled using pesticides.

Experimental Site and Experimental Design

The study was carried out in the Macan Kumbang sub-district (2°58'24.2 "S 104°43'12.5"E), Palembang, Indonesia, from April to July 2018. This study was arranged in a factorial randomized block design. The first factor was biochar doses consisting of 0, 1, and 2 $kg.m^{-2}$; the second factor was one control, and two different water table depths were 20 cm below the soil surface and 10 cm below the soil surface for 1 week. Each treatment was repeated three times. Three samples for proline and sucrose analysis and 12 for all destructive analysis (0, 3, 6, and 7 days after transplanting, DAT. In the shallow water table treatment, plants were prepared in a 3.0m x 1.5m x 0.25m pond at their generative stage, i.e., when about 50% of the plants have developed fully bloomed flowers).

Proline and Sucrose Analysis

Leaf proline and sucrose analysis was conducted at the Integrated Research Laboratory of the Faculty of Agriculture, Universitas Sriwijaya. The analyses were carried out in several stages: before water table treatments (D0), three days after treatment (D3), six days after treatment (D6), and 7 days of the recovery period (R7). The method of proline analysis followed the method of Bates (1973). Meanwhile for method of sucrose analysis followed Somogyi Method (Nelson, 1944).

Plant growth and yield measurements

The relative leaf expansion rate (RLER.) was measured based on leaf area data, which was measured continuously for 14 days starting at the day the treatment was initiated until the recovery period. The leaves observed are healthy young leaves that have been in full bloom. Leaf area was measured every morning from 6 to 6:30 am. Leaf area measured non-destructively following Lakitan et al. (2017) formula in which LA = 1.5198 x leaf length x leaf width.

Specific leaf fresh weight (SLFW) and specific leaf water content (SLWC) were measured based on data, i.e., leaf area, fresh weight, and leaf dry weight. Leaf samples were taken simultaneously with leaf sampling to measure proline contents. SLFW and SLWC were measured by weighing fresh leaves immediately after being picked, then drying in an oven at 80°C until reaching the constant dry weight. SLFW is a comparison between the fresh weight of leaves (FW) and leaf area (LA), SLWC is calculated based on the difference in fresh weight (FW) and dry weight (DW) divided by leaf area (LA) (Meihana et al., 2017; Widuri et al., 2017). The weight and number of pods were measured continuously from the 1st harvest to the 17th harvest.

Statistical analysis

Differences in the effect of biochar applications and shallow water table were analyzed of variance (ANOVA) using Statistical Analysis System (S.A.S.) 9.2. For significant treatments on any measured parameters, differences among mean values were tested using the Least Significant Differences (L.S.D.) test at p ≤ 0.05

Result and Discussion

Proline Accumulation Due to Shallow Water Table and Biochar Application

The results showed that the shallow water table affected the proline content of the leaves. The significant effects started showing three days after treatment (DAT). On the water table 20 cm below the soil surface and control, proline accumulation was decreased and gradually increased on the 6 DAT (Figure 1). Contrary to the water table 10 cm below the soil surface (W2), proline accumulation significantly increased on the 3 DAT The proline increase was likely due to the dry season when the temperature was in the range of 32-35°C, and plants accumulated the proline as a response to the high temperature.

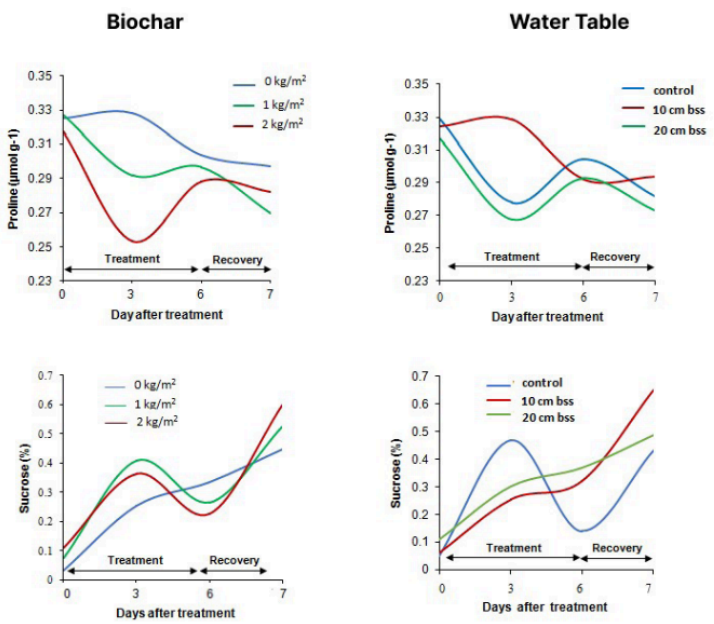


Figure 1. Proline and Sucrose contents are affected by biochar application and shallow water table.

