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Root growth, hydraulic conductance and cell wall properties of rice root under interactive effect of growth regulator and limited water

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Abstract: A field experiment was conducted to study the effect of Indole Acetic Acid (IAA) under limited soil water on root growth and root hydraulic conductance of rice (cv. BRRIdhan28). The treatments were H_0: Control; H_1: 50 ppm IAA; H_2: 100 ppm and H_3: 150 ppm IAA for growth regulators and irrigation frequencies were W_1: always flooded condition and W_2: flooded 20 days interval for imposing drought. The root dry weight, root fresh weight, root volume, root surface area, root length, diameter at soil base were increased markedly for 100 ppm IAA under flooded water condition. It was also observed that the combined application of IAA and flooded irrigation were more effective and found to be superior to others treatments for root growth parameters. There is no marked variation in root anatomy and suberin lipid deposition in root cell endodermis layer. The hydraulic conductance of rice cv. BRRIdhan28 under flooded condition had higher than that of drought condition. The plant due to drought or limited water showed low hydraulic conductance.

Key words: Root, hydraulic conductance, rice, root, limited water, IAA

Introduction

Rice is grown in more than hundred countries of the world. It is the most important crops in Asian countries. Among the growing countries, Bangladesh rank's 4th on the basis of area and production (FAO, 1994) while 39th on yield of rice (IRRI, 1995). In Bangladesh average yield of rice is very low, i.e., 2.83 tons/ha, where as in other countries it is very high, the average yield is 6.3, 6.5 and 6.4 tons/ha, in Japan, Korea and Australia, respectively (Karim, 1992). This indicates scope fairly exists to increase yield per unit area. Use of plant growth regulator (PGR) in the interface of moisture availability might be an useful measure to increase rice crop productivity. It is globally convinced today that PGR plays central role in crop growth and yield (Prasad and Paudel, 1994). Many countries like South Korea and China have already started research and use of PGRs in different crops to increase yield. The commercial production of plant growth regulators are used mainly in horticultural crops and considerably in rice (Park, 1995). Plant growth regulators (PGR) are being used as an aid to enhance yield (Nickell, 1982). Naphthalene Acetic Acid (IAA) is one of the growth promoting hormones, may play significant role to change growth character and yield in rice.

Foliar application of growth regulator such as, Indole Acetic Acid (IAA), Naphthalene Acetic Acid (NAA), Ethrel, 2,4-D, Gibberellic Acid (GA_3) and Malic Hydrazide (MH) produce more fertile grain/hill. Foliar application of NAA has also found to increase plant height, number of leaves per plant, fruit size with consequent enhancement in seed yield in different crops (Lee, 1990).

IAA is environment friendly. IAA is of advanced broad spectral and highly effective root growth promoting agent used for improving the production of cereal, vegetables and some especial economically important crops (Tao and Shiying 1993). The studies of NAA on crop production in Bangladesh for cereal crops are very scanty.

Most of our land is medium high land. Boro rice needs continued water logging condition in the rabi season. During this period water supply is limited either by limited supply of irrigation pumps or electricity. The latter along with poor socioeconomic condition, farmers cannot supply required irrigation to boro rice. If it is possible to cultivate boro rice by applying Plant Growth Regulators (PGRs), it will be very helpful and rewarding for our farmer in increasing yield of boro rice. PGRs also increase the root growth and also help promoting new roots. Rice spraying with 10 and 100 ppm NAA at tillering stage significantly increased root dry weight (Wang and Deng, 1992). Boro rice has gained more importance here in Bangladesh, since its average yield per hectare is much higher than that of Aman. This study thus used boro rice variety BRRIdhan28. Examining the combination effect of IAA and water stress for better rice yield is still in initial stage. Work on IAA is limited in our country; while those studied in other countries of the world although provides useful information, but that cannot be recommended or practiced without trial in our local condition.

There is a little information available on root hydraulic conductance, resistance and suberin synthesis under water stress condition. The present approach is to examine the response of hydraulic conductance and resistance of the rice root system for water translocation in plant under water stress. This finding might be useful for studying the plant and soil water relations and water management practices in the agricultural farm for crop selection in arid and semi-arid regions as well as in areas where soil water shortage are in common. The proposed research has focused on measurement of root conductance and also observed the anatomical structure of root and qualitative properties of cell wall. Therefore, a research work has been undertaken to investigate the root growth characteristics of rice under interactive effect of growth regulator and limited water and o quantify the hydraulic conductance of rice root systems to flow water at different developmental stages.

