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Effect of growth hormone and cold hardening on summer rice

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ABSTRACT

Four summer rice cultivars, viz. Jyotiprasad, Bishnuprasad, Joymoti and Kolaboro were grown in the farmers field at Kohar Goan, Jorhat during 2004-05. Four different treatments given to 40 days old seedlings before transplanting were control, 4^0 C cold hardening treatment in light for 48 hours, GA_3-10 ppm treatment five days before transplanting and ABA-10 ppm treatment five days before transplanting. Various morphological and physiological parameters, viz plant height, tiller number, leaf number, leaf area, relative leaf water content, leaf chlorophyll, nitrogen, phosphorus and potash content of shoot at harvest were significantly influenced by the treatments. Seedlings treated with GA_3 exhibited better growth and plant vigour in comparison to control. Bio-chemical traits, viz leaf proline content, total soluble sugar in stem, grain yield and yield attributes such as panicle length, grains panicle^{-1}, sterility percentage were also found to be influenced significantly due to GA_3, ABA and 4^0 C hardening treatment. The adverse effect of low temperature on growth and development of summer rice can be reduced with suitable cold hardening treatments. Among the different treatments GA_3 was found to be the most suitable treatment for improvement of cold tolerance in terms of growth and yield in summer rice.

Key words: *rice, cold hardening, growth, hormone, yield*

Injury due to low temperature is reported to be a major constraint of rice production in eastern India while In north-eastern part of India, the third season rice crop (summer rice) experiences chilling temperatures during November to February resulting in yield loss. In Assam cold period prevails at the vegetative phase (Dec-Jan) and sometimes at the flowering stage of summer rice. Low temperature of air and water in irrigated rice and temperature of air and soil in upland rain fed rice causes various types of cold injuries. Low temperature during seedling and vegetative growth stage of summer rice crop affects the establishment of seedling and prolongs the duration of the crop (Gogoi and Baruah, 1999) and it affects the transplanting of subsequent autumn rice crop. Chilling temperature at flowering stage of rice causes spikelet sterility and reduces yield. Therefore, it is desirable to have cold tolerance in high yielding varieties of summer rice during vegetative and reproductive phase for sustainable rice production. Cold hardening treatments have some beneficial effects on metabolic, bio-chemical and physiological processes of plant, which improves the cold tolerance in different crops. (Peter *et al*, 2003; Kaarina *et al*, 2003; Dionne *et al*, 2001). The present investigation was undertaken to study the effect of cold hardening and growth hormone at seedling stage on growth and yield of field grown summer rice.

MATERIALS AND METHODS

An experiment was conducted in farmer's field at Kohar Goan, Jorhat, during 2004-05. The experiment was laid out in split plot design with four summer rice varieties (Jyotiprasad, Bishnuprasad, Joymoti and Kolaboro). Four different treatments were given to the seedlings before transplanting. The first set was kept as control, second set was given 4^0 C cold hardening treatment in light for 48 hours, third set was given GA_3- at a dose of 10 ppm treatment five days before transplanting and fourth set was given abscisic acid (ABA) at a dose of 10 ppm treatment five days before transplanting. Treatments were replicated four times in a split plot design. Forty days old seedlings of these varieties were transplanted in the field. Samples were collected during tillering and panicle initiation stage for various morphological, physiological and bio-chemical analyses. Leaf area was recorded in leaf area meter (LICOR, LI 3000, USA). Leaf chlorophyll content and total soluble sugar of stem were recorded by the methods of Anderson and Boardman (1964) and Yem and Willie (1954), respectively. Proline in the leaves and relative water content were determined by the methods of Bates *et al* (1973) and Matin *et al*. (1989), respectively. Nitrogen, phosphorus and potassium content of shoot were determined by Microkjeldahl, Colorimetric and Flame photometry methods, respectively. Grain yield and yield attributing parameters were recorded at maturity.

