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Soil hydric excess and soybean yield and development in Brazil

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Abstract

Soybean is the main crop of Brazil and the country is the first largest producer in the world. Soybean is cultivated in several different climates, soil types and, lately, in lowland soils in rotation with flooded rice. The objective of this work was to evaluate the effect of hydric excess by flooding on the soybean growth and yield of the cultivar BMX Potência RR. The experimental design used was randomization, with six treatments and five replications consisting of pots of 7.5 L (6.0 dm³ soil per pot), consisting of two experiments: hydric excess at the flowering stage (R2); and hydric excess at the beginning of the grain-filling stage (R5). The six treatments of the hydric excess were 0, 2, 4, 8, 16 and 32 days of flooding, in R2 (R2.0, R2.2, R2.4, R2.8, R2.16 and R2.32, respectively) and R5 (R5.0, R5.2, R5.4, R5.8, R5.16 and R5.32, respectively), at 55 and 76 days after emergency, respectively. The results showed that the plant height, number of leaves, stem diameter, height of insertion of the first pod, number of pods per plant, number of grains per pod and number of grains per plant were not altered by the soil flooding up to 32 days. It was determined that the mass of a thousand grains and soybean grains yielded decreased gradually with an increase in the number of days of flooding, in stages R2 and R5, with grain yield reduction of 17% and 29%, after 16 days, and 41% and 36% after 32 days of flooding.

Keywords: Flooding; hypoxia; *Glycine max* (L.) Merrill.
Abbreviations: pH_hydrogen potential, OM_organic matter, N_nitrogen, P_phosphorus, K_potassium, Ca_calcium, Mg_magnesium, Al_aluminium, V_basis saturation, Mn_manganese, Fe_iron.

Introduction

Brazil is the first largest producer of soybeans in the world. This crop covers the largest area in the country [30.1 million ha (74.3 million acres; Conab, 2014)]. In the 2012-2013 harvest, the increase in soybeans' commercial value in the international market has resulted in an increase in the cultivated area (Conab, 2014), in a range of climatic conditions and soils, particularly flooded soils that occur in many other areas of the world. In the state of Rio Grande do Sul (RS), Brazil, there are extensive areas of flooded soil, with natural drainage deficiency, used for livestock or irrigated rice. In addition, this area is the largest national producer of the flooded rice. Although rice has a low commercial value, the need for crop rotation to control weeds, pests, and diseases has induced the use of alternative crops, such as soybeans. Soybean cultivation in flooded soils of the West Frontier of RS state is still incipient, and with low yield, because of the hydric deficit or excess, but it requires a low cultivation investment and the grain has a high economic value (Sosbai, 2012). For the cultivation and to obtain a higher yield of soybeans, sufficient water content is essential. This water is derived from the precipitation that fluctuates during cultivation, resulting in periods of soil water deficit or excess to plants. The effects of water stress from water deficiency in the soybean development and yield is clear in several studies (Dogan et al., 2007; Lobato et al., 2008; Mastrodomenico et al., 2013), considering its frequent occurrence during cultivation, worldwide. Therefore, excess water that occurs in soils flooded or compacted generates hypoxia, i.e. insufficient oxygen (O_2) concentration to maintain a normal rate of root respiration. Several studies have been conducted to improve the soybean management (Lanza et al., 2013). However, there is a need for additional research to enable the cultivation in conditions of temporary soil flooding, where hypoxia affects the growth of soybeans in various ways (Henshaw et al., 2007a; Fante et al., 2010; Lanza et al., 2013). Flooding decreases the amount of O_2 in the soil, causing hypoxia. This limits the soybean growth. With hypoxia, biological nitrogen (N) fixation decreases because the nodes need O_2 to maintain aerobic respiration, necessary to supply a large amount of adenosine triphosphate (ATP) essential for nitrogenase activity. Although there are biochemical adjustments, with protein production in conductive O_2, most Fabaceae family plants cannot maintain their supply of O_2 to plant nodes in flooded conditions (Loureiro et al., 1995). In addition, a number of effects on biochemical and physiological processes take place, such as the reduction of photoassimilate transport to roots and reduced photosynthetic rate (Davanso et al., 2002), inhibiting the absorption of nitrogen and other minerals and an increased absorption of iron that can cause toxicity (Pires et al., 2002). Effects of the flooding stress are complex, depending on the developmental stage of the plant and the duration of stress (Schöffel et al., 2001; Pires et al., 2002). During its developmental cycle, soybeans are morphologically adapted to conditions of excess water and partially recover growth when soil drainage occurs, conferring greater tolerance to soil flooding (Pires et al., 2002). Therefore, soybean is a promising crop for the cultivation and crop rotation in flooded soils. However, the magnitude of yield reduction, due to flooding and the duration of excess water, needs to be determined for the better crop management. Soybean is a crop tolerant to excess water because of the creation of adventitious roots and aerenchyma, a mechanism for adaptation to excess water (Pires et al., 2002). However, the tolerance and ultimate yield of soybean BMX Potência RR, recommended for several Brazilian States and the Western Frontier of RS State, in conditions of flooding for an extended period, is not well understood. This study aimed to evaluate the effects of excess water on the yield and development of soybean cultivar BMX Potência RR.

