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Genotype-dependent and flooding-induced root growth and flooding tolerance in the early growth stage of sugarcane

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Abstract Flooding conditions reduced the root dry weight, the root/shoot ratio, the root length, and the total dry weight. Genotype variation was found in all the root growth traits and the adventitious roots under flooding conditions, and genotype differences were observed for flooding tolerance. A positive and significant correlation between root dry weight and total dry weight (r = 0.76**) was found. The tolerance index (TI) of the root dry weight also correlated positively with the TI of the dry weight for different parts of the sugarcane (r=0.84** to 0.93**). Additionally, the correlation between the number of aboveground roots and the TI of the total dry weight was a significantly positive result(r = 0.68*). These findings suggested that root growth and the ability to form adventitious roots in flood-prone sugarcane genotypes are crucial factors for promoting flooding tolerance.

Keywords: Genotype-dependent, Root traits, Adventitious root, Flooding stress, Tolerance index

Introduction

Flooding is a natural problem that affects crop production in agricultural areas around the world, and it is a major issue for sugarcane cultivation in the tropics and subtropics, which are frequently affected by climate change and rainfall variability (Pipitpukdee *et al.*, 2020). Sugarcane is Thailand's most important economic crop, serving as the primary raw material for the sugar industry. In addition to being used for domestic consumption, it is a significant export product for the country (USDA Foreign Agriculture Service, 2020). Thailand's sugarcane planting area is likely to expand as a result of the Thai government's agricultural land management policy for zoning and planting

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crops appropriate for the area. This encourages farmers in areas where rice cultivation is not feasible to switch to other crops, particularly sugarcane, to replace rice. Sugarcane has been planted in greater numbers in lowland areas that were previously rice fields. As a result, as the rainy season approaches, sugarcane cultivated in lowland areas faces an increased risk of flooding stress, which affects sugarcane growth and yield (Jaiphong *et al.*, 2016; Jaiphong *et al.*, 2017).

Flooding conditions impact plant growth and yield, and sugarcane growth and yield as well as the sugar yield in both plant cane and ratoon cane might be reduced (Gomathi *et al.*, 2015). The extent of sugarcane damage caused by flooding conditions is determined by the environment, the sugarcane growth stage, the water level height, and the time of the flooding, as well as the tolerance level of sugarcane genotypes to flooding (Jain *et al.*, 2017; Fazle *et al.*, 2015; Glaz and Lingle, 2012). Sugarcane in the early growth stage (3–4 months), is the most susceptible to flooding compared to the other growth stages. Flooding stress during the early growth stage could impact the quantity and quality of the sugarcane yield at the end of the growing season (Sanghera and Jamwal, 2019). Sugarcane cultivation in lowland and irrigated areas in Thailand usually takes place in the pre-rainy season until the beginning of the rainy season.Sugarcane planting startsbetween February and April, and the amount of rain increases during the rainy season from mid-May to mid-October. Sugarcane cultivation in lowland areas has a high risk of flooding in the early stages of growth (Jaiphong *et al.*, 2016; Jaiphong *et al.*, 2017). Flooding occurs when water replaces gaps in the soil, resulting in a lack of oxygen. As a result, sugarcane roots are depleted of oxygen, resulting in decreased root respiration. Due to insufficient energy, the roots' ability to absorb water and nutrients suffers until the roots are hampered in their growth or die (Gomathi *et al.*, 2015). Consequently, various physiological anomalies influence sugarcane growth and productivity.

Adaptation of root traits might result in sugarcane being more tolerant to flooding conditions. A difference in root weight and root volume was discovered when sugarcane genotypes were exposed to flooding conditions (Soleh *et al.*, 2018). Flooded sugarcane genotypes are more resistant to floods and have greater root density (Avivi *et al.*, 2016). Flooding enhanced the root number and volume in some sugarcane varieties (Anitha *et al.*, 2016). Moreover, under flood conditions, some sugarcane genotypes form adventitious roots around the flooded node to increase the functional capacity and oxygen determination of the roots (Puspitasari *et al.*, 2017; Jaiphong *et al.*, 2016). Therefore, root traits might be related to the flooding tolerance of sugarcane.

Flooding-tolerant sugarcane varieties could be cultivated to effectively mitigate the effects of flooding on sugarcane production. Furthermore, root growth traits might be used in the selection of sugarcane genotypes for flood tolerance. Thus, the objectives were to investigate the effect of flooding conditions on sugarcane root growth, assess variations in root traits and tolerance for flooding conditions among sugarcane genotypes, and to investigate a relationship between root traits and sugarcane tolerance to flooding.