RESULTS AND DISCUSSION

Significant improvement in shoot growth in terms of plant height, tiller number, leaf number, leaf area was observed due to application of GA_3, ABA and cold hardening treatment (Table 1). Commonly observed cold injuries in rice are failure to germinate, delayed seedling emergence, stunting, leaf discoloration, panicle tip degeneration, incomplete panicle exertion, delayed flowering, high spikelet sterility and irregular maturity. Low temperature adversely affects the growth of shoots by some secondary effects which include (i) nutrient deficiencies or toxicities, (ii) increase in resistance to water flow and hence water deficits and/or (iii) changes in growth regulators (Nielsen and Humphris, 1966). Exogenous application of GA_3 increases plant height, inter node elongation and leaf area. (Carlson *et al*, 1990; Kim and Heu, 1990; Gogoi and Baruah, 2000). Most reports have demonstrated that exogenous application of ABA and low temperature hardening treatment provides tolerance to various stress condition (Chen and Li, 2005; Kazemitabar *et al*, 2002; Kerepesi *et al*, 2004). Increased plant height, tiller number, leaf area in present experiment revealed that these treatments increased the cold tolerance ability of summer rice. Low temperature causes reduction in plant height and increases growth duration in rice (Lee, 1989) which was observed in non-hardened plants (Table 1). Significant increase in leaf water content was recorded in plants treated with GA_3, ABA and cold hardening at tillering and panicle initiation stage (Table 2). This is because of maintenance of a higher amount of sugar and proline in hardened plants, which helps in water relation and maintains the osmoticum of the cell. (Guinchard *et al*, 1997; Gogoi and Baruah,1999). Accumulation of solutes in the cell sap leads to osmotic

Table 1. Effect of growth hormone and cold hardening treatments on plant height, tiller number, leaf number and leaf area during maximum tillering stage of selected rice varieties.

Treatment	Cultivars	Plant height (cm)	Tiller number $hill^{-1}$	Leaf numberhill^{-1}	Leaf area(square cm $hill^{-1}$)
Control	Jyotiprasad	35.48	14.25	49.75	412.00
	Bishnuprasad	35.68	13.25	49.50	383.50
	Joymoti	36.30	15.50	56.50	443.25
	Kolaboro	39.63	10.75	39.50	304.25
4^0 C	Jyotiprasad	37.48	14.75	55.60	462.50
	Bishnuprasad	36.43	13.75	54.75	426.33
	Joymoti	37.55	16.50	61.92	484.29
	Kolaboro	41.18	11.50	41.75	331.38
GA_3-10ppm	Jyotiprasad	41.08	16.75	60.00	474.00
	Bishnuprasad	40.10	15.75	57.50	447.56
	Joymoti	42.23	18.00	65.00	503.53
	Kolaboro	45.38	12.00	46.75	343.49
ABA-10ppm	Jyotiprasad	37.60	15.75	58.50	469.75
	Bishnuprasad	37.18	14.75	56.00	431.09
	Joymoti	38.50	17.50	63.50	492.27
	Kolaboro	42.30	12.5	44.50	336.83
	Treatment	CD(5%)	CD(5%)	CD(5%)	CD(5%)
	Variety	1.93	1.19	3.97	20.06
	Treatment x Variety	1.76	2.57	13.98	23.40
		2.47	2.37	7.94	43.140

Maximum tillering stage (Days after transplanting; DAT): Jyotiprasad-30 DAT, Bishnuprasad-30 DAT, Joymoti-32 DAT, Kolaboro -35 DAT

Table 2. Effect of growth hormone and cold hardening selected biochemical parameters of rice plant.