Results and Discussions

Excess water and soybean growth

Soybean plant height, number of leaves, and stem diameter were not affected by flooding until 32 days, in stages R2 and R5 (Table 1). In addition, Pires et al. (2002), in flood soil conditions for 21 days at the vegetative stage (V2 to V5), found no differences in the plant height. However, these researchers observed the death of the main root, enlargement (hypertrophy) and formation of a spongy region at the stem base, emergence of adventitious roots, presence of soybean nodulation; and reduction in the levels of nitrogen (N), potassium (K^+), magnesium (Mg^{2+}), and manganese (Mn^{2+}) and an increase in levels of iron (Fe^{2+}) in leaves. According to these authors, soybean adventitious roots under flooding showed high rupture of cortex cells, creating aerenchyma voids used as pores for O_2 transfer to roots, being a plant survival mechanism under these conditions.

Morphological changes by flooding

Although no changes were observed in the plant height and stem diameter in soybean plant roots and stems after eight days of flooding, more pronounced effects were observed at 32 days (Fig 1). The formation of a large number of adventitious roots, appearance of a few nodules on the soil surface, formation of cracks in the stem just above the water surface and in submerged stems and deformation was pronounced. This may have occurred because the culture was already in the reproductive stage, when the vegetative growth is slow, and because of cultivar characteristics, no spongy tissue was formed, and there were no increases in the stem diameter, as verified by Pires et al. (2002). These authors observed the increased stem diameter when the flooding occurred in the V2 stage, and this increase differed among soybean cultivars FT-Abyara and Br 4, demonstrating an important adaptation in differentiating resistant cultivars to hypoxic conditions. The emergence of the adventitious roots (Henshaw et al., 2007b) and aerenchyma tissues (Shimamura et al., 2003; Visser and Voesenek 2004) constitutes a strategy of plant survival to hypoxia but does not allow full plant growth, resulting in the reduction in height and shoot dry matter. These morphological adaptations are important for the soybean tolerance to water stress because the effects of soil flooding during the vegetative stage are less damaging (Scholles and Vargas, 2004).