Materials and methods

Experimental design and treatments

The experiment was conducted at the research fields of the Department of Agronomy, Kasetsart University, KamphaengSaen Campus, NakhonPathom, Thailand from May to December 2018. The experiment usedtwo (2 × 5) factors factorial in completelyrandomizeddesign (CRD) with three replications.The first factor involved two different watering conditions, and the second involved five sugarcane genotypes.A comparison between the control and flooding conditions, where sugarcane was flooded in the early stages of growth, tested five sugarcane improved sugarcane genotypes in Thailand: KK07-037, KK07-250, KK07-599, K95-84, and LK 92-1.One-month-old healthy sugarcane seedlings were prepared for testing. The seedlingswere transplanted into pots with a diameter of 30 cm and a height of 27 cm, using soil from the sugarcane plantation as the planting material. The potting soil was placed to almost fill the pot, with the remaining space from the edge of the pot being approximately 2 cm; each pot contained 8 kg of soil. A fertilizer formula of 15-15-15 at a rate of 11 g/plant was applied after the seedlings had been transplanted for one month. To continue simulating flooding conditions, we selected plants that were complete and similar in size in each genotype two months after transplantation.

The sugarcane plant pots were immersed in a cement pond with a diameter of 80 cm and a height of 90 cm, in which the water in the cement pond floodedto a height of 60 cm, and the sugarcane plant was flooded to about 30 cm above the soil surface. A steady flood level was maintained throughout the simulation by supplying water to compensate for evaporation, and the sugarcane was in a state of flooding for 30 days. Then, the sugarcane potswere removed from the cement pond and placed in control conditions for another 30 days to allow the sugarcane to recover, simulating the non-flooded state for comparison. In the control conditions, the sugarcane was watered by filling the pot with water until the water began to seep out from the bottom. Water was given every three days throughout the experiment.

Data collection

The root growth traits and dry weight of sugarcane genotypes were collected both after the flooding and after the recovery from the flooding. Three sugarcane plants were randomly assigned to each treatment. Each sugarcane plant was washed in water, and the soil was separated from the root. We recorded the root-length data by measuring the length of the root from the first node of the stalk to the longest part of the root. Then, we separated the sugarcane leaves, stalks, and roots. The sugarcane leaves, stalks, and roots were freshly weighed and placed in an 80 ℃ hot air oven. The leaves and roots were dried for 72 hours, while the stalks were dried for 120 hours. Once the drying was complete, we weighed and recorded the root dry weight, the shoot dry weight (the overall dry weight of the leaves and stalks), and the total dry weight of the whole cane plant. The root/shoot ratio was calculated by dividing the root dry weight by the shoot dry weight. Each plant's fresh weight and dry weight data were used to calculatethe moisture percentage using the following formula: (fresh weight – dry weight) / fresh weight) $\times$ 100. In the flooded sugarcane, the adventitious roots were generated at the root primordia at nodes under the water and from aerial nodes, which were aboveground. We separated the aboveground and underground root parts and recorded their dry weights. The ratio of the aboveground root dry weight to the underground root dry weight (aboveground/underground root ratio) was then computed.

Statistical analysis

Analysis of the variance of the root traits and the sugarcane dry weight after the flooding and after the recovery from flooding wasperformed. The factorial in CRD experimental design was used to assess the impact of the flooding conditions and genotype variability in sugarcane. The variances of each trait were then analyzed separately for the control and flooding conditions using a CRD experimental design. Mean comparisons were performed using Duncan's multiple range test (DMRT).

For each genotype, the flooding tolerance index (TI) was calculated as follows:TI = (measured plant parameter under flooded conditions/measured plant parameter under control conditions) $\times$ 100 (Jain *et al.*, 2017).

Simple correlations were computed between the sugarcane root traits and flooding tolerance.

Results

Effects of the flooding conditions on the root growth and dry weight of the sugarcane

After 30 days of flooding, the early flooding conditions had a statistically significant effect on the root dry weight, root/shoot ratio, and root length (Table 1). The root dry weight was reduced by 53.74%, the root/shoot ratio was reduced by 58.33%, and the root length was reduced by 14.18%. However, no effect of the flooding conditions on the shoot dry weight or the total dry weight was observed. Furthermore, except for the root dry weight, the differences between the genotypes were found to be statistically significant in all the traits studied. There was also an interaction between the water regimes and the genotypes in all the traits studied, except the root length.

