SCREENING OF COLD TOLERANT, SHORT DURATION AND HIGH YIELDING RICE LINES SUITABLE FOR NORTHERN PART OF BANGLADESH

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Abstract

Rice, like many crops is lack of acclimatization ability to cold temperature that limits reducing crop yield worldwide. Three cold tolerant, long duration, and short bold grain (Manjusree-2, Komol-7 and Komol-9) exotic varieties were backcrossed with popular high yielding, short duration, long slender grain, Binadhan-17 to develop cold tolerant, short duration with fine grain quality rice varieties for ensuring stable yields over cold stress environments in Bangladesh. Eighteen BC₁F₄ backcross lines along with check variety were evaluated over three locations, viz. Nilphamari, BINA Substation Rangpur, and BINA HQs farm, Mymensingh at reproductive stage. Among them six lines of Manjusree-2, three lines of Kamol-7 and six lines of Kamol-9 showed cold acclimatization with high yield potential. Crop duration was 144-160 days depending on the locations and varieties. Although the plant height of these lines was tall but did not lodge. Yield attributing characteristics of these lines were found better than parents with higher number of effective tillers, filled grains, long panicle and higher yield. Therefore, these advanced lines could be released as cold tolerant rice varieties along with high yield for Northern cold prone areas of Bangladesh after further trials and following variety release procedure.

Keyword: Cold tolerant, short duration, high yield, rice

Introduction

In Bangladesh, around 2 million hectare of rice area becomes affected by low temperature during winter season causing seedling mortality in some years up to 90% and thereby increases cost of cultivation. The population is rising at 1.37 per cent annually but arable land is decreasing at 0.4 percent (BBS, 2019-20) in Bangladesh. In north-eastern haor areas of Bangladesh, the land remains submerged most of the time and around 1.26-million hectares of cultivable land are restricted to grow only boro paddy every year. Boro season usually begins in mid-November but many farmers start sowing in late October to avoid flash floods. As a result, the harvesting time fall sometime in January-February that triggers yield loss due to cold stress (Rashid and Yesmin, 2017). However, extreme climate change such as cold stress has already raised a radical change in northern regions and north-eastern haor regions in Bangladesh and threatened the productivity of the existing rice varieties (Hakim et al., 2014). Therefore, to utilize these areas for rice cultivation, development of suitable cultivars could be the best approach.

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Rice is widely cultivated around the world in both temperate and high-elevation (Andaya and Mackill, 2003) environments in tropical and subtropical areas as well as in irrigated areas which rely on the use of cold water (Ye *et al.*, 2010). But low temperatures is a major limitation for rice production in those zones that can reduce up to 25% of the final yield in rice genotypes (Lima *et al.*, 2012). Two types of cold stress observed in rice- at young vegetative stage: heading-delay type occur which results in low spikelet fertility and at reproductive stage, spikelet-sterility type occurs which result in poor grain filling of rice crop (Andaya and Tai, 2006). Although rice is sensitive to low temperature stress, a range of cold tolerance exists among rice cultivars (Lou *et al.*, 2007) and it is reported that Indica rice is more sensitive to cold stress than Japonica rice (Lv *et al.*, 2016) and the damage at reproductive stages is usually significantly greater than damage at the seedling stage (Pan *et al.*, 2015). It has been shown that, spikelet sterility increases by up to 90% if reproductive stages exposed at 15°C (day) and 10°C (night) temperature and reduces grain eating quality (Jacobs and Pearson, 1999).

The objectives of this study were to screen rice lines showing high levels of cold stress tolerance, short duration and higher yield.

