Indian J. Genet., 68(4): 426-434 (2008)

Studies on environmental conditions and pollination management in hybrid rice seed production

Bui Viet Thu[1], S. K. Chakrabarty, S. P. Sharma and M. Dadlani

Indian Agricultural Research Institute, New Delhi 110 012
[1]Hybreed Rice Research and Development Centre, Hw Khauh-Honi Duc, HaTay, Vietnam

(Received: June 2008; Revision: December 2008; Accepted: December 2008)

Abstract

In order to understand the effect of environmental conditions in development of hybrid seed production technology of Pusa Rice Hybrid 10 (PRH-10), an experiment was conducted in the Indian Agricultural Research Institute, New Delhi during 2005. The results indicated that in hybrid seed production of PRH-10, application of GA_3 @ 90g/ha and two time supplementary pollinations at 10:30 hrs and 11:30 hrs in a favourable condition during flowering period would give the highest hybrid seed yield upto 4.2 tons/ha. Most favourable ecological conditions for maximizing hybrid rice seed production were temperature ranging from 18.6-34.5 ^{o}C, relative humidity ranging from 34-91%, mean sunshine as 9.5 h/day and mean wind velocity of 4.1 km/hr during the flowering period. Higher hybrid seed germination and seedling vigour were recorded in seeds produced under favourable condition than that under unfavourable condition. Incidence of seed borne fungi was higher in seed lots with higher dose of GA_3 application and in seeds produced under unfavourable growing conditions.

Key words: Environmental parameters, seed set, seed yield, germination, seedling vigour, seed health, hybrid seed production

Introduction

Hybrid technology in rice with an yield advantage of about 15-20% over the best commercial varieties has been recognized as a promising technology for enhancing rice production. Indian Agricultural Research Institute has released Pusa Rice Hybrid 10 (PRH-10), the first ever hybrid in aromatic fine grain rice in 2001. PRH-10 was developed by crossing the CMS line, Pusa 6A with restorer line, PRR-78 (Basmati type) maturing in 115-120 days. Large-scale adoption and cultivation of PRH-10 is likely to bring a revolutionary change in the fine grain rice production. One of the limitations of hybrid rice is low production of F_1 hybrid seed per unit area leading to high seed cost. It is influenced by factors such as characteristics of parental lines, ecological conditions, package of agronomic practices etc. A major constraint in achieving high hybrid seed production is incomplete panicle exsertion of the female parental lines currently being used including Pusa 6A. GA_3 has been widely used for enhancing panicle exertion of CMS lines.

Hybrid rice seed production being a complex activity requires understanding of various factors which contribute maximum hybrid seed yield *viz*., time of towards flowering, GA_3 application, supplementary pollination and their combined effect with environmental conditions such as temperature and relative humidity, and the genetic background of the parental line. Therefore, a study was undertaken with the objective to compare and understand the effect of environmental factors at flowering time on i) flowering characteristics ii) seed set, yield and its quality in seed parent of PRH-10 under two different conditions.

Materials and method

The experimental material comprised of parental lines of Pusa rice hybrid-10 (PRH-10) i.e., Pusa 6A (A line) and PRR-78 (R line). Experiments were conducted at two locations in the fields of Indian Agricultural Research Institute, New Delhi during *Kharif*, 2005-2006. These two locations are designated as site I and site II. The weather conditions in these two sites were different because of differential sowing and transplanting dates of the parental lines for hybrid rice seed production (Table 1). Sowing of seeds and transplanting were done as follows:

(i) early sowing of male line by seven days in first fortnight of June and transplanting on 14th July at site I, and

(ii) late sowing of male line in splits of 3 and 6 days after female line sowing during June 30 to July 6

and transplanting both male and female lines simultaneously on 27th July at site II.

27-30 days old seedlings of female line and 21-37 days old seedlings of male line were transplanted on the same day in the field. The seedlings were transplanted with a plot size of 3.5m x 1.75m following row ratio of 2:8 (R:A). The recommended agronomic practices were followed to raise a good crop.