After the sugarcane recovered from the 30 days of flooding, the flooding conditions had a statistically significant effect on the root/shoot ratio and root length (Table 1), with the effects on the total dry weight also included. Because of the flooding, the root dry weight reduced by 53.64%, the total dry weight reduced by 20.32%, the root/shoot ratio reduced by 57.30%, and the root length reduced by 11.87%. However, no effect of the flooding on the shoot dry weight was found. Moreover, except for the root dry weight, the difference between the genotypes was found to be statistically significant in all the traitsstudied. Furthermore, an interaction between the water regime and the genotypes was found in the root dry weight and root length. However, there was no interaction between the water regime and the genotypes in the root dry weight, total dry weight, and root/shoot ratio.

A comparison of the moisture content of the sugarcane between the control and flooding conditions found that at 30 days after the flooding, the flooded sugarcane had a root moisture content of 36.47% (Figure 1A), lower than in the control conditions with a root moisture content of 45.40%. However, the flood sugarcane had shoot and total moisture contents of 73.87% and 71.25%, respectively, which was higher than in the control conditions, which had shoot and total moisture contents of 71.35% and 66.41%, respectively. After 30 days, the sugarcane had a root moisture content of 55.58% (Figure 1B), higher than in the control conditions thathad a root moisture content of 45.76%, including the flooded sugarcane, which had shoot and total moisture contents of 74.26% and 70.95%, respectively, higher than in the control conditions, in which the sugarcane had shoot and total moisture contents of 69.78% and 62.17%, respectively. As a result of the flooding conditions, the root moisture of the sugarcane decreased, while the shoot moisture increased. Furthermore, after the sugarcane had recovered from the flooding, the previously flooded sugarcane had higher moisture than in the control conditions, both in the root and in the shoot.

Genotype variation in the root traits of the sugarcane

Analysis of the variance of root traits was performed separately for the control and flooding conditions. After 30 days of flooding, there was no difference between the genotypes in the root dry weight under control conditions, with values ranging from 14.21 g/plant to 25.14 g/plant (Table 2), but there was a statistically significant difference between the genotypes in the root dry weight under flooding conditions, with K 95-84 and KK 07-037 having the highest root dry weight, 13.59 g/plant and 12.09 g/plant, respectively. The TI of the root dry weight ranged from 20.04% to 95.62%. K 95-84 had the highest root dry weight tolerance. Under the control conditions, no differences in the root lengths, which ranged from 65.67 cm to 90.33 cm, were observed between the genotypes. However, under the flooding conditions, a statistically significant difference in root length was observed between the genotypes. The longest root length was 89.00 cm in LK 92-11. The TI of the root length ranged from 74.07% to 104.71%. The genotype with the highest root dry weight tolerance was LK 92-11. Statisticallysignificant differences between the genotypes were found in the root/shoot ratio in boththe control and the flooding conditions. In the control conditions, K95-84 had the highest root/shoot ratio, with a value of 0.71, and in the flooding conditions, KK07-250 had the highest root/shoot ratio, with a value of 0.29 and a TI of the root/shoot ratio ranging from 27.81% to 68.51%. KK 07-250 was the genotype with the highest TI of the root/shoot ratio.

After the sugarcane recovered from the flooding for 30 days, there was no difference in the root dry weight between the genotypesbetween 38.10 g/plant and 52.43 g/plant under the control conditions (Table 3). There was a statistically significant difference in the root dry weight between the genotypes under the flooding conditions. With a root dry weight of 27.93 g/plant, LK 92-11 had the highest root dry weight. The TI of the root dry weight ranged from 35.94% to 56.62%. LK 92-11 was the genotype with the best root dry weight tolerance. In both the control and the flooding conditions, the difference in root length across the genotypes was statistically significant. LK 92-11 had the longest root length in the control conditions, at 104.00 cm, whereas the genotypes LK92-11 and KK07-037 had the longest root length in the flooding conditions, at 81.33 and 75.69 cm, respectively. The TI of the root length ranged from 74.70% to 120.74%, with KK07-037 having the highest TI in root length. Under the control conditions, no differences inthe root/shoot ratio were found between the genotypes, with the values ranging from 0.67 to 1.49. However, there was a statistically significant difference in the root/shoot ratio under the flooding conditions, with K 95-84 having the highest root/shoot ratio of 0.63 and a TI of root/shoot ratio ranging from 35.16% to 52.73%, and LK92-11 having the highest TI in root dry weight.