Treatment	Cultivars	Total Chlorophyll (mg g-fr.wt)	Leaf praline (μg g^{-1} fr.wt.)	Total soluble sugar (mg g^{-1} fr.wt.)	Nitrogen (%)	Phosphorus (%)	Potash (%)	Relative leaf water content (%) Tillering stage	Relative leaf water content (%) Panicle initiation stage
Control	Jyotiprasad	1.72	493	3.18	2.13	0.338	1.835	79.50	71.15
	Bishnuprasad	1.87	487	3.23	2.10	0.375	1.793	80.38	72.20
	Joymoti	1.68	495	4.03	2.24	0.458	1.978	80.03	71.03
	Kolaboro	1.67	514	3.20	2.00	0.383	1.293	80.08	69.48
4^0 C	Jyotiprasad	2.15	512	3.36	2.33	0.373	2.041	82.18	73.23
	Bishnuprasad	2.10	520	3.47	2.30	0.417	1.974	82.58	73.53
	Joymoti	1.86	536	4.28	2.43	0.514	2.220	81.38	73.16
	Kolaboro	1.79	546	3.27	2.15	0.420	1.400	81.18	71.25
GA_3 (10ppm)	Jyotiprasad	2.21	574	3.54	2.93	0.420	2.246	83.40	74.08
	Bishnuprasad	2.22	596	3.68	2.87	0.471	2.158	83.28	74.38
	Joymoti	1.96	590	4.67	2.93	0.565	2.340	82.48	74.13
	Kolaboro	1.84	584	3.41	2.31	0.446	1.453	82.28	72.15
ABA (10ppm)	Jyotiprasad	2.12	566	3.51	2.82	0.431	2.236	83.28	74.70
	Bishnuprasad	2.14	574	3.56	2.76	0.472	2.248	83.65	74.53
	Joymoti	1.81	568	4.59	2.86	0.568	2.412	81.68	74.08
	Kolaboro	1.80	571	3.46	2.28	0.452	1.420	81.37	71.48
CD (P=0.05)	Treatment	0.172	9.11	0.31	0.195	0.033	0.169	0.51	0.77
	Variety	0.277	13.66	0.42	0.147	0.082	0.183	0.74	1.12
	TreatmentxVariety	0.346	18.21	0.63	0.390	0.078	0.338	1.02	1.30

Maximum tillering stage (Days after transplanting; DAT): Jyotiprasad-30 DAT, Bishnuprasad-30 DAT, Joymoti-32 DAT, Kolaboro -35 DAT.
Panicle initiation stage : Jyotiprasad-53 DAT, Bishnuprasad-56 DAT, Joymoti-68 DAT, Kolaboro -56 DAT.

adjustment, which helps to maintain the water content and turgor of living cells (Steponkus, 1984). Increased osmoticum in terms of soluble sugars (Gonzales *et al*, 1990) and proline leading to a better hydration status in cold hardened plants suggested by Guinchard *et al*, (1997), may be the reason for higher relative leaf water content observed in the study (Table 2).

Significant variation in chlorophyll content due to GA_3, ABA and cold hardening treatment was recorded among the varieties (Table 2). Decrease in chlorophyll content in leaves is reported to be associated with decrease in nitrogen in the leaf blades of cold intolerant rice cultivars (Setter and Greenway, 1988). GA_3, ABA and 4^0 C cold hardening treatment have recorded significant improvement in nitrogen content of leaves (Table 2) which may be related to the increased chlorophyll content in leaves. All the four varieties showed variation in nitrogen, phosphorus and potassium content (Table 2) when subjected to hardening treatments. The low temperature induced nutrient deficiency in root affects the level of growth regulators and therefore, reduces the growth of shoot (Setter and Greenway, 1988). Nitrogen, Phosphorus and Potassium content can be used as indices for cold tolerance in rice. Higher phosphorus content is associated with low temperature tolerance (Gogoi and Baruah, 1999). Total soluble sugar content increased significantly in GA_3, ABA and 4^0C cold hardening treatments in comparison with non-hardened plant (Table 2). These findings are in conformity with the findings of Rosens *et al* (1993) in clover and Gogoi and Baruah (1999) in rice. Sugar accumulation during acclimation and foliar application of GA_3 has been reported in different plant species (Guinchard *et al*, 1997; Chetterjee *et al*, 1975) resulting in elevation of osmotic concentration. Carbon compounds released from starch hydrolysis is associated with low temperature tolerance for osmotic regulation, re-growth and maintenance. Higher amount of sugar in Jyotiprasad, Bishnuprasad and Joymoti indicated their acclimation to cold. Significant increase in proline content was recorded in the plants treated with GA_3, ABA and 4^0C cold hardening in the present experiment (Table 2). GA_3

Table 3. Effect of growth hormone and cold hardening on yield attributing parameters and yield of summer rice.