The shallow water table treatment showed positive impacts until the 3 DAT, as indicated by the reduction of proline content compared to the others treatment. However, bean growth decreased after 6 DAT, and proline content significantly increased. The mechanism of survival in bean was by producing proline in the leaves, or osmotic adjustment. Increasing proline accumulation is a mechanism for beans to survive in excess water stress conditions. In stress conditions, plants accumulate proline rapidly because of proline's high solubility (Bhaskara et al., 2015) and compatibility with the cell environment (Kishor et al., 2015), so proline is an indicator to detect plant adaptability to stress.

The proline content in bean leaves decreased when the plant entered the recovery period, i.e., when the shallow water table treatment was stopped. Generally, proline accumulates on young leaves (Claussen, 2005; Zegaoui et al., 2017); proline accumulation indicates the physiological adaptation mechanism of the plant to excess water conditions (hypoxia) (Ashraf, 2012). By accumulating proline in the cell, the osmotic potential of the cell decreases, thereby increasing the cell's capacity to maintain turgidity (Turner, 2018). Cell turgidity is essential for photosynthesis, enzyme activity, and cell enlargement (Claussen, 2005).

Compared to the water table of 20 cm below the soil surface, the water table of 10 cm below the soil surface makes more parts of roots to be in a submerged condition. The closer the water table is to the soil surface, the more parts of the roots are submerged in the water. In excess water condition, gas exchange between soil and air becomes limited (Subbaiah and Sachs, 2003). As a result, the supply of O_2 in the soil becomes low (hypoxia), and in a long time, it will become anoxia (Ashraf, 2012). The limited supply of O_2 causes metabolic disorders in plants. To survive in this condition, plants can adapt anatomically and morphologically and make osmotic adjustments (Ashraf, 2012). Osmotic adjustment occurs by increasing the concentration of proline in leaf tissue (Singh et al., 2000; Abraham et al., 2003; Fichman et al., 2015).

Another finding of this research indicated proline content of beans which growing in the soil without biochar application increased (Figure 1); on the contrary proline content of plants growing in the ground mixed with 1 kg.m^{-2} of biochar and 2 kg.m^{-2}of biochar sharply decreased at the 3 DAT However, proline content increased at the 6 DAT of the shallow water table for 1 kg.m^{-2} of biochar and 2 kg.m^{-2} of biochar but decreased for controlled plants. Biochar application on 2 kg.m^{-2} gave a positive effect on plant resistance under shallow water table stress, likely due to the role of biochar in the physical properties of the soil. Some benefits of biochar applications on soil physical properties, i.e., facilitating drainage in poorly drained soils (Herath et al., 2013), forming macro aggregates in the soil (Jien et al., 2013; Wong et al., 2016), and increasing soil pore space (Sun and Lu, 2014; Hardie et al., 2014). The 2 kg.m^{-2} of biochar is better in providing aeration on the part of the soil that is not submerged in water (10 cm near the soil surface) than the soil mixed with 1 kg.m^{-2} of biochar. Therefore, the oxygen supply in well-aerated soil (that is not submerged in water is maintained for root respiration.

The change of proline and sucrose contents are physiological mechanisms in the plant when experiencing environmental stress conditions. This research showed that shallow water tables also affected the sucrose contents of the leaves (Figure 1). Sucrose contents increased at 3 DAT and decreased at the 6 DAT of stress. Despite this, in the control conditions, sucrose content fell from 3 to 6 DAT The decrease in sucrose content is likely due to the translocation of sucrose from the source (leaves) to the sink (pods). Biochar application at 1 kg.m^{-2} produced higher proline and sucrose than the 2 kg.m^{-2} (Figure 1).