All the sugarcane genotypes studied had adventitious root formation at the node of the flooded portion, which was the aboveground root, in addition to the underground root, under the flooding conditions. As a result, only the flooded sugarcane data were evaluated for the separation of the aboveground and underground roots. After the 30 days of flooding, the differences in the number of aboveground roots, the aboveground root dry weight, the underground root dry weight, and the aboveground/underground root ratio between the genotypes were statistically significant (Table 4). K 95-84 and KK 07-037 had the most aboveground roots, with 96.50 and 88.00 roots/plant, respectively. KK 07-037 had the highest aboveground root dry weight of 4.88 g/plant, K 95-84 had the highest underground root dry weight of 10.18 g/plant, and KK 07-037 had the highest aboveground/underground root ratio of 0.69. After 30 days, the aboveground root dry weight, the underground root dry weight, and the aboveground/underground root ratio were found to show statistically significant differences between the genotypes. KK07-599 and KK07-037 had the highest aboveground root dry weights of 4.06 g/plant and 3.83 g/plant, respectively, LK92-11 had the highest underground root dry weight of 26.42 g/plant, and KK07-037 and KK07-599 had the highest aboveground/underground root ratios of 0.27 and 0.26, respectively. However, there were no differences in the number of aboveground roots between the genotypes, which rangedfrom 53.00 roots/plant to 84.67 roots/plant.

Tolerance to flooding conditions in the sugarcane genotypes

After flooding the sugarcane for 30 days, the difference in the total dry weight between the genotypes was statistically significant in both the control and the flooding conditions (Table 5). Under the control conditions, the genotypes with the highest total dry weight were KK07-599, LK92-11, KK07-037, and KK07-250, with total dry weights of 86.29g/plant, 81.87g/plant, 73.41g/plant, and 69.59 g/plant, respectively. Under the flooding conditions, the genotypes with the highest total dry weight were KK07-037, KK07-599, and K95-84, with total dry weights of 77.53 g/plant, 71.65g/plant, and 65.66 g/plant, respectively. The TI to the flooding conditions was observed to be between 57.35% and 191.87%. K 95-84 was the genotype with the highest flood TI. Furthermore, after the sugarcane had recovered from being flooded for 30 days, under the control conditions, the total dry weight did not differ between the genotypes, ranging between 78.76 g/plant and 118.13 g/plant. However, when the sugarcane was flooded, there was a statistically significant difference in the total dry weight between the genotypes. The genotypes with the highest total dry weights were KK 07-599 and LK 92-11, with total dry weights of 101.69 g/plant and 99.78 g/plant, respectively. When the TI to flooding conditions was considered, with values ranging from 66.90% to 103.29%, the tolerance genotype to the highest flooding condition was KK07-599.

Relationship between the root traits and the flood tolerance of the sugarcane

The pooled correlations were analyzed after the flooding and after the recovery from flooding in boththe control and the flooding conditions (n = 20). A strong positive correlation was found between the root dry weight and the total dry weight (r = 0.76**) (Figure 2). Furthermore, when the correlation was examined separately for each water regime (n=10), a strong positive correlation between the root dry weight and the total dry weight in both the control (r = 0.81**) and the flooding conditions (r = 0.70*) was observed (data not shown). Moreover, the TI of the root dry weight had a significant positive correlation with the TI of the leaf dry weight, the stalk dry weight, the shoot dry weight, and the total dry weight (r = 0.84** to 0.93**) (Table 6). This experiment also found a relationship between the adventitious roots of the sugarcane that were created in the flooding conditions and the sugarcane's flooding tolerance. A positive correlation was observed between the number of aboveground roots and the TI of the total dry weight (r = 0.68*) (Figure 3). In addition, for each study date (n=5), the correlation between the total dry weight and the adventitious root traits of sugarcane was examined separately. A positive correlation between the total dry weight and the aboveground root dry weight and the aboveground/underground root ratios after the 30 days of flooding (r=0.88* and 0.90*, respectively) was found (data not shown), but there was no relationship between the total dry weight and the adventitious root traits after the 30 days of recovery from flooding.