GA_3 was sprayed @90 and 180 g/ha on two consecutive days in the proportion of 2:3 i.e. 40 and 60% of the total dose. It was sprayed at 5% heading stage of A line plants on plot basis. GA_3 solution was prepared by first dissolving it in 70% ethanol @ 1g of GA_3 1g of GA_3/20ml. To this required quantity of water was added before spraying. To avoid losses due to drift, spraying was done in the late afternoon when there was a relatively calm weather with low wind velocity.

Supplementary pollination through pulling a rope of about 5m was done during anthesis at different times of the day (10.30 am only, 10.30 and 11.30 am and 11.30 am only). It was continued for a week.

Observations

Effective Accumulated Temperature (EAT)

For estimation of EAT, 12°C was taken as the lower limit of temperature and 30°C as the optimum temperature [1]. The following formula was used to calculate the EAT:

$$EAT = \Sigma (T-H-L)$$

Where,

T = Mean daily temperature from date of sowing to flowering

H = Temperature above upper limit (T-30°C)

L = Temperature of lower limit (12°C)

Daily weather data obtained from Division of Agricultural Physics, Indian Agricultural Research Institute, New Delhi was used for estimation of EAT.

Air borne pollen load estimation

Four cross sticks were fixed in the first row to 4th row of A line in each treatment. In early morning two glass slides coated with glycerin on one side was fixed to each cross stick facing east and west with the help of cellophane tape. This operation was completed before 9 am on the day of pollination. Rope pulling was done following the treatment to facilitate pollen movement from R line to A line. Slides were collected carefully after the pollination treatment was over in tray and pollens were counted with the help of a compound microscope on the same day in laboratory. A drop of acetocarmine (1%) was applied on the glycerin side of the slide and a cover slip was put on it. The number of pollen grains under cover slip was counted under the microscope (an area of one square centimeter).

Pollen deposited on the stigma

Ten to fifteen spikelets opened in the early morning were randomly selected and marked in each plot. Stigma was carefully removed immediately after pollination treatment on that day. Stigmas were fixed on the slide with acetocarmin dye. Number of pollen grain deposited on the bifid stigma were observed and counted under compound microscope.

Spikelet opening time and duration

Ten to fifteen spikelets of both A and R line to be opened on a day were selected and marked in each plot in the morning and the time of spikelet opening and closing was recorded for each spkelet randomly in the whole plot for 4 days.

Panicle exsertion

The panicle exsertion was calculated as the proportion of spikelet emerged out of the leaf sheath.

Stigma exsertion

The stigmas generally protrude outside the lemma and palea after the spikelet opens. It remains outside irrespective of pollination and fertilization. The stigma exsertion was calculated as (number of spikelets with stigma exerted (visible)/ total number of spikelets per panicle) x 100.

Percent seed set

Three panicles comprising of a main tiller and two side tillers in each row and in each plot were randomly harvested. The numbers of filled and unfilled spiklets were counted separately for each panicle and percent seed set was calculated as {number of filled spikelets (grain) per panicle/total number of spikelets per panicle (filled + unfilled)} x 100.

Seed yield

Harvesting and threshing of each row of a plot was done separately. Seeds were cleaned and weighed in grams to express the seed yield/row. Due to non-synchrony of

flowering between the parental lines and highly unfavorable weather condition in site I, seed yield/plot was not recorded. However, percent seed set of the labeled plants of seed parent, which synchronized with the flowering of pollen parent plants were recorded.

Seed quality

Germination of cleaned and dried seeds were tested in four replications of 100 seeds each following between paper (BP) method [2]. Germination testing was done one-month after harvest and then at six months after harvest. The hybrid seeds were kept in cloth bags and stored at ambient condition. Incidence of seed borne pathogens was recorded on freshly harvested seeds and seeds after six months of storage following Blotter test method. After the incubation, plates were examined under a stereo binocular microscope for the detection and identification of fungi.

Results and discussion

Flowering time

There was non synchrony of flowering when the male line was planted early. The female line flowered 5 days earlier than male line and the mean seed set was only 18.9 percent. However, when the male line was planted later than female in 2 splits 3-6 day later, there was most optimum synchrony of flowering of parental lines i.e. the female line flowered 1-3 days before the male parent leading to very high hybrid seed set of 49.6 percent (Table 1).