Materials and Methods

Plant culture and treatment description: The experiment was conducted at the Farm of Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur during the Boro season, 2007- 2008. A high yielding variety of boro rice, BRRIdhan28, was used as test variety. Forty days old seedlings were raised and transplanted in the experimental plots on January 25, 2008. Healthy seedlings were uprooted carefully from the seed bed and three seedlings were transplanted in each hill following interows and hill distances of 15×20 cm. Indole acetic acid (IAA) was used in four doses, were: H_0: Control; H_1: 50 ppm IAA; H_2: 100 ppm and H_3: 150 ppm IAA under two different irrigation frequencies, were W_1: always flooded condition and W_2: flooded 20 days interval for imposing drought. The irrigation facilities were provided from the deep tubewell. A total of 24 plots were arranged each with 3 m length and 2 m wide under a RCBD-factorial experimental design. Plant growth regulator (IAA) solution was prepared with an adhesive, namely 1% Tween 20 following the procedure of Roy *et* al., 1992. Spraying was done two times at 20 days interval by using a hand sprayer from tillering to booting stages. The chemical fertilizers Urea, TSP and MOP and Gypsum were applied @ 215 kg, 180 kg, 100 kg, and 20 kg, respectively, per hectare. At the beginning of land preparation one half of urea, full dose of TSP and MOP were applied to the experimental plot. The remaining half of urea was applied in two splits, one at tillering and other at booting stages.

Root data: For growth analysis, data were collected at seediling, vegetative and harvesting stages for root length, root volume, root weight (fresh and dry weight), and root diameter at soil base of rice plants. Roots were collected through careful washing of the soil by spraying water on root mass on a sieve to minimize root loss and damage. The fresh root was gently blotted out surface water for root surface area measurement. The roots were spread on a contrast background and photographs were taken with digital camera. Root surface area (RSA) was calculated by image analysis with LIA32 software using the formula

$$R_{SA} = \frac{I_{TR} \times A_{Ri}}{I_{AR}} \times \pi \quad (m^2) \qquad [1]$$

where I_{TR}, A_{Ri} and I_{AR} are the image area of total root measured by software, actual reference index area on background, and average image of reference index area measured by software, respectively while π is used for 3-D factor. The collected roots were dried at 65 ^{0}C in an oven for 96 h until a constant dry biomass reached. Root dry weight data was taken by digital balance and recorded for analysis.

Root anatomy: Root systems of rice plant under different conditioned treatments were collected from the soil through careful washing. About 10 root samples of secondary root were blotted well and preserved in a preservative of 50% ethanol. For anatomical observation, free hand cross-sections were prepared with a sharp razor blade and stained with Sudan Red 7B (Brundrett *et al.*, 1991). Random cross sections were made at 0.07 to 0.12 m distance from root apex. Stained suberin lamella in the root cell was observed under a light microscope and photographs were taken by a digital camera.

Measurement of hydraulic conductance by aspirator apparatus: The designed rice plant pot was maintained at full saturation by putting the pot in a bigger bucket containing tap water 2 h prior to root excise on the day of experiment (Fig. 1). The root was excised at 4-5 cm above the soil base by a sharp knife. An aspirator was used for measuring root water flow separately at full saturation (Fig. 1). The suction pressure from the aspirator was applied at 0.60 MPa. The $C_{R[-]}$ was calculated using the following formula

$$C_{R[-](SS\ or\ FW)} = \frac{F_{A[-]}}{(\Psi_{Plant} - \Psi_{Out}) \times R_{SA}} \quad (kg\ m^{-2}\ s^{-1}\ MPa^{-1}) \qquad [2]$$

where $C_{R[-]}$, $F_{A[-]}$, Ψ and R_{SA} represent hydraulic conductance, total amount of volume flux per plant, pressure potential and root surface area, respectively.

Fig. 1. Volume flux per plant ($F_{A[-]}$), measurement using aspirator in excised rice root at submerged condition in a bucket.

The data collected on different parameters under the experiment were statistically analyzed to obtain the level of significance using the MSTATC- computer software developed (Russell 1986) and the treatments means were compared by LSD.

Results and Discussion

The effect of different concentration of IAA in combination with different irrigation frequencies on root growth pattern, root anatomy, root hydraulic conductance during different growth stages were markedly varied. The result recorded that the different concentration of IAA both alone and in combination with water significantly increased root weight (Fresh and dry) root length, volume, and surface area over control (Table 1 and 2). At maturity stage, maximum root growth was recorded due to the treatment of 100 ppm IAA under flooded condition. Control plants recorded the lower root length and diameter (Table 3). Result also revealed that the combined applications of IAA and water were more effective to root growth than the treatment of IAA alone. Wang et al. (1992) observed that, rice spraying with 10 and 100 ppm NAA at tillering stage significantly increase root dry weight. In the present study, IAA promoted the root growth in rice BRRIdhan28. Ratna *et. al.* (1995) observed an increase root dry weight with ABA-6 at 20 ppm in soyabean. Tao and Shiying (1992) found higher root volume due to application of ABA, which supported the result of the present study. Root dry weight was recorded at seedling, vegetative and harvesting stages. The result revealed that the stimulation effects on roots were significantly positive (Table 1). Results further revealed that IAA with flooded was more effective than IAA under drought for root production. In contrast, control plants produced the lowest dry weight of roots. 50 ppm IAA showed poor effect on root dry weight. Similarly, root fresh weight of BRRIdhan28 was induced by different concentration of IAA at different growth stages.