Table 1. Root dry weight, shoot dry weight, total dry weight, root/shoot ratio, and root length of sugarcane genotypes under different water regimes at 30 days after flooding and 30 days after recovering

Treatment	30 days after flooding										30 days after recovering									
	Root dry weight (g/plant)		Shoot dry weight (g/plant)		Total dry weight (g/plant)		Root/shoot ratio		Root length (cm)		Root dry weight (g/plant)		Shoot dry weight (g/plant)		Total dry weight (g/plant)		Root/shoot ratio		Root length (cm)	
Water regime																				
Control	21.01	A	48.06		69.07		0.48	A	80.20	A	46.38	A	57.85		104.22	A	0.89	A	79.43	A
Flooding	9.72	B	51.01		60.73		0.20	B	68.83	B	21.50	B	61.54		83.04	B	0.38	B	70.00	B
F-test	**		ns		ns		**		*		**		ns		**		**		*	
Genotype																				
KK 07-037	18.45		57.02	ab	75.47	ab	0.34	bc	78.33	ab	35.64		62.74	a	98.37	a	0.57	b	69.17	b
KK 07-250	14.76		40.97	c	55.73	c	0.36	b	60.00	c	32.31		62.09	a	94.41	a	0.52	b	64.83	b
KK 07-599	14.64		64.32	a	78.96	a	0.23	d	83.50	a	28.82		71.25	a	100.07	a	0.46	b	73.67	b
K 95-84	13.90		36.05	c	49.94	c	0.49	a	63.75	bc	34.29		32.06	b	66.36	b	1.06	a	73.25	b
LK 92-11	15.09		49.32	bc	64.41	bc	0.28	cd	87.00	a	38.63		70.33	a	108.96	a	0.56	b	92.67	a
F-test	ns		**		**		**		**		ns		**		**		**		**	
Water regime × Genotype																				
F-test	**		**		**		**		ns		ns		**		ns		ns		*	
CV (%)	25.05		17.60		17.76		16.14		17.20		28.73		17.83		16.60		38.18		12.28	
Mean	15.37		49.54		64.90		0.34		74.52		33.94		59.69		93.63		0.63		74.72	

Mean in the same column with the same letters are not significantly different byDuncan's multiple range test (DMRT).
ns, not significant at P<0.05; *, significant for P<0.05; **, significant for P<0.01.

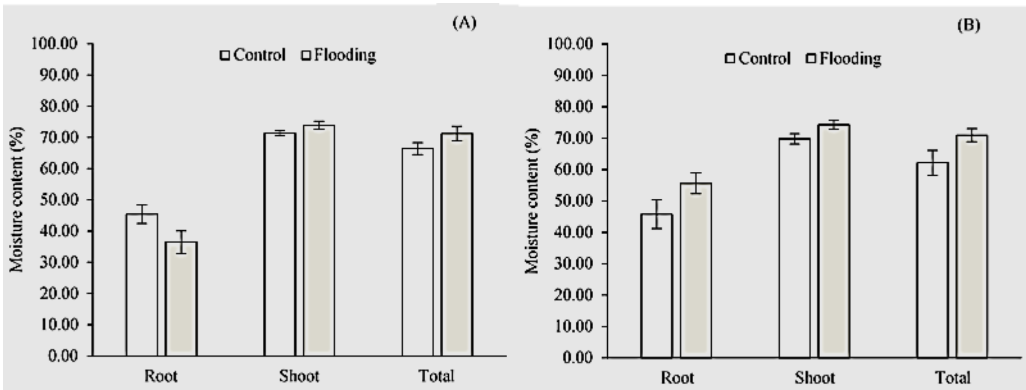

Figure 1. Comparison in moisture content of root, shoot, and total plant of sugarcane between the control condition and flooding condition at 30 days after flooding (A) and 30 days after recovering (B), Error bars represent ± SD

Table 2. Root dry weight, root/shoot ratio and root length of sugarcane genotypes under different water regimes and tolerant index (TI) at 30 days after flooding

Genotypes	Root dry weight (g/plant)				Root length (cm)				Root/shoot ratio				
	Control	Flooding		TI	Control	Flooding		TI	Control		Flooding		TI
KK 07-037	24.80	12.09	a	48.77	90.00	66.67	bc	74.07	0.50	b	0.18	bc	36.42
KK 07-250	20.60	8.92	b	43.31	65.67	54.33	c	82.74	0.42	b	0.29	a	68.51
KK 07-599	20.32	8.96	b	44.11	90.33	76.67	ab	84.87	0.31	c	0.15	c	46.82
K 95-84	14.21	13.59	a	95.62	70.00	57.50	bc	82.14	0.71	a	0.26	ab	36.45
LK 92-11	25.14	5.04	c	20.04	85.00	89.00	a	104.71	0.44	b	0.12	c	27.81
F-test	ns	**			ns	*			**		**		
CV (%)	25.07	14.15			18.20	15.60			10.79		29.14		
Mean	21.01	9.72			80.20	68.83			0.48		0.20		

Mean in the same column with the same letters are not significantly different by Duncan's multiple range test (DMRT).
ns, not significant at P<0.05; *, significant for P<0.05; **, significant for P<0.01.