Components of the yield

Visual morphological changes in soybean roots and stems during flooding did not reflect any significant damage to yield components, which was reflected in the height of the first pod, number of pods per plant, number of grains per pod and number of grains per plant (Table 2). Schöffel et al. (2001) observed a decrease in the number of pods per plant, when the flooding occurred during 10 days from the R4 stage, attributing to excessive dropped pods. However, this did not occur when flooding occurred at the V6 vegetative stage. The thousand grain mass and yield decreased gradually following quadratic regression adjustments, with an increase in the number of days of flooding in stages R2 and R5 (Fig 2). This is possibly due to O_2 reduction in the soil by flooding that causes immediate reduction in the respiration of plant roots (Liao and Lin, 2001), decreasing nodulation and symbiotic nitrogen (N) fixation in the soybean plant, N uptake and other nutrients (Bacanamwo and Purcell, 1999; Pires et al., 2002; Scholles and Vargas, 2004) and water and assimilates translocation in the stressed plant (Taiz and Zeiger, 2013). Reduction in the soybean yield was 17% and 29% at stages R2 and R5 respectively, 16 days after the flooding, indicating reduced yield with increased crop exposure time to the flooded soil. Yield reduction was higher in the R5 stage. Rhine et al. (2010), in a field experiment with two soil types, obtained reductions from 20% to 39% in the grain yield of different soybean cultivars when subjected to eight days of flooded soil in the R5 stage. According to Rhine et al. (2010), flooding at the R5 stage caused more damage to the soybean grain yield compared with flooding at the R2 stage. Schöffel et al. (2001) found no difference in the thousand grain mass and yield of four soybean cultivars when the flooding occurred for 10 days at the R2 and R4 stages. This may be due to the short period of flooding, i.e. 10 days, since eight days of flooding in the R2 stage (Fig 2) caused a slight reduction in soybean grain yield in the pot experiment. In this context, it is important to consider that in experiments conducted in pots, there is a rapid drainage of excess water, after the set period of flooding, compared to field conditions where drainage is slower, prolonging the period of hypoxia. Therefore, a few days of field flooding are sufficient to cause more severe grain yield reduction, as observed in studies by Rhine et al. (2010). At 32 days, the decrease of soybean grain yield was 41% and 36% at stages R2 and R5, respectively, with larger reduction when prolonged excess water was applied at the R2 stage (at full flowering), when the plant has increased demand for water and nutrients to yield potential express. The thousand grain mass was the yield component that decreased with increasing time of flooding, although the number of pods per plant and number of grains per plant decrease more than 14% but did not significantly influence grain yield. The decrease in the soybean yield, with an increase in the number of days of flooded soil, may have occurred because of N deficiency, at the stage R5 (beginning seed filling). The yield reduction was small at 16 to 32 days of soil flooding, a period when culture is at the end of grain filling, and a lower demand for N exists. Several studies have shown a lower biological N_2 fixation and contribution of N for soybeans under conditions where there is an absence of O_2 (Pires et al., 2002). Bacanamwo and Purcell (1999) found that the decrease in soybean dry matter production was 34% in soil flooding condition and with biological N_2 fixation. Therefore, when nitrate was added, the reduction was only 12%, indicating lower biological N_2 fixation in flooded conditions. This indicates the key factor responsible for soybean growth reduction. They also observed that inhibition of N_2 fixation occurred earlier and more significantly in relation to dry matter production, when flooding did not occur until 21 days, during the V4 vegetative stage. Seven days of soil flooding caused no reduction in the soybean

Table 1. ANOVA on data generated using six excess water treatments, five replications and their interaction on plant height, number of leaves/plant and stem diameter, in soybean R2 and R5 stage, after the beginning of the excess water treatments at 7, 14, 21, 28 and 35 days.

Source of variation	Degree of freedom	Mean square								
		R2 stage					R5 stage			
		7	14	21	28	35	7	14	21	28
		Plant height (cm)								
Treatment	5	1.79^{ns}	7.65^{ns}	39.71^{ns}	32.06^{ns}	33.07^{ns}	60.46^{ns}	49.52^{ns}	63.76^{ns}	54.19^{ns}
Error	24	54.50	33.48	20.02	21.55	19.50	30.60	36.18	31.10	38.02
CV (%)		11.19	8.35	6.30	6.68	6.33	8.24	8.85	8.4	9.17
		Number of leaves/plant								
Treatment	5	53.39^{ns}	57.50^{ns}	116.72^{ns}	113.97^{ns}	67.34^{ns}	93.58^{ns}	90.53^{ns}	78.21^{ns}	147.17^{ns}
Error	24	50.00	54.37	54.15	48.32	41.57	49.51	47.95	215.7	41.15
CV (%)		18.59	17.85	16.96	15.75	15.24	16.10	15.50	16.96	18.94
		Stem diameter (cm)								
Treatment	5	1.39^{ns}	0.72^{ns}	1.99^{ns}	2.12^{ns}	1.59^{ns}	0.36^{ns}	0.68^{ns}	3.18^{ns}	2.60^{ns}
Error	24	1.40	0.96	1.57	0.84	1.23	0.91	1.80	1.70	0.99
CV (%)		10.45	7.64	5.77	6.96	8.97	7.61	10.44	10.07	7.81

^{ns} No significance at $p \leq 0.05$. CV= coefficient of variation.