Materials and method

Thirteen genotypes (10 from cold prone area of Nepal and 3 from Bangladesh) of 21 day old seedlings were subjected to cold stress (10 °C) in a growth chamber for three weeks to evaluate their cold response. Three genotypes viz. Manjusree-2, Kamol-7 and Kamol-9) were selected based on their cold tolerant and hybridized with high yielding short duration variety Binadhan-17 to develop cold tolerant progeny. The F₁ seeds of each cross progenies were backcrossed with Binadhan-17 as the selected exotic genotypes were low yielding and long duration varieties. This study was carried out at the pot (crossing), laboratory (molecular) and the experimental field of Bangladesh Institute of Nuclear Agriculture (BINA) Headquarters, Mymensingh, BINA Sub-station Rangpur and farmer's field Nilphamari. The pot experiments were set to produce F₁ and backcross generations since 2018. Crossing materials were grown at BINA HQs farm Mymensingh to advance the generation and their performance was evaluated. Based on yield and yield attributing characters preliminary screening was done from BC₁F₁₋₃ generation at BINA Headquarters farm, Mymensingh. 120 lines from BC₁F₂ generation and 49 lines from BC₁F₃ generation were selected. Next year further evaluation was done from BC₁F₄ generation at two locations of the northern part of Bangladesh e.g. Rangpur and Nilphamari including BINA Headquarters, Mymensingh and the result was compiled accordingly. In experimental field, plant to plant and row to row distance was maintained by 20 cm and 15 cm, respectively. The experiment was followed by non-replicated design. A unit plot size was 2m × 1m. Recommended doses of nitrogen, phosphorus, potassium, sulphur and zinc were applied in the form of Urea, TSP, MoP, Gypsum and Zinc Sulphate. Cultural and intercultural practices were followed as and when necessary. The data of different parameters were taken from five plants of each plot and the average value was recorded. Statistical analysis was done by t-test. The yield data were recorded according to plot basis. Two cold responsive genes ICE and DREB were survey to investigate the cold tolerant gene introgression into the BC₁F₄ population from exotic variety. Genomic DNA was isolated according to CTAB method (Doyle and Doyle, 1987). PCR analysis was performed in 10 μ l reaction sample containing 50 ng of DNA template of 2 μ l, 5 μ l of master mix, 2 μ l nuclear free water, 1 μ l each of 10 μ M forward and reverse primers using Biometra T₃ thermal cycler with single 96-well. After initial denaturation for five minutes at 94°C, each cycle comprised one minute denaturation at 94°C, one min annealing at 55°C, and two min extension at 72°C with a final extension for 7 min at 72°C at the end of 35 cycles. The PCR products were analyzed by electrophoresis on 8% polyacrylamide gel using mini vertical polyacrylamide gels for high throughput manual genotyping (CBS Scientific Co. Inc., CA, USA). Two μ l of amplification products were resolved by running gel in 1X TBE buffer for 2-2.5 hrs depending upon the allele size at around 80 volts and 400 mA current. The gels were stained in 0.5 mg ml⁻¹ ethidium bromide and were documented using Whatman Biometra Gel Documentation System (prod nr: 1603209).

Result and discussion

From previously harvested 49 lines of BC₁F₃ generation (data not shown) was grown at BINA HQs, Mymensingh in boro season 2019-2020. Based on yield performance the best lines were selected for further evaluation. During 2020-2021 a non-replicated experiment was conducted with selected 18 lines of BC₁F₄ generation for each three locations. Almost all locations, plant height was observed taller for all lines compared to Binadhan-17 but lodging was not found in any location. It has been reported that although plant height is the major contributor to the lodging tolerance but not only genetic gain in lodging tolerance can be obtained, also long with strong culm resists lodging that is independent of plant height (Navabi *et al.*, 2006: Nomura *et al.*, 2019).

From BC₁F₃ generation, 21 lines of Monjusree-2, ten lines of komol-7, and 18 lines of komol-9 have been selected according to the higher tiller numbers, longer panicle length, maximum filled grains plant⁻¹ and yield hill⁻¹. Among them 18 (seven from Monjusree-2, five from komol-7 and six from komol-9) promising lines from BC₁F₄ generation were transplanted in BINA Hqs farm, Mymensingh, Sub-station farm, Rangpur and farmer's field, Nilphamari.

Crop duration was observed medium in all population and significant variation was observed in no. of effective tillers comparing to the both parents (Table 1). At Mymensingh, M2-P-15, M2-P-16 produced higher number of effective tillers plant⁻¹. Longer panicle length (25-27 cm) was observed in M2-P-5, 8, 10 and 15. M2-P-10 exhibited highest no. of filled grains (208.2) and less no. of unfilled grain panicle⁻¹ (26.4) comparing to the both parents and gave higher yield plant⁻¹ comparing to the recipient parent (Table 1). Considering the agronomic performance including yield, M2-P-5, 8, 10, 15 and 16 were the best. The superior performance of backcross lines may have resulted from breakage of undesirable linkages or loss of negative interactions as it went through one round of

backcrossing and advancement. The field based phenotypic selections based on different agro-morphological characters at each generation helped in identification of lines with superior performance over recurrent parent in terms of quality and abiotic stress tolerance (Kush and Zena, 2009).