Table 3. Root dry weight, root/shoot ratio and root length of sugarcane genotypes under different water regimes and tolerant index (TI) at 30 days after recovering

Genotypes	Root dry weight (g/plant)				Root length (cm)					Root/shoot ratio			
	Control	Flooding		TI	Control		Flooding		TI	Control	Flooding		TI
KK 07-037	52.43	18.84	b	35.94	62.67	b	75.67	a	120.74	0.83	0.32	bc	38.31
KK 07-250	44.09	20.53	b	46.57	64.67	b	65.00	b	100.51	0.71	0.35	b	49.06
KK 07-599	38.10	19.54	b	51.29	84.33	ab	63.00	b	74.70	0.67	0.24	c	35.16
K 95-84	47.93	20.66	b	43.10	81.50	ab	65.00	b	79.75	1.49	0.63	a	42.06
LK 92-11	49.33	27.93	a	56.62	104.00	a	81.33	a	78.20	0.73	0.39	b	52.73
F-test	ns	*			*		**			ns	**		
CV (%)	28.84	15.58			15.86		4.46			38.10	14.77		
Mean	46.38	21.50			79.43		70.00			0.89	0.38		

Mean in the same column with the same letters are not significantly different by Duncan's multiple range test (DMRT).
ns, not significant at P<0.05; *, significant for P<0.05; **, significant for P<0.01.

Table 4. Number of the aboveground root, aboveground root der weight, underground root dry weight, and aboveground/underground root ratio of sugarcane genotypes under flooding conditions at 30 days after flooding and 30 days after recovering

Genotype	**30 days after flooding**								**30 days after recovering**						
	Number of aboveground root		Aboveground root dry weight (g/plant)		Underground root dry weight (g/plant)		Aboveground /underground root ratio		Number of aboveground root	Aboveground root dry weight (g/plant)		Underground root dry weight (g/plant)		Aboveground /underground root ratio	
KK 07-037	88.00	a	4.88	a	7.21	bc	0.69	a	67.67	3.83	a	15.02	b	0.27	a
KK 07-250	47.67	b	1.00	d	7.93	ab	0.14	c	84.67	2.45	b	18.09	b	0.14	b
KK 07-599	70.67	ab	2.31	c	6.65	bc	0.37	b	77.67	4.06	a	15.49	b	0.26	a
K 95-84	96.50	a	3.40	b	10.18	a	0.33	b	53.00	1.76	bc	18.90	b	0.09	b
LK 92-11	13.67	c	0.06	e	4.98	c	0.01	c	60.33	1.51	c	26.42	a	0.06	b
F-test	**		**		**		**		ns	**		*		**	
CV (%)	23.96		19.51		18.17		30.60		29.71	16.32		17.68		33.59	
Mean	63.30		2.33		7.39		0.31		68.67	2.72		18.78		0.16	

Mean in the same column with the same letters are not significantly different byDuncan's multiple range test (DMRT).

ns, not significant at P<0.05; *, significant for P<0.05; **, significant for P<0.01.

Table 5. The total dry weight of sugarcane genotypes under different water regimes and tolerant index (TI) at 30 days after flooding and 30 days after recovering

Genotypes	Total dry weight (g/plant)									
	30 days after flooding					30 days after recovering				
	Control		Flooding		TI	Control	Flooding		TI	
KK 07-037	73.41	a	77.53	a	105.61	117.88	78.87	b	66.90	
KK 07-250	69.59	a	41.87	b	60.17	107.90	80.91	b	74.99	
KK 07-599	86.27	a	71.65	a	83.05	98.45	101.69	a	103.29	
K 95-84	34.22	b	65.66	a	191.87	78.76	53.96	c	68.51	
LK 92-11	81.87	a	46.95	b	57.35	118.13	99.78	a	84.47	
F-test	**		**			ns	**			
CV (%)	20.29		13.71			19.85	8.96			
Mean	69.07		60.73			104.22	83.04			

Mean in the same column with the same letters are not significantly different by Duncan's multiple range test (DMRT).not significant at P<0.05; **, significant for P<0.01.