Fig 1. Image of a soybean plant, and the soil surface with adventitious roots, stem cracks and deformation of the stem that was submerged at the depth of 2 cm in water in the R2 stage, at 32 days after soil flooding.

biomass production, but the N content decreased significantly. In addition, it was observed that the decrease in the biomass is due to N deficiency and the supply of mineral N may increase soybean tolerance to soil flooding. Scholles and Vargas (2004) observed that soil flooded at 14 days after emergence, for 30 days, decreased biological N_2 fixation, reducing the number and mass of nodules in soybean plants. However, after removal of excess water, soybean nodulation and N_2 fixation recovered. According to these authors, soybean plants fertilized with mineral N were also influenced by soil flooding but had higher shoot dry matter production compared to plants inoculated with bacteria, indicating that other factors, to a lesser extent, are involved in decreased dry matter production, beyond N deficiency. It is noted that soybeans inoculated with the bacterium *Bradyrhizobium elkanii* had the same effect on biological N_2 fixation as *Bradyrhizobium japonicum,* under soil flooding conditions (Scholles and Vargas, 2004). This is important because it demonstrates that there are no differences among gender and strains of bacteria used for inoculation. In addition, it shows that biological N_2 fixation is reduced by an insufficient O_2 supply to nodules and by hormonal changes in plants subjected to hypoxia, such as an increase in the endogenous production and concentration of ethylene (Fukao and Bailey-Serres, 2004). The O_2 deficiency in roots and nodules in soil flooding conditions is evident at Fig 1, showing that adventitious roots and some nodes are at the soil surface after 32 days in the R2 stage. This may have contributed to a reduction in N_2 fixation, N supply to the plant and yield reduction. This is in agreement with the results of Pires et al. (2002), who found a lower N content in soybean leaves at the seventh day of soil flooding. Soybean yield decrease may have occurred due to flooding having been experimentally applied in R2 and R5 stages, when carbohydrates are directed to the reproductive parts of the plant. However, with soil flooding, the soybean plant tends to displace the photoassimilates, preferably for the development of adventitious porous roots (Bacanamwo and Purcell, 1999) damaging grain filling and upon termination of water stress, are partly involved in the vegetative recovery and during the closure of plant cycle. In addition, Schöffel et al. (2001) and Rhine et al. (2010) found more pronounced effects of soil flooding on the yield when the flooding occurred in soybean reproductive subperiods. Besides the possible deficiency of N under flood soil conditions, other factors may have contributed to the soybean yield reduction. Among these are the reductions in photosynthetic rate of leaves, partially attributed to decreased stomatal conductance (Davanso et al., 2002). This consequently reduces the rate of growth and yield (Rhine et al., 2010). Also, once the condition of hypoxia has occurred, a deviation from aerobic to anaerobic pathway takes place, inducing changes in the respiratory metabolism

Table 2. ANOVA on data generated using six excess water treatments, five replications and their interaction on the height of first pod, pods/plant, grains/pod, grains/plant, thousand grains mass and grains yield/plant, in soybean R2 and R5 stage.

Source of variation	Degree of freedom	Mean square							
		Soybean stage							
		R2	R5	R2	R5	R2	R5	R2	R5
		Height first pod		Pods/plant		Grains/pod		Grains/plant	
Treatment	5	14.15^{ns}	4.59^{ns}	65.92^{ns}	31.47^{ns}	0.04^{ns}	0.04^{ns}	296.93^{ns}	218.03^{ns}
Error	24	6.50	4.68	29.57	30.83	0.04	0.02	145.95	182.13
CV (%)		23.96	22.15	16.28	15.10	7.27	5.98	13.97	14.14
				Thousand grains mass		Grains yield/plant			
Treatment	5			1757.26^{**}	1816.19^{**}	42.70^{**}	31.52^{**}		
Error	24			152.75	121.47	3.98	3.22		
CV (%)				6.92	6.69	12.69	11.23		

^{ns} No significance at $p \leq 0.05$. ** Significance at $p \leq 0.01$. CV= coefficient of variation.