At BINA Sub-station, Rangpur crop duration and no. of effective tillers plant⁻¹ were higher than Mymensingh. The range of no. of effective tillers was 11.8 to 18.8 per plant. Longer panicle was observed in M2-P-5. The highest no. of filled grains was observed in M2-P-5 and M2-P-16 than the both parent (Table 1). The lines M2-P-5, M2-P-8, M2-P-10, M2-P-14, M2-P-15 and M2-P-16 produced higher yield comparing to the both parent (Table 1). These seven lines can be evaluated for the next trial for Rangpur region where even small amounts of temperature fluctuation lead to decline a large amount of rice yield and Cold tolerant lines would be best possible solution of higher rice production (Rokonuzzaman *et al.*, 2018).

Table 1. Evaluation of BC_1F_4 generation of Monjusree-2 based on grain yield and yield components during boro season 2020-21 at three locations

	•	O					
		Duration	Plant	Effective	Panicle	Filled grain	Yield
Location	Variety/Line	(days)	height	tiller plant ⁻¹ length		panicle ⁻¹	(t ha ⁻¹)
-			(cm)	(no.) (cm)		(no.)	
	M2-P-5	146 ± 0.71	102.6±1.88	7 ± 0.45	26.6 ± 0.32	163.6 ± 4.82	7.5 ± 0.34
	M2-P-8	145 ± 0.71	110.2±1.88	6.8 ± 0.45	27.4 ± 0.32	146.8 ± 4.82	6.6 ± 0.34
Ζ	M2-P-10	147±0.71	122.4±1.88	7 ± 0.45	25.4 ± 0.32	208.2 ± 4.82	7.6 ± 0.34
УĦ	M2-P-14	147±0.71	99.6±1.88	7.8 ± 0.45	23.2 ± 0.32	137.8 ± 4.82	5.1 ± 0.34
en	M2-P-15	146±0.71	112.2±1.88	10.4 ± 0.45	25.8 ± 0.32	149.4 ± 4.82	7.5 ± 0.34
Mymensingh	M2-P-16	148 ± 0.71	107.4 ± 1.88	8.6 ± 0.45	24 ± 0.32	159.4 ± 4.82	7.2 ± 0.34
gh	M2-P-19	150 ± 0.71	102.6±1.88	7.6 ± 0.45	23.6±0.32	136.2 ± 4.82	3.4 ± 0.34
	Monjusree 2 (P)	150 ± 0.71	117.8 ± 1.88	7.6 ± 0.45	22.6±0.32	148.2 ± 4.82	3.2 ± 0.34
	Binadhan-17 (P)	158 ± 0.71	98.4 ± 1.88	13.8 ± 0.45	23.0 ± 0.32	156.8 ± 4.82	7.1 ± 0.34
	M2-P-5	157±0.92	124.4±6.59	15±0.91	24.82±0.87	145.4±9.34	8.3±0.65
	M2-P-8	156 ± 0.92	137±6.59	14.6 ± 0.91	22.8 ± 0.87	94.4 ± 9.34	9.7 ± 0.65
	M2-P-10	157 ± 0.92	135.8±6.59	16.6 ± 0.91	22.2 ± 0.87	107.8 ± 9.34	8.1 ± 0.65
Rangpur	M2-P-14	157 ± 0.92	85.4 ± 6.59	16 ± 0.91	19.7 ± 0.87	115.2 ± 9.34	8.5 ± 0.65