Plant Growth Analysis

Biochar application and shallow water table treatments significantly interact with plant growth analysis except on the relative water content (RWC) in 6 DAT (Table 1). Leaves and water content variables tend to be more affected by water table conditions rather than biochar application. A shallow water table generally decreased specific leaf fresh weight (SLFW) and specific leaf water content (SLWC) but increased specific leaf area (SLA) and leaf area (LA). SLFW and SLWC showed a downward trend during exposure to shallow water tables until the recovery period; however, the decline was not significant. This condition was a physiological response of plants to the shallow water table. This decrease is not only caused by excess water but also affected by the age of the leaf. After entering the recovery period, the plant is not in the treatment pond. In that condition, the plant cannot compensate for the water loss rate due to high transpiration. After being outside the pond, soil moisture decreased due to evaporation. In recovery time, the bottom root section has not been able to absorb water properly because the bottom roots are damaged during exposure to a shallow water table. In this situation, water absorption rests on the upper root not submerged. Because of the upper part of the soil loses a lot of water due to evaporation, the water content in plants, especially in leaves, decreases. Water is needed by plants to maintain leaf turgidity (Turner, 2018). In this case, the SLWC of the plant becomes low, so the SLFW decreases. This phenomenon has been reported on the previous study. Lakitan et al. (2018b) found those leaves of beans reached it maximum size in approximately nine days.

Since proline accumulation is one of the osmotic adjustment mechanisms for plants that experience excess water, SLFW and SLWC are essential variables that respond to these conditions. However, in this study, the increase in leaf proline content was not followed by the rise of these two variables (Figure 1), showing that SLFW and SLWC are more affected by disruption of water absorption due to root damage that experiences shallow water table conditions.

At the beginning of treatment, 1 kg.m^{-2} of biochar in the control plant showed significantly higher LA than the 0 kg.m^{-2} of biochar-water table treatment. After three days, 1 kg.m^{-2} of the biochar-water table 20 cm below the soil surface and 2 kg.m^{-2} of the biochar-control plant than the other treatments. During the recovery period, the interaction of 0 kg.m^{-2} of the biochar-water table of 10 cm below the soil surface reached higher LA compared with 0 kg.m^{-2} of biochar-control, 1 kg.m^{-2} of biochar-water table 10 cm below the soil surface, and 2 kg.m^{-2} of biochar-water table 20 cm below the soil surface.

On the day treatment started, higher RWC were obtained in the biochar application compared to controls. At the 3 DAT, 2 kg.m^{-2} of biochar-water table 10-cm below the soil surface got too higher RWC. than the others. It showed that 2 kg.m^{-2} biochar was better for guaranteeing water availability in the media for plant growth. In this research, we recognized the benefit of biochar for improving soil physic characteristics such as facilitating soil drainage (Herath et al., 2013), creating macro aggregate in soil (Jien et al., 2013; Wong et al., 2016), and increasing soil pore (Sun and Lu, 2014; Hardie et al., 2014). RWC in each treatment did not show a significantly different at the 6 DAT, whereas 2 kg.m^{-2} of biochar-water table 10-cm below the soil surface resulted in the highest of RWC Application 2 kg.m^{-2} of biochar 10-cm below the soil surface showed the most elevated of RWC in 3, 6, and 7 DAT, respectively.

At the beginning of treatment SLA in 1 kg.m^{-2} of biochar increased. At the 3 DAT, SLA in 1 kg.m^{-2} of biochar - 10 cm below the soil surface significantly increased compared to 0 kg m^{-2} of biochar-20 cm below the soil surface. It revealed that the water table at 10 cm below the soil surface in three days positively affected leaf area development. The temperature on the day before treatment is relatively high (32^{o}C–35^{o}C), increasing the evapotranspiration rate. Common beans obtained sufficient water supply for leaf development at the beginning shallow water table period. However, at the 6 DAT, the highest SLA was reached on 2 kg.m^{-2} of biochar-control plant, indicating that biochar could increase water retention of soil media to provide water for the plant. Sufficient water for plants increases leaf turgor and leaf area expansion. After the recovery period, 0 kg m^{-2} of biochar - 20 cm below the soil surface got too higher SLA than 0 kg.m^{-2} of biochar-10 cm below the soil surface and control. The effect of the water table 20 cm below the soil surface was not as severe as the water table 10 cm below the soil surface. The water table 10 cm below the soil surface causes the roots to be submerged and damaged. During the recovery period, common beans might be in optimal condition, but plant roots could completely absorb the water supplied during the watering time. While in the 20 cm below the soil surface, fewer roots were damaged, so they recovered more quickly.