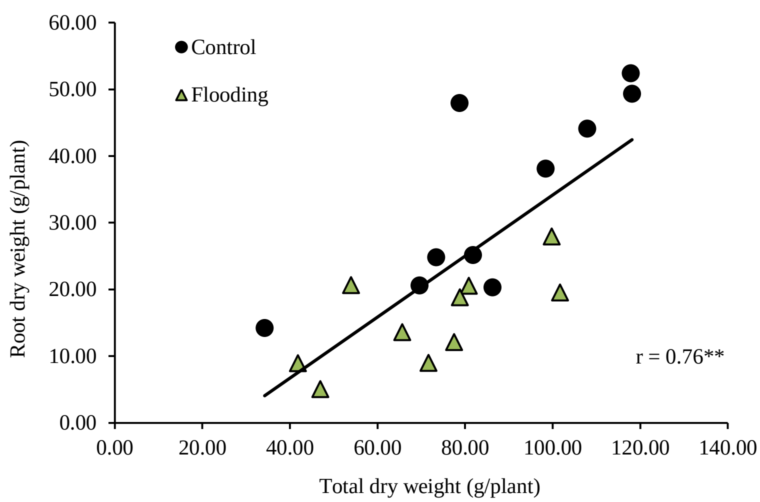

Figure 2. Relationships between root dry weight and total dry weight of sugarcane genotypes across two sampling dates in both control and flooding conditions (n = 20). ** Significant at 0.01 probability level

Table 6. Correlation coefficients (r) between the tolerant index of root dry weight with the tolerant index of leaf dry weight, stalk dry weight, shoot dry weight and total dry weight across two sampling dates (n = 10)

Tolerant index	Tolerant index of root dry weight
Leaf dry weight	0.84 **
Stalk dry weight	0.91 **
Shoot dry weight	0.91 **
Total dry weight	0.93 **

** Significant at the 0.01 probability levels.

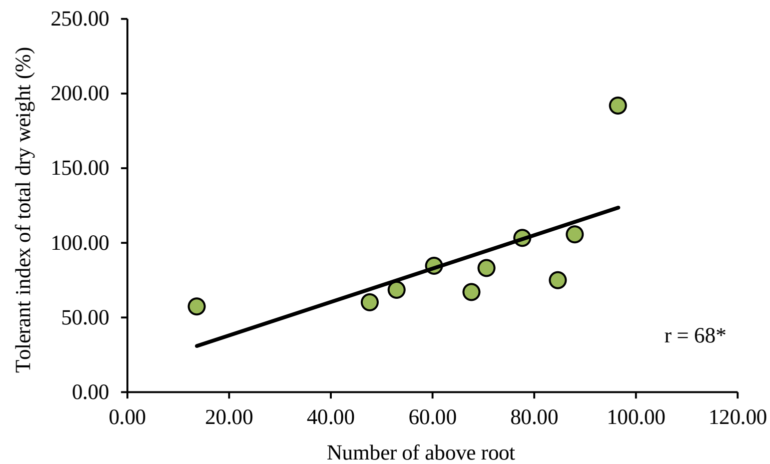

Figure 3. Relationships between the tolerant index of total dry weight and the number of above root of sugarcane genotypes across two sampling dates under flooding conditions (n = 10). * Significant at 0.05 probability level

Discussion

Flooding impacts the growth of sugarcane roots and different effects of floodingconditions on sugarcane root growth have been reported. According to Jain *et al*. (2017) and Bajpai and Chandra (2015), flooding conditions increased the root weight in sugarcane compared to sugarcane grown under normal conditions. Conversely, Misra *et al.* (2020) reported that, while flooding conditions reduced the underground root weight, the sugarcane aboveground roots compensated for the total root dry weight, which was no different from the sugarcane root dry weight under normal conditions. However, in the current study, although all the genotypes of sugarcane used in the study were able to form adventitious roots above the soil, the flooding conditions also resulted in a decrease in the total root dry weight of the sugarcane. The flooding also reduced the root/shoot ratio and the root length. This is consistent with the findings of Jaiphong *et al*. (2016, 2017), who revealed that flooding conditions caused a decrease in sugarcane root dry weight. Flooding depletes oxygen levels in the soil. Sugarcane roots are thus deprived of oxygen, resulting in decreased root respiration. Consequently, root function and growth are impaired, and root death occurs (Gomathi *et al.*, 2015). In this study, even after the sugarcane was flooded, the effects of the flooding on the root growth persisted. Although the flooded sugarcane recovered in the control conditions for 30 days, the root growth of the sugarcane was still unable to return to normal. In addition, flooding conditions affected the root growth and caused the dry weight of the sugarcane to decrease. In the present study, although there was no effect of the flooding on the dry weight of the sugarcane after 30 days of flooding, the effect of the flooding caused the total dry weight of the sugarcane to decrease after the sugarcane had passed the flooding period of 30 days.