Table 1. Root weight (dry and fresh) of rice in response to IAA and soil water.

Growth stage	IAA	Root weight (fresh), g		Root weight (dry), g	
	Concentration	Flooding	Stress	Flooding	Stress
Seedling (Initial)	Control	0.2567 a	0.5733 a	0.05 a	0.05 a
	50 ppm	0.2500 a	0.6067 a	0.05 a	0.05 a
	100 ppm	0.2433 a	0.6267 a	0.05 a	0.05 a
	150 ppm	0.2533 a	0.5467 a	0.05 a	0.05 a
	LSD	0.063	0.109	0.063	0.063
	CV	14	13	5	5
Seedling	Control	0.4267 c	1.033 b	0.100 a	0.1000 a
	50 ppm	0.6433 b	1.067 b	0.100 a	0.1000 a
	100 ppm	0.8500 a	1.277 a	0.100 a	0.1000 a
	150 ppm	0.8333 a	1.157 ab	0.100 a	0.1000 a
	LSD	0.089	0.200	0.063	0.063
	CV	14	15	6	6
Vegetative	Control	49.50 d	53.61a	12.37 c	8.833 a
	50 ppm	56.08 c	52.61a	18.78 b	9.947 a
	100 ppm	71.89 a	53.89a	22.33 a	10.33 a
	150 ppm	61.66 b	51.72a	18.55 b	10.06 a
	LSD	3.09	2.87	2.80	1.48
	CV	9	13	12	14
Harvesting	Control	63.08 c	57.94 b	18.50 c	10.07 a
	50 ppm	74.66 b	59.37 a	24.44 b	10.13 a
	100 ppm	78.89 a	59.47 a	28.67 a	10.20 a
	150 ppm	75.47 ab	59.33 a	24.55 b	10.07 a
	LSD	3.91	0.85	2.34	0.24
	CV	12	15	15	14

There is a significant variations in rice root weight (dry and fresh) amongst the treatment but 100 ppm showed statistically similar to 150 ppm IAA at vegetative stage. Root growth significantly increased using 100 ppm IAA. This was evident in dry and fresh root weight in seedling, vegetative and harvesting stage. Under soil water stress, a statistically similar result was observed. In case of root volume, 100 ppm IAA showed a significant variation for flooded conditions.

The interaction of IAA and limited water or drought showed little changes in the root structure of rice cv BRRIdhan28, producing endodermis without a visible amount of suberin lamellae (Fig. 2). The deposition of suberin lamellae in exodermis and especially in endodermis in roots of stressed plants might be responsible for resisting radial conduction of water from soils to xylem vessels. Under drought condition, a litle amount of suberin lamellae at the exodermis was observed for all the root structure irrespective of their soil water. Plant age and water stress had the principal cause for this suberin deposition in the root cells. There is no marked variation in root structure and suberin deposition in endodermis layer.

Table 2. Root volume and surface area of rice in response to IAA and soil water.

Growth stage	IAA	Root volume (cm3)		Root surface area (cm2)	
	Concentration	Flooding	Stress	Flooding	Stress
Seedling (Initial)	Control	0.4167a	0.4167a	42	42
	50 ppm	0.4067a	0.4067a	41	41
	100 ppm	0.4167a	0.4167a	40	40
	150 ppm	0.4033a	0.4033a	42	42
	LSD	0.063	0.063		
	CV	10	15		
Seedling	Control	1.267 c	2.033 b	129	207
	50 ppm	1.467 b	2.143 b	151	203
	100 ppm	1.823 a	2.710 a	193	221
	150 ppm	1.543 b	2.457 ab	172	227

Table 3. Root diameter and root length of rice in response to IAA and soil water.