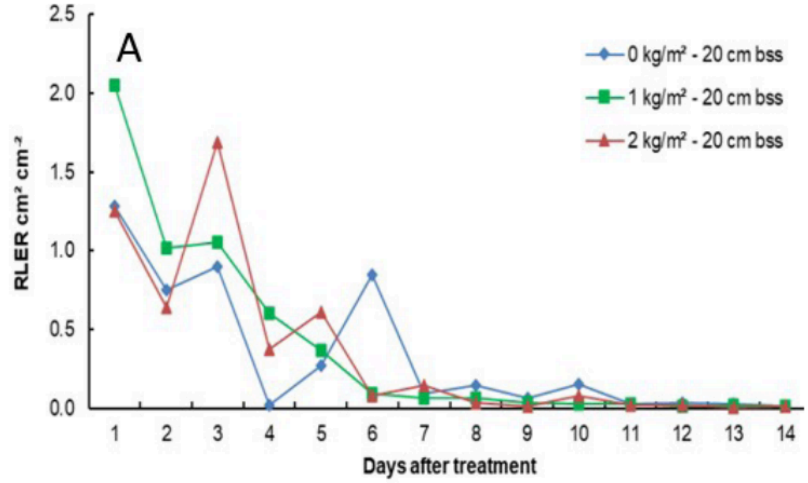
Leaf turgidity also revealed the effect of biochar and water table treatment. Low leaf turgidity affects the relative leaf expansion rate. The turgor of actively dividing cells determines leaf turgidity. If the cell turgor decreases, cell elongation becomes obstructed, and RLER decreases. RLER on treated and non-treated plants (control plants) began to drop dramatically until the 6 DAT and finally stopped at the end of the recovery period (Figure 2). Genetic factors might also cause the termination of the leaf expansion rate. This research revealed that in less than 14 days, the leaves had reached their maximum size. Adding biochar increased RLER at the control and water table 20 cm below the soil surface.

The shallow water table stress condition significantly affected plant dry weight on the 6th day of treatment (Figure 3). It showed a rapid dry weight increase on the water table 20 cm below the soil surface due to an increment of leaf, stem, and fruit weight. The dry weight increased continuously until 7 DAT The dry weight of the plants increased due to the application of biochar. The higher the dose of biochar, the higher the dry weight obtained (Figure 3).

Table 1. Leaves and water content-related variables during water table stress condition and recovery