ng	M2-P-15	157 ± 0.92	131.2±6.59	11.8 ± 0.91	16 ± 0.87	121 ± 9.34	8.5 ± 0.65
ımd	M2-P-16	157 ± 0.92	141.8±6.59	18.8 ± 0.91	22.9 ± 0.87	182.8 ± 9.34	9.9 ± 0.65
•	M2-P-19	157 ± 0.92	101.6±6.59	12.2 ± 0.91	22 ± 0.87	116.4±9.34	6.2 ± 0.65
	Monjusree 2 (P)	151±0.92	115.2±6.59	9.8 ± 0.91	22.6 ± 0.87	148.2 ± 9.34	4.6 ± 0.65
	Binadhan-17 (P)	162 ± 0.92	99.5±6.59	14.1±0.91	23.8 ± 0.87	156.8 ± 9.34	7.0 ± 0.65
	M2-P-5	160±0.82	103.6±6.13	19.8±1.27	21.7±0.43	148±10.93	8.9±0.59
Nilphamari	M2-P-8	159 ± 0.82	130.2±6.13	14.2 ± 1.27	22.6±0.43	97.2±10.93	9.3 ± 0.59
	M2-P-10	160 ± 0.82	135.4±6.13	16.2 ± 1.27	23.3±0.43	81±10.93	9.6 ± 0.59
	M2-P-14	159±0.82	84.8 ± 6.13	13±1.27	19.4±0.43	118.2±10.93	9.5 ± 0.59
	M2-P-15	159±0.82	129.2±6.13	11.8±1.27	21.6±0.43	133.2±10.93	6.0 ± 0.59
	M2-P-16	160±0.82	138.6±6.13	19.6±1.27	22.5±0.43	181.2±10.93	9.2 ± 0.59
≦.	M2-P-19	159±0.82	112±6.13	10.4±1.27	21±0.43	139±10.93	8.5±0.59
	Monjusree- 2 (P)	153±0.82	115±6.13	8.8 ± 1.27	22.5±0.43	145.2±10.93	4.3±0.59
	Binadhan-17 (P)	162±0.82	98.7 ± 6.13	14 ± 1.27	23.7±0.43	160.18±10.93	6.1±0.59
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At Nilphamari, crop duration was little higher than Rangpur. All the lines produced higher No. of effective tillers plant⁻¹, among them M2-P-5, M2-P-8, M2-P-10 and M2-P-16 had the highest no. of effective tillers⁻¹ (Table 1). Panicle length was the same for all the populations comparing to the both parents. M2-P-16 had highest no. of filled grain panicle⁻¹ (181.2) comparing to the both parents. The Backcross lines M2-P-5, M2-P-8, M2-P-10, M2-P-14, M2-P-16 and M2-P-19 produced higher yield compare to the recipient parent (Table 1). These lines can be evaluated for the next trial. Agriculture of Nilphamari district is highly vulnerable in weather patterns and is therefore extremely at risk from climate change in aspect of temperature fluctuation, dew point temperature and sunshine period that contribute to the spikelet sterility in cold susceptible rice either in vegetative and reproductive phase (Islam *et al.*, 2021).