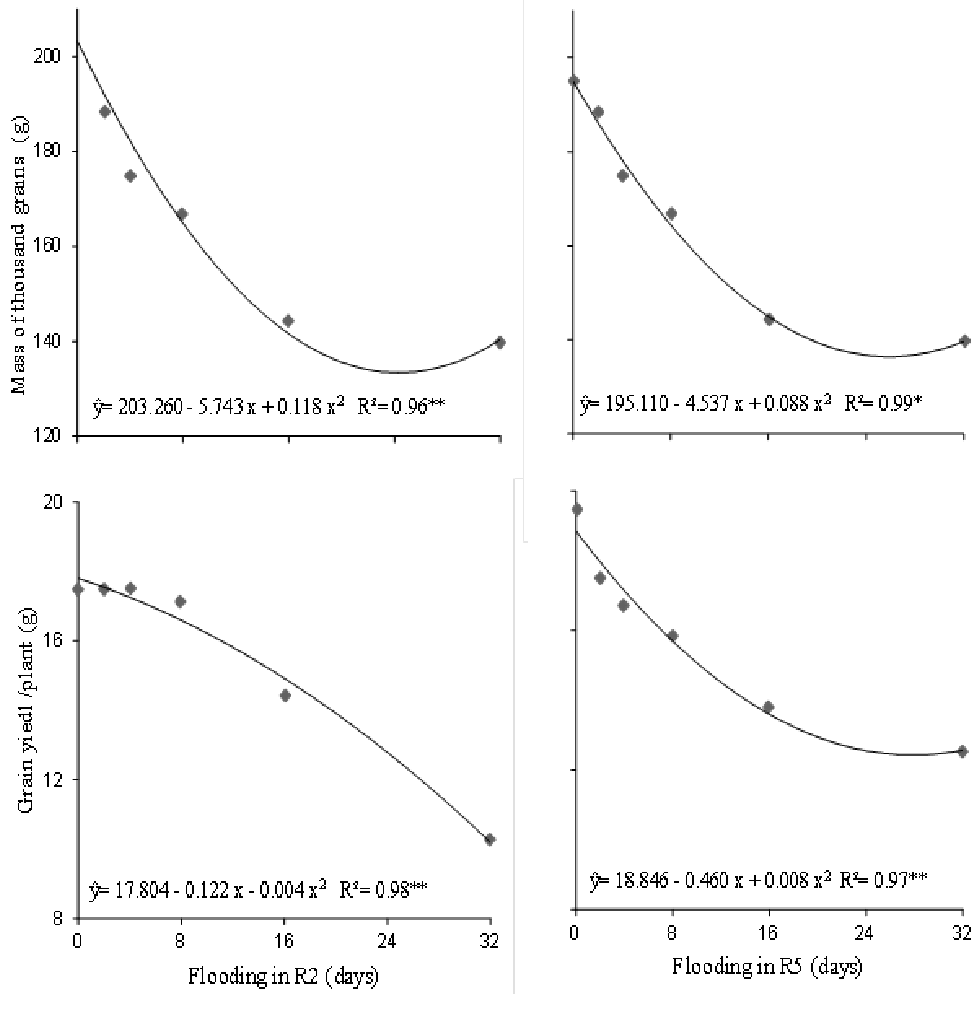

Fig 2. Soybean mass of thousand grain and yield per plant/pot in soil flooding periods of 0, 2, 4, 8, 16 and 32 days with 2 cm water above the soil surface, in the stages of full flowering (R2) and beginning grain filling (R5), at 55 and 76 days after emergence, respectively.

of the root system that, in turn, produces toxic substances such as ethanol and lactate, beyond low energy efficiency (Kolb and Loly, 2009), and an abrupt increase in the activity of fermentative enzymes (Borella et al., 2013).