Treatment	Cultivars	Plant height (cm)	Grain panicle^{-1}	High density grain (%)	1000 grain wt. (g)	Yield (t ha^{-1})
Control	Jyotiprasad	10.0	106.75	72.53	24.06	3.58
	Bishnuprasad	9.0	102.50	71.70	23.90	3.52
	Joymoti	10.25	112.50	72.38	24.10	3.74
	Kolaboro	8.0	75.0	71.48	22.00	2.51
4^0 C cold hardening	Jyotiprasad	11.0	109.00	73.68	24.30	3.60
	Bishnuprasad	10.0	105.75	73.35	24.17	3.57
	Joymoti	11.0	116.00	74.45	24.30	3.76
	Kolaboro	8.0	77.50	72.38	22.16	2.53
GA_3 (10ppm)	Jyotiprasad	12.0	113.75	75.93	24.46	3.64
	Bishnuprasad	11.00	110.00	75.28	24.24	3.63
	Joymoti	12.00	120.00	76.38	24.49	3.77
	Kolaboro	9.00	81.50	73.13	22.27	2.55
ABA (10ppm)	Jyotiprasad	11.0	112.00	74.33	24.39	3.61
	Bishnuprasad	10.0	108.50	74.15	24.27	3.60
	Joymoti	11.0	117.50	74.73	24.36	3.76
	Kolaboro	8.5	81.50	72.95	22.24	2.54
CD (P=0.05)	Treatment	0.46	4.35	0.79	0.34	0.11
	Variety	0.60	3.90	0.45	2.70	0.77
	Treatment x Variety	0.91	6.91	1.07	0.57	0.23

and low temperature induced accumulation of proline has been reported by Charest and Phan (1991) in wheat and Gogoi and Baruah (1999) in rice. Proline accumulation provides a mechanism for the maintenance of osmoticum in cells and tissues (Levitt, 1980).

In the present study application of GA_3, ABA and 4^0C cold hardening significantly increased the panicle hill^{-1}, improved the grain filling capacity in terms of increasing percentage of high density grain, increased thousand grain weight and yield (Table 3). Low temperature in vegetative and reproductive phases ultimately results in less number of spikelet per panicle, increased spikelet sterility and decreased grain weight leading to yield reduction (Gogoi and Baruah, 1999). In the present investigation spikelet sterility was found to be higher in non-hardened plants (Table 3). Low temperature decreases the filled grain percentage by affecting sink capacity or by increasing the length of ripening period (Tanaka, 1962). Hardening of rice seedling is reported to increase rice yield in cold climate (Gogoi and Baruah, 2000) and we also obtained yield improvement due to GA_3, ABA and 4^0 C cold hardening treatments.

REFERENCES

Anderson JN and Boardman NK 1964. Studies on greening of dark brown bean plants. IV. Development of photochemical activity. Aust J Biol Sci.,17:93-101

Bates LS, Waldren RP Teare ID 1973. Rapid determination of free proline for water stress studies. Plant Soil, 39:205-207

Charest C and Phan CT 1991. Kinetic properties of four enzymes involved in cold acclimated wheat. J Exp Bot., 42 :673-678.

Chatterjee A, Mandal RK and Sircar M 1975. Effect of growth substances on productivity, photosynthesis and translocation of rice varieties. Indian J. Plant Physiol. 19(1&2): 131-138

Carlson RD, Adair HM, Fugiel J and Monsalud M 1990. Effect of GA on semi dwarf rice : commercial application. Proc. Plant Growth Regulators Soc.; America 1:208.