Five lines from BC₁F₄ generation of parent, Komol-7 were evaluated at all three locations. Maturity date was shorter in Mymensingh than in Northern part of Bangladesh. Total number of effective tillers plant⁻¹ was observed higher than parent Komol-7 but lesser than Binadhan-17 in all locations (Table 2). The backcross lines K7-P-1, K7-P-2, K7-P-3, K7-P-4, K7-P-7 and K7-P-9 had longer panicle (24.2 cm to 26.6 cm) than the recipient (23.4) and donar parent (23.8) at Mymensingh but had the similar length at Rangpur and

Table 2. Evaluation of BC_1F_4 generation of Komol-7 based on grain yield and yield components during boro season 2020-21 at three locations

		Duration	Plant	Effective	Panicle	Filled grains	Yield
Location	Variety/Line	(days)	height	tillers plant ⁻¹	length	panicle ⁻¹	(t ha ⁻¹)
			(cm)	(no.)	(cm)	(no.)	
	K7-P-1	149 ± 1.16	138.4 ± 4.63	7.6 ± 0.74	26.6 ± 0.49	176.4 ± 7.95	6.8 ± 0.25
Z	K7-P-2	148 ± 1.16	121.2±4.63	7.8 ± 0.74	24.2 ± 0.49	180 ± 7.95	7.5 ± 0.25
[ym	K7-P-3	150±1.16	146 ± 4.63	6.2 ± 0.74	26.6 ± 0.49	229.4±7.95	7.2 ± 0.25
ıen	K7-P-5	149±1.16	128 ± 4.63	6.4 ± 0.74	21.8±0.49	213.4±7.95	7.1 ± 0.25
Mymensingh	K7-P-9	153±1.16	129.6±4.63	7.4 ± 0.74	25.8±0.49	224 ± 7.95	8.9 ± 0.25
gh	Komol-7(P)	157±1.16	113.2 ± 4.63	5.6 ± 0.74	23.4 ± 0.49	170.8 ± 7.95	5.3 ± 0.25
	Binadhan-17(P)	158±1.16	98.4 ± 4.63	13.8 ± 0.74	23.8±0.49	156.8±7.95	7.2 ± 0.25
	K7-P-1	152±1.16	116.4±4.63	7.8±0.71	22±0.49	131.2±7.95	6.0±0.25
	K7 -P-2	158±0.24	140.2±9.23	12.2±1.25	24.2±0.31	164.8±11.28	9.4 ± 0.74
Ra	K7-P-3	148±1.16	121.2±4.63	6 ± 0.72	20.2±0.49	180 ± 7.95	5.3 ± 0.25
Rangpur	K7-P-5	157±0.24	151.6±9.23	11.6±1.25	22.4 ± 0.31	141.4±11.28	9.1±0.74
nuc	K7-P-9	157±0.24	131.8 ± 9.23	11.6±1.25	23.4±0.31	118.6±11.28	9.4 ± 0.74
	Komol-7(P)	157±0.24	115.1±9.23	6.4 ± 1.25	23.5 ± 0.31	175.1±11.28	5.2 ± 0.74
	Binadhan-17(P)	158 ± 0.24	99.4 ± 9.23	13.8 ± 1.25	23.9±0.31	160.3±11.28	6.5 ± 0.74
Nilphamari	K7-P-1	155±1.16	118.4±4.63	7.8±0.31	22±0.49	139.2±7.95	6.3±0.25
	K7-P-2	160±0.37	135±7.11	11.4±1.21	22.6±0.25	146.6 ± 9.74	9.2 ± 0.74
	K7-P-3	157±1.16	129.2±4.63	8 ± 0.72	20.2±0.49	140 ± 7.95	5.2 ± 0.45
	K7-P-5	159±0.37	135.8±7.11	11.6±1.21	23.9±0.25	154.8±9.74	9.3 ± 0.74
	K7-P-9	160±0.37	132.2±7.11	10.8±1.21	23.8±0.25	116.4±9.74	8.8 ± 0.74
₽.	Komol-7(P)	159±0.37	115.1±7.11	6.4 ± 1.21	23.5±0.25	175.1±9.74	5.3±0.74
	Binadhan-17(P)	160±0.37	99.4±7.11	13.8±1.21	23.9±0.25	160.34±9.74	6.3±0.74

Nilphamari. All backcross lines had significantly higher no. of filled grains panicle⁻¹ (176-229) than both parent but this result was not found similar at Rangpur and Nilphamari (Table 2) as cold temperature prevail more at Northern part than Mymensingh (Table 4) which hinder grain filling properly by incomplete panicle exertion, spikelet abortion along with late and incomplete grain (Satake and Hayase, 1970). Although yield, the most important and complex traits in rice is regulated by QTLs and influenced by external environmental factors (Wang *et al.*, 2012; Zeng *et al.*, 2017) three lines e.g. K7-P-2, K7-P-5 and K7-P-9 were selected from three locations as high yielding than the recipient parent.

Six lines from BC₁F₄ generation of parent, Komol-9 were also evaluated at all three locations. At Mymensingh, K9-P-17-17 line was observed short duration and plant height than other lines. Most of the backcross lines had significantly higher number of effective tillers plant⁻¹ than the recipient parent (7.8). K9-P-16-3, K9-P-16-16 and K9-P-18 exhibited longer panicle (25.4 cm, 25.8 cm, and 25.4 cm) than the both parents. The number of filled grains panicle⁻¹ was similar with both parents. K9-P-16-3 line produced higher yield comparing to the recipient parent (Table 3). At Rangpur Sub-station, all the back cross lines had taller plant height comparing to the donor parent (Table 3).