Day	Treatment	SLFW (mg cm⁻²)		SLWC (mg cm⁻²)		SLA (cm² mg⁻¹)		LA (cm²)		RWC (%)	
D0	B0W0	30.02±8.94	a	27.20±7.85	a	0.035±0.010	b	134.77±35.51	b	90.58±2.94	ab
	B0W1	22.67±5.14	ab	20.61±4.44	ab	0.045±0.010	ab	136.50±26.06	b	85.91±4.26	b
	B0W2	27.90±1.45	ab	25.01±0.93	ab	0.035±0.001	ab	151.61±17.62	ab	91.93±5.72	ab
	B1W0	23.67±3.05	ab	20.90±3.22	ab	0.042±0.006	ab	189.65±13.57	a	93.29±1.93	a
	B1W1	20.71±4.33	b	19.15±3.92	b	0.049±0.009	a	162.13±15.83	ab	91.77±5.61	ab
	B1W2	22.89±5.26	ab	20.84±4.77	ab	0.045±0.009	ab	142.10±37.90	b	92.24±4.84	ab
	B2W0	21.36±1.54	b	19.00±1.35	b	0.047±0.003	ab	149.05±36.20	ab	85.64±2.46	b
	B2W1	21.02±4.35	b	19.26±3.62	b	0.049±0.012	ab	146.37±20.74	ab	94.08±5.45	a
	B2W2	21.94±3.00	b	19.87±2.65	b	0.046±0.006	ab	169.94±28.98	ab	89.87±3.58	ab
D3	B0W0	23.91±4.15	bdc	21.44±3.43	bc	0.042±0.007	abcd	179.40±27.73	abc	88.93±2.55	ab
	B0W1	30.06±5.32	a	26.72±4.68	a	0.034±0.007	d	178.80±25.58	abc	91.79±3.25	ab
	B0W2	26.88±1.03	ab	23.87±0.83	ab	0.037±0.001	dc	141.81±3.650	c	90.19±3.15	ab
	B1W0	20.20±0.92	d	18.13±0.92	c	0.049±0.002	a	187.04±18.13	ab	91.93±4.65	ab
	B1W1	24.28±3.05	bdc	21.37±2.71	bc	0.042±0.006	abcd	207.87±27.60	a	92.57±2.99	ab
	B1W2	20.49±2.33	d	18.50±2.00	c	0.049±0.005	a	143.84±23.18	c	88.18±7.07	ab
	B2W0	22.09±2.30	bdc	19.31±1.83	bc	0.046±0.004	abc	219.57±30.82	a	86.77±9.00	b
	B2W1	21.09±0.66	dc	18.93±0.62	c	0.047±0.002	ab	149.93±11.97	bc	93.26±2.27	ab
	B2W2	26.23±4.69	abc	23.64±4.11	ab	0.039±0.006	bdc	152.17±36.34	bc	95.26±1.05	a
D6	B0W0	27.88±5.25	a	24.21±4.14	a	0.037±0.007	b	132.12±30.02	d	89.11±1.51	a
	B0W1	24.70±3.32	ab	21.65±3.02	abc	0.040±0.005	ab	171.00±0.265	bcd	86.25±2.87	a
	B0W2	21.32±2.89	b	18.84±2.37	bc	0.047±0.006	ab	230.15±57.15	a	87.86±2.45	a
	B1W0	21.55±4.21	b	18.93±3.47	bc	0.047±0.010	ab	183.16±25.48	abc	85.29±1.02	a
	B1W1	26.51±2.25	ab	23.67±2.05	ab	0.038±0.003	ab	185.28±5.620	ab	88.52±0.89	a
	B1W2	23.51±4.71	ab	20.34±4.06	abc	0.044±0.009	ab	133.42±19.90	cd	85.48±0.61	a
	B2W0	20.45±0.83	b	17.99±0.63	c	0.049±0.002	a	183.34±31.65	abc	86.99±8.56	a
	B2W1	23.79±3.21	ab	21.07±2.98	abc	0.043±0.005	ab	156.24±27.68	bcd	89.01±4.65	a
	B2W2	25.90±3.91	ab	21.81±3.36	abc	0.039±0.006	ab	187.46±30.86	ab	91.42±1.65	a
D7	B0W0	25.06±4.45	ab	20.74±4.19	ab	0.040±0.008	bc	132.82±11.87	bcd	87.73±3.27	abc
	B0W1	19.29±1.59	c	15.61±1.01	c	0.052±0.005	a	152.33±37.82	abcd	81.23±5.53	cd
	B0W2	26.07±0.66	a	22.07±0.50	a	0.038±0.001	c	186.27±46.42	a	88.16±1.45	abc
	B1W0	21.54±1.69	abc	17.63±1.20	abc	0.046±0.004	abc	154.85±13.34	abcd	76.83±7.62	d
	B1W1	21.81±1.66	abc	18.28±1.29	abc	0.046±0.004	abc	178.69±9.230	ab	88.83±2.33	abc
	B1W2	21.89±2.41	abc	18.44±2.17	abc	0.046±0.005	abc	114.03±14.74	cd	87.35±0.37	abc
	B2W0	20.49±4.88	c	17.29±4.16	c	0.050±0.013	ab	159.36±28.69	abc	84.76±2.51	bcd
	B2W1	23.75±3.60	abc	19.9±3.94	abc	0.043±0.006	abc	107.78±12.32	d	95.55±14.2	a
	B2W2	21.68±2.50	abc	17.93±2.41	abc	0.047±0.006	abc	142.62±44.05	abcd	91.92±3.13	ab

[z] Means followed by the same letters within columns of each treatment, and the measured trait is not significantly different based on L.S.D. at P < 0.05. B = Biochar; B0 = 0 kg m⁻²; B1 = 1 kg m⁻²; B3 = 2 kg m⁻². W = water table; W0 = control; W1= 20 cm bss, W2= 10 cm bss; bss = below soil surface