Growth stage	IAA	Root diameter (cm)		Root length(cm)	
	Concentration	Flooding	Stress	Flooding	Stress
Seedling (Initial)	Control	0.60	0.60	80	80
	50 ppm	0.60	0.60	80	80
	100 ppm	0.60	0.60	82	82
	150 ppm	0.60	0.60	80	80
Seedling	Control	0.70	0.65	302	320
	50 ppm	0.70	0.55	305	311
	100 ppm	0.70	0.60	321	340
	150 ppm	0.80	0.60	329	349
Vegetative	Control	0.8	0.60	4086	3470
	50 ppm	0.9	0.60	4127	3830
	100 ppm	0.9	0.60	4380	3880
	150 ppm	0.8	0.60	4212	4918
Harvesting	Control	0.90	0.65	5429	4222
	50 ppm	0.95	0.60	5582	4337
	100 ppm	1.00	0.60	5600	4390
	150 ppm	0.90	0.70	5690	4475

The measured flux volume ($F_{A[-]}$) coming out through the root systems at a specific applied hydrostatic pressure varied considerably in response to the stress intensity. The F_A of drought imposed rice roots at applied pressures were always lower than those flooded rice roots. This is also evident in the case of different concentration of IAA. Under different concentration of IAA, the stress experienced rice plants showed lower F_A than those under flooded while the control plant always had the highest. The F_A increases linearly with the increase in time. A similar phenomenon occurs in the roots of many species like rice, maize and olive, where applied hydrostratic driving force changes the contribution of water flow (Lo Gullo *et al.*, 1998, Ranatunge *et al.*, 2003). In the present study it is evident that the F_A of roots of stressed root was always smaller than that of flooded. Under 100 ppm IAA, the F_A of roots was always smaller than that of control. This might be due to some anatomical changes which resist the water permeability of roots under stress and effect of IAA.

The calculated hydraulic conductance (C_R) following equation [2] for different conditioned plants was presented in Table 4.

There were marked variations in C_R among different conditioned treatments. Rice plants under stress showed the lowest conductance than those of control. C_R of test plants using IAA under stress was lower than those of the plants grown under flooded conditions. The mean $C_{R[+]}$ of H_0, H_1, H_2 and H_3 rice plants grown under flooded was 2.92×10^{-5}, 1.15×10^{-5}, 1.27×10^{-5} and 1.35×10^{-5} kg m^{-2} s^{-1} MPa^{-1}, respectively, while corresponding C_R under drought condition were 0.90×10^{-5}, 1.05×10^{-5}, 1.00×10^{-5} and 0.97×10^{-5} kg m^{-2} s^{-1} MPa^{-1}, respectively.

Table 4. Root hydraulic conductance of rice (cv. BRRIDhan-28) in response to different concentration of IAA and soil water.

Growth stage	Concentration of IAA	Root hydraulic conductance (kg m^{-2} s^{-1} Mpa^{-1}) Flooding	Stress
Harvesting	Control	$2.92*10^{-5}$	$1.10*10^{-5}$
	50 ppm	$1.15*10^{-5}$	$1.05*10^{-5}$
	100 ppm	$1.27*10^{-5}$	$1.00*10^{-5}$
	150 ppm	$1.35*10^{-5}$	$0.97*10^{-5}$

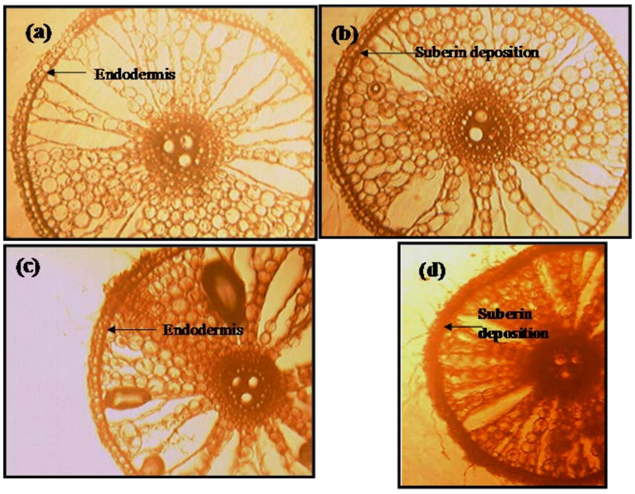

Fig. 2. Free hand cross-sections of a part of the rice roots for showing location of the endodermis and suberin lipids at the root cells grown using IAA interacting with water stress. Figure 2(a) and (b) indicate root section under flooded condition and 2(c) and (d) indicate root section under drought condition

A field experiment was carried out to study the effect of different doses of IAA in combination with soil water on root growth and to ascertain the effective dose of IAA of boro rice cv. BRRIdhan28. Results revealed that all the root growth parameters were improved in the treated plants compared to the control. Amongst the doses, 100 ppm under flooded condition showed the best performance in all morphological characters. The root growth (length, weight, volume, diameter and surface area) increased with the advancement of plant age in all treatments. The 100 ppm IAA under flooded condition had significant positive effect on root length, weight, diameter and surface area over control at all growth stages. There is no marked variation in root structure and suberin deposition in endodermis layer. Limited water or drought in BRRIdhan28 had the reduced hydraulic conductance at different concentration of IAA while flooded condition showed increased conductance in response to IAA at different growth stages.

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