Table 3. Evaluation of BC₁F₄ generation of Komol-9 based on grain yield and yield components during boro season 2020-21 at three locations

		Duration	Plant	Effective	Panicle	Filled grain	Yield
Location	Variety/Line	(days)	height	tiller plant ⁻¹	length	panicle ⁻¹	(t ha ⁻¹)
			(cm)	(no.)	(cm)	(no.)	
	K9-P-11	155±1.02	136.2±6.45	13 ± 0.74	24.8 ± 0.41	136.8±8.96	6.9 ± 0.35
-	K9-P-12	149 ± 1.02	117.8 ± 6.45	8.8 ± 0.74	23.2 ± 0.41	166.4±8.96	5.7 ± 0.35
$\frac{1}{2}$	K9-P-16-3	151±1.02	145.6±6.45	6.8 ± 0.74	25.4 ± 0.41	163.8±8.96	7.4 ± 0.35
me	K9-P-16-16	153±1.02	140.2±6.45	8.8 ± 0.74	25.8 ± 0.41	183±8.96	6.4 ± 0.35
nsi	K9-P-17-17	146±1.02	91.2±6.45	10.8 ± 0.74	21.4 ± 0.41	153.4±8.96	4.0 ± 0.35
Mymensingh	K9-P-18	156±1.02	145.6±6.45	6.2 ± 0.74	25.4 ± 0.41	101±8.96	4.8 ± 0.35
b	K9 (P)	155±1.02	135.4±6.45	7.8 ± 0.74	24.6 ± 0.41	183.8±8.96	5.1±0.35
	Binadhan-17 (P)	158±1.02	98.4 ± 6.45	13.8 ± 0.74	23.8 ± 0.41	156.8±8.96	7.2 ± 0.35
	K9-P-11	155±0.32	104.8±5.98	13.8±0.61	22.1±0.34	155.2±12.29	9.7 ± 0.53
Rangpur	K9-P-12	157 ± 0.32	138.4 ± 5.98	12.2 ± 0.61	24.8 ± 0.34	145.4±12.29	9.8 ± 0.53
	K9-P-16-3	157 ± 0.32	111.5±5.98	13.2 ± 0.61	24.4 ± 0.34	152.8±12.29	8.3 ± 0.53
	K9-P-16-16	156 ± 0.32	126±5.98	13.2 ± 0.61	25 ± 0.34	190.8±12.29	10.4 ± 0.53
	K9-P-17-17	156 ± 0.32	108 ± 5.98	15.2 ± 0.61	24.6 ± 0.34	92±12.29	8.7 ± 0.53
Ħ	K9-P-18	157 ± 0.32	141.2±5.98	11 ± 0.61	23.4 ± 0.34	107±12.29	9.3 ± 0.53
	K9 (P)	157 ± 0.32	136.4±5.98	9.8 ± 0.61	24.6 ± 0.34	185.1±12.29	5.6 ± 0.53
	Binadhan-17 (P)	158 ± 0.32	98.4 ± 5.98	13.8 ± 0.61	23.6 ± 0.34	167.1±12.29	8.1 ± 0.53
	K9-P-11	158 ±0.38	97±5.74	13.4±0.55	23.7±0.39	163 ±11.43	7.7±0.45
Nilphamari	K9-P-12	159 ± 0.38	112.6±5.74	11 ± 0.55	22.7±0.39	110.6±11.43	9.4 ± 0.45
	K9-P-16-3	160 ± 0.38	111±5.74	12.2 ± 0.55	24.7±0.39	180.4±11.43	9.2 ± 0.45
	K9-P-16-16	159 ± 0.38	121.8±5.74	12.4 ± 0.55	22.0±0.39	148.4±11.43	8.5 ± 0.45
	K9-P-17-17	159±0.38	104.2 ± 5.74	14.6 ± 0.55	22.8 ± 0.39	94±11.43	9.3 ± 0.45
ıarı	K9-P-18	160 ± 0.38	139.6±5.74	11.6±0.55	21.7±0.39	148±11.43	8.6 ± 0.45
•	K9 (P)	157±0.38	136.4±5.74	9.8 ± 0.55	24.6±0.39	185.1±11.43	5.5 ± 0.45
	Binadhan-17 (P)	160 ± 0.38	98.4 ± 5.74	13.8 ± 0.55	23.6±0.39	167.1±11.43	7.0 ± 0.45