Chen WP and Li PH 2005. Membrane stabilization by abscisic acid under cold aids proline in alleviating chilling injury in maize (Zea mays L) cultures cell. Plant, Cell and environment. 25:95-98

Dionne J, Castonguay Y, Nadeau P and Desjardins Y 2001. Freezing tolerance and carbohydrate changes during cold acclimation of green-type annual bluegrass (*Poa annua* L.) ecotypes. Crop Sci 41:443–451

Gonzalez B, Boucaud J, Salette J and Langlois J 1990. Fructan and Cryoprotection in rye grass (*Lolium perenne* L)

New Phytol., 155:319-323

Gogoi Nirmali and Baruah KK 1999. Effect of cold and chemical hardening on growth , yield and some biochemical characters in summer rice rice (*Oryza sativa* L.) Indian J Plant Physiol., 4:179-184

Gogoi Nitumoni and Baruah KK 2000. Effect of cold hardening and GA_3 on growth and yield of 'boro' rice. Indian J Plant Physiol. 5:339-343

Guinchard MP, Robin Ch., Grieu, Ph. and Guckert A 1997. Cold acclimation in clover subjected to chilling and frost : changes in water and carbohydrate status. European J Agron 6:225-233

Kaarina Pihakaski, Tamminen I, Milla Pietianen and Marilyn Griffith 2003. Antifreez proteins are secreted by winter rye cell in suspension culture. *Physiologia Plantarum*. 118:390-393

Kazemitabar SK, Tomsett AB, Collin HA, Wilkinson M. and Jones MG 2002. Effect of short term cold stress on rice seedlings. *Euphytica*. 129 (2):193-200

Kerepesi I, Banyai-Stefanovits E, Galiba G 2004. Cold acclimation and abscisic acid induced alterations in carbohydrate content in calli of wheat genotypes differing in frost tolerance. J Plant Physiol 161:131-133

Kim YK and Heu MH 1990. Segregation of culm length and GA response in crosses of dwarf cultivars. Korean J Crop Sci 35(2): 165-170

Levitt J 1980. Responses of plants to environmental stresses. Vol.I, Chilling, Freezing and high temperature stresses. Academic Press New York. pp.218-350.

Lee JH 1989. Screening methods for cold tolerance at Crop Experiment Station Phytotron and at Chuncheon. Report of Rice Cold Tolerance Workshop, IRRI.

Matin MA, Brown JH and Ferguson H 1968. Leaf water potential, relative leaf water content and diffusive resistance as screening techniques for drought resistance in barley. Agron J., 81:161-168

Neilsen KF and Humphris EC 1996. Effects of root temperatures on plant growth, soils and fertilizers. Abs. World Literature. 29:1-7

Peter Streb, Serge Aubert, Elisabeth Gout and Richard Bligny 2003. Cold and light induced changes of metabolic and anti-oxidant levels in two high mountain plant species *Soldanella alpine* and *Ranunculus glacialis* and lowland species Pisum sativum. Physiologia Plantarum, 118:96-104.

Rosen K, Juntilla O, Ernsten A and Sandli N 1993. Development of cold tolerance in white clover Trifolium ripens L in relation to carbohydrate and free amino acid content. Acta Agric Scand Sect B. Soil Plant Sci., 43 : 151-155

Setter TL and Greenway H 1988. Growth reduction of rice at low root temperature; decrease nutrient uptake and development of chlorosis. J.Expt Bot, 39:811-829.

Steponkus PL 1984. Role of the plasma membrane in freezing injury and cold acclimation. Annu. Rev Plant Physiol 35:543-584.

Tanaka M 1962. Studies on the growth of low land rice caused by cool water irrigation and delayed heading. Bull.Aomori.Agric.Exp. Stn., 7:1-107 N

Yem EW and Willie AJ 1954. The estimation of carbohydrates in plant extract by anthrone. Biochem. J, 57: 5008