The level of flood tolerance of sugarcane genotypes determines the extent of damage caused by flooding conditions (Jain *et al.*, 2017; Glaz and Lingle, 2012). The response of sugarcane genotypes to root growth traits influences their ability to withstand flooding conditions (Soleh *et al.*, 2018; Avivi *et al.*, 2016). Variations in root growth traits, including the root dry weight, root length, and root/shoot ratio, were discovered in this study between the sugarcane genotypes under the flooded conditions, both after the flood and the flood recovery time. Furthermore, the TI of the root growth traits revealed variation in flooding tolerance between the sugarcane genotypes. The root systems in the genotypes with a high TI were more resistant to flooding stress than the genotypes with a low TI. The TI was also calculated to assess the flooding tolerance of the sugarcane genotypes for other growth characteristics, such as cane weight, dry weight, leaf pigment content, leaf greenness, specific leaf weight, phosphorus (P), and potassium (K) content in the leaves (Singh *et al.*, 2019; Jain *et al.*, 2017). During a flood, some sugarcane genotypes adapt by growing adventitious roots above the soil surface in the flooded nodes to improve root vitality and oxygenation (Puspitasari *et al.*, 2017; Jaiphong *et al.*, 2016). As a result, adventitious root formation has evolved to improve sugarcane's ability to tolerate flooding (Sanghera and Jamwal, 2019). Therefore, the adventitious root formation of sugarcane under flooded conditions might be used in the selection of sugarcane genotypes for tolerance to flooding conditions (Gilbert *et al.*, 2008). Our study revealed that in flooded conditions, all the sugarcane genotypes studied were able to produce adventitious roots above the soil surface. The traits of the adventitious roots differed between the genotypes in terms of the number of aboveground roots, the aboveground root dry weight, and the aboveground/underground root ratio. These traits might be related to sugarcane's ability to tolerate flooding. In a previous study, sugarcane breeding for flooding stress tolerance could be selected based on the sugarcane biomass or yield under flooded conditions (Sanghera and Jamwal, 2019; Krishna *et al.*, 2018). In addition, the flooding TI was used as an indicator of the genotypes' ability to maintain growth without deterioration when affected by flooding. When the TI was low, there was a significant reduction in growth compared to normal growth. However, if the TI was high, there was little or no decrease in growth compared to normal growth (Singh *et al.*, 2019; Jain *et al.*, 2017). Thus, the total dry weight under the flooding conditions and the TI of the total dry weight were used to classify the sugarcane genotypes that are tolerant to flooding. In this study, variance was found between the genotypes and the total dry weight under the flooding conditions and the TI of the total dry weight, indicating that the sugarcane genotypes had different abilities to tolerate flooding stress. The highest flooding tolerance capability was demonstrated by KK 07-599, which had the highest total dry weight under the flooding conditions, both after the flooding and after the recovery. It also had a high flooding TI both during and after the flooding.

Plant roots play an important role in the survival, growth, and productivity of plants in flood conditions (Mustroph, 2018). In our experiment, a positive correlation was found between the root dry weight and the total dry weight of the sugarcane in both the control and the flooding conditions, indicating that the root growth was related to the biomass generation capacity of the sugarcane in both the control and the flooding conditions. Furthermore, the TI of the root dry weight had a strong positive relationship with the TI of the dry weight in different parts of the plant, including the total dry weight of the sugarcane plant. This suggests that if the sugarcane roots were highly tolerant to flooding conditions, this would result in higher tolerance in other parts of the cane plant and the overall tolerance of the plant. It was also discovered that the adventitious root traits were positively correlated with the sugarcane's tolerance to flooding. It should be noted that the ability of sugarcane to form adventitious roots under flooded conditions has resulted in sugarcane genotypes that are more tolerant to flooding conditions. As a result, sugarcane adventitious root traits could be used to select sugarcane genotypes for flooding tolerance.

The sugarcane root growth was affected by the flooding during the early growth stage. There was variation among the genotypes in all the root traits.

Flooding induced the formation of adventitious roots above the soil surface in all the sugarcane genotypes used in the study. The tolerance to flooding conditions varied amongthe sugarcane genotypes. A positive relationship was found between the root traits, the total dry weight, and the TI. The adventitious root traits were also found to be positively related to the flooding tolerance of the sugarcane. As a result, root growth and the ability to form adventitious roots under flooding conditions enhanced the sugarcane's tolerance to flooding conditions.

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