All had higher number of effective tillers than the recipient parent (9.8). RM-K9-P-16-16 had longer panicle (25cm) and maximum filled grain panicle (190.8) hence yield was highest among the lines. All backcross lines showed higher yield than the both parents. Although scientists have tried to explore the mechanism of cold tolerance in rice for a long time, its genetic mechanism is still not well understood. Both additive and non-additive gene interaction are known to be present for controlling the attributes that directly or indirectly influence on yield and cold susceptibility (Chen *et al.*, 2006).

At Nilphamari, K9-P-11 had shorter plant height (97 cm), K9-P-17-17 had higher no. of effective tiller (14.6), K9-P-11, K9-P-16-3 had longer panicle length (23.7cm and 24.7 cm) comparing to the both parent (Table 3). All six lines were showed higher yield than the both parents among them K9-P-12, K9-P-16-3, K9-P-16-16, K9-P-17-17 and K9-P-18 produced higher yield comparing to the recipient parent. Molecular analysis revealed that all selected 18 lines responded positively with the survey of two cold responsive genes ICE and DREB (Fig.1).

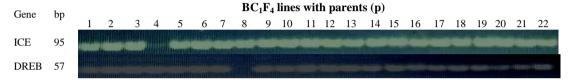


Fig 1: Two cold responsive genes (ICE and DREB) survey among 22 genotypes: Lane = Monjusree-2 (P); Lane2=Komol-7 (P); Lane3=Komol-9 (P); Lane4 = Binadhan-17 (P); Lane5-11= Monjusree-2 (BC₁F₄); Lane12-16 = Komol-7 (BC₁F₄); and Lane17-22 = Komol-9 (BC₁F₄)

Table 4. Monthly average temperature (°C) of three experimental districts

District	Temperature	Month							
District	(⁰ C)	Oct-20	Nov-20	Dec-20	Jan-21	Feb-21	March-21	April-21	May-21
Mymensingh	Day	31	28	25.8	29	32	35	35	35
	Night	24	17	13.5	17	19	22	25	26
Rangpur	Day	33.1	29.5	25.1	22.7	24.1	31	33.3	31.8
	Night	24.8	17.9	13.7	12. 2	14	19	22.3	23.9
Nilphamari	Day	30.5	32.2	30	25.1	21.8	24.1	28.6	32
	Night	22.7	21.2	15	13	11.2	12.6	16.6	23.7

Source: Weather office, Mymensingh, Rangpur and Nilphamari

The day night temperature was different in Mymensingh region and the average night temperature was higher comparing to the northern part of Bangladesh (late Dec. to early March). However, the mean minimum temperature in Northern part at reproductive stage was recorded <20 °C (Table 4) in mid-February to mid-March that enhance spikelet sterility if below 20 °C prevails 5-6 days (Biswas *et al.*, 2011). The reported genotypes could escape the cold injury due to having cold tolerance gene (Rahman *et al.*, 2020) which was supported by molecular analysis (Fig. 1). So, the Selected 15 lines [6 (Monjushree)+3(K7)+6(K9)] from Rangpur and Nilphamari would be the good material as cold-tolerant variety for North area of Bangladesh through further trials.

Conclusion

From the different sets of backcross population of BC₁F₄ generations 15 lines were acclimatized with low temperature without compromise the yield than their both parents. At all three locations, nine lines e.g. three (RM-M2-P-5, RM-M2-P-8, RM-M2-P-16) from Manjusree-2, three (RM-K7-P-2, RM-K7-P-5, RM-K7-P-9) from Komol-7 and three (RM-K9-P-11, RM-K9-P-12, RM-K9-P-16-3) from Komol-9 were performed better based on yield and yield contributing traits. Crop duration of the selected lines was 144-160 days that might be fitted at Northern cold prone area of Bangladesh and lodging tolerant gene might be involved these lines as lodging was not observed during the experimental period though the plant height is taller.

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