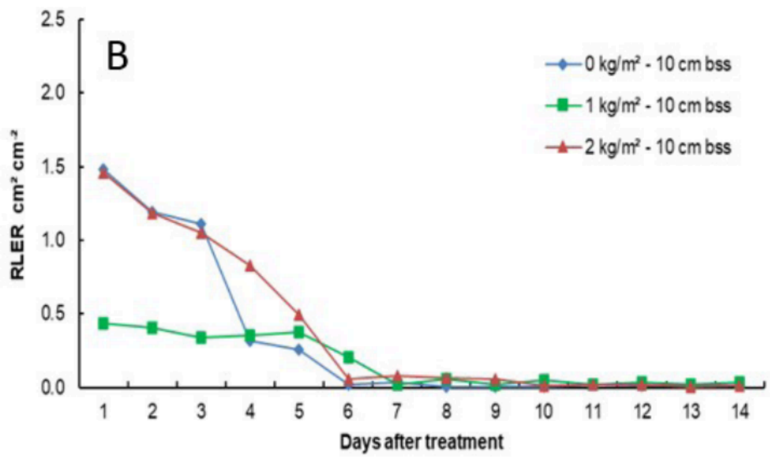
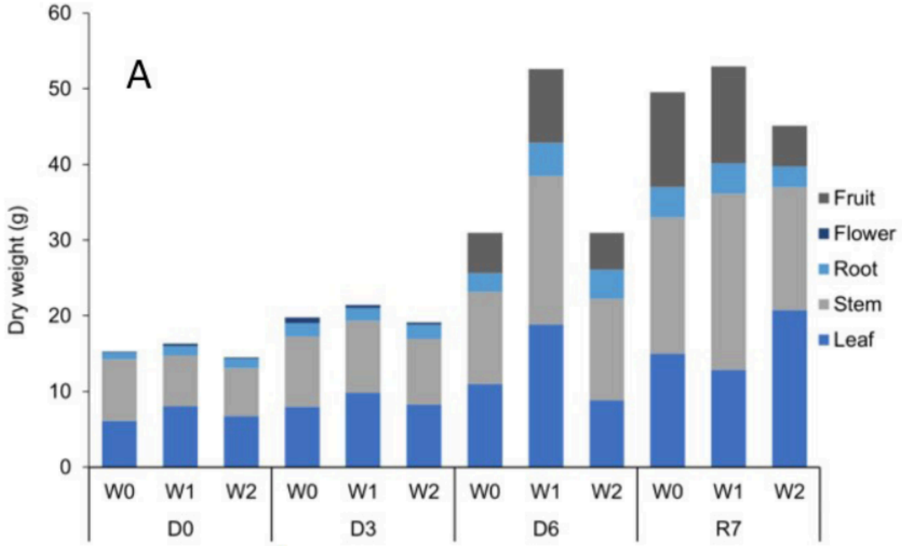


Figure 2. Continuous measurement of relative leaf expansion rate (RLER) in beans at 1 to 14 days after treatments. RLER began to drop significantly on the sixth day after treatment and finally stopped at the end of the recovery period. Two water table depths were 20 cm (A) and 10 cm below the soil surface (B) for one week; bss = below soil surface.



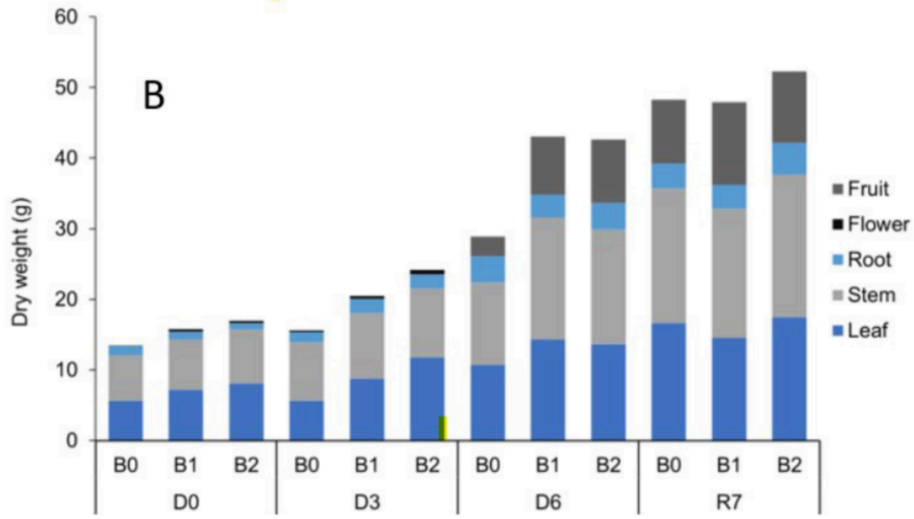


Figure 3. Plant biomass at different water table (A) and biochar (B) applications. At day 0 (D0) and three days after treatment (D3) water table had no effects on plant dry weight, but at six (D6) and seven days (D7) after treatment plant dry weight significantly increased with water table 20 cm bss (W1). Biochar application increased plant dry weight at D0, D3, D6, and R7. The highest dry weight was obtained by application of 2 kg.m-2 of biochar (B2).

Conclusion

Proline accumulation is an adaptation mechanism of beans to the shallow water table. The biochar application of 2 kg.m^{-2} provides good aeration in soil, enhancing the bean's ability to survive under shallow water table conditions. Beans can be categorized as tolerant crops to shallow water tables for vegetable cultivation in the riparian wetlands on the rainy season.

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