Materials and Methods

Location and soil characterization

The experiment was carried out at geographic coordinates 29º12'28''S and 56º18'28''W, 74 m altitude, 'Cfa' climate, subtropical with no dry season and a hot summer, according to the classification of Köppen-Geiger. Ultisol containing 190 g kg^{-1} of clay, collected at a layer of 0 - 20 cm depth, and passed through a sieve of 0.4 cm was used. Ultisol's chemical characteristics were: pH H_2O = 5.1, P = 12.6 mg dm^{-3}, K^+ = 0.153, Ca^{2+} = 2.7, Mg^{2+}=0.7, Al^{3+} = 0.6 cmol$_c$ dm^{-3}, V = 50% and OM = 1.6%. The pH of the soil was increased in through a liming process at 180 days before sowing (Tedesco et al., 2004).

Experimental design and treatments

The experimental design used was completely randomized with six treatments and five replicates of pots of 7.5 L (6 dm³ soil per pot), by performing two experiments: excess water in the flowering stage (R2); and in the beginning of the grain-filling stage (R5). The six treatments consisted of excess water of 0, 2, 4, 8, 16 and 32 days of flooding, in stage R2 (R2.0, R2.2, R2.4, R2.8, R2.16 and R2.32, respectively), and R5 (R5.0, R5.2, R5.4, R5.8, R5.16 and R5.32, respectively) at 55 and 76 days after emergence, respectively. The excess water was created by adding an external pot, without holes, and the application of water was performed 2 cm above the soil surface. After the period of application of excess water, the external pots were removed to drain the excess water through holes in the bottom of the pot being used for cultivation.

Soybean cultivation

Soybean sowing with cultivar BMX Potência RR was performed on 10/01/2013 with eight seeds per pot at 4 cm depth. The fertilizations with macronutrients were: phosphorus = 165 mg dm^{-3} and potassium = 188 mg dm^{-3}, in the form of triple superphosphate and potassium chloride, respectively. These fertilizers were triturated in a Willey mill and homogenized all soil that was placed in the pot. Soybean seeds were treated with insecticides and fungicides: Fipronil (Standak® 100 mL 100 kg^{-1} of seed) and Carboxina + Tiran (Vitavax Thiram 200 SC® 300 mL 100 kg^{-1} of seed). In addition, seeds were inoculated with *Bradyrhizobium elkanii* with 120 g 100 kg^{-1} of seed, in a concentration of 5 x 10^9 colony-forming units per gram (g). At 14 days after sowing, thinning was performed, leaving one plant per pot. These plants were watered daily and rotation of pots was performed weekly.

Evaluations performed

Plants were evaluated after the beginning of the excess water treatments. These evaluations consisted of the measurement of the plant height, number of leaves per plant and stem diameter at 2 cm above the soil surface every seven days, totaling five and four evaluations in experiments R2 and R5, respectively.

At the harvest, plants were evaluated for the height of the first pod, number of pods/plant, number of grains/pod, grains/plant, thousand grains mass, and grains yield/plant.

Statistical analysis

Statistical analyses were performed by analysis of variance (ANOVA) and randomized completely design with six treatments and five replicates and were applied in each of two experiments to yield components: excess water in the flowering stage (R2) and in the beginning of the grain-filling stage (R5). The ANOVA were made at each assessment date in the two experiments for the plant height, number of leaves per plant and stem diameter. The probability for ANOVA was set at 0.05 level and when F was significant, treatment mean was submitted to polynomial regression analysis with significance level at $p \leq 0.05$.

Conclusion

Soybean plant height, leaf number, stem diameter, height of the first pod, number of pods per plant, number of grains per pod and number of seeds per plant were not affected by soil flooding until 32 days in stage R2 (full flowering) and R5 (beginning of grain filling). The soybean thousand grain mass and grain yield reduced gradually with an increase in the number of days of soil flooding, in stages R2 and R5, reducing grain yield 17% and 29%, after 16 days and 41% and 36%, after 32 days of excess water, respectively.

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