At locations where fluctuations in temperature are very frequent, the difference in days to flowering is not very effective in determining the seeding interval. Under these circumstances, the Effective Accumulated Temperature (EAT) method can be conveniently used.

Table 1. Effect of temperature on flowering time

Items	Site I		Site II		
	6A	R	6A	R1*	R2*
Flowering date	11 Sept.	6 Sept.	29 Sept.	30 Sept.	2 Oct.
Duration from sowing to flowering (day)	89	91	91	89	88
EAT from sowing to transplanting (T^0C)	441.8	500	442.5	392.0	339
EAT from sowing to flowering (T^0C)	1434.9	1410.2	1480.0	1463	1439

*: First and second set of R line sown

Viraktamath and Ramesha [1] observed that every genotype requires a specific quantum of temperature to reach the flowering stage, which generally remains relatively stable irrespective of time of seeding. Pandey [3] reported that EAT required for flowering was reduced in parental lines as sowing was delayed. The results of present study confirmed with previous studies that EAT in early sowing was higher than later sowing. Interestingly EAT of female line was higher than male line by about 17-26^{0}C. It was equal to 1-2 days accumulated heat for different time of sowing. The results also indicated that the response of parental lines to temperature was not obvious as these took almost same number of days to flower irrespective of sowing time and seedling age (Table 2). However, using the estimated EAT it can be inferred that to achieve synchronization of flowering between male and female line for seed production of Pusa RH-10 the female line should be sown 2-3 days earlier than the male line, when seed sowing is done in the first week of July and transplanting in the last week of July.

Panicle and stigma exsertion

Poor panicle exsertion in the Wild Abortive (WA) type CMS line in rice is one of the main factors for low seed set in hybrid seed production. Yuan [4] suggested application of GA_3 to enhance panicle exsertion. It was observed that panicle exsertion increased from 62.4% (control) to 79.5% and 80.8% (control) to 88.8% when 180g GA_3/ha was applied and supplementary pollination was done at 10.30 and 11.30 am in site I and II respectively (Table 3). Maximum panicle exsertion was recorded in the treatment with GA_3@ 180g/ha from 76% to 88% in two growing conditions. About 10% higher panicle exsertion in site II could be due to more favorable weather condition particularly low RH, which facilitates better absorption of GA_3 in the plant. Panicle exsertion increased from 77.03 per cent in control to 84.95 per cent at GA_3@ 180g/ha as recorded by Pandey [5]. Jose [6] also reported that panicle exsertion in IR58025A increased from 72% in control to 86% in three time application of GA_3 @180g/ha.

GA_3 application significantly increased percent stigma exsertion in two conditions (28.4 to 44.3% in site I and 54.4 to 70.2% in site II) (Table 3). In weather condition such as no rain, low RH and good sunshine hours, the percent stigma exsertion would be higher than in unfavorable condition. A comparison of two conditions on stigma exsertion showed that in favorable weather condition it increased to almost double than in control plot and more than 60% when 180g GA_3/ha was

Table 2. Temperature and relative humidity during flowering period of parental lines of PRH-10

	Site I								Site II							
					Temp. (°C) at		RH(%) at						Temp. (°C) at		RH(%) at	
Date/line	SO	SC	Dura-tion	MOS	SO	SC	SO	SC	Date/line	SO	SC	Dura-tion	MOS	SO	SC	SO	SC
12 Sept.									26 Sept.								
Female	9:20-9:25	11:40	140	10:15-10:30	34	36	71	NR	Female	10:10-10:15	12:35	135	10:30	33	33.5	57	56
Male	10:05	NR	NR	>10.35	NR	NR	71	NR									
13 Sept.									Male	11:05	NR	NR	11:45	33	33	56	56
Female	9:40-9:45	12:00	140	10:45	32.7	35	77	71	1 Oct.								
Male	10:45	NR	NR	> 11.15	NR	NR	NR	NR	Female	9:40-9:45	11:55	130	11:05	32	33	51	50
14 Sept.									Male	10:45	NR	NR	11:30	33	33	50	50
Female	9:45-9:50	12:15	150	10:00	31	33.5	75	75	2 Oct.								
Male	10:30	NR	NR	11:05	31	33.5	75	75	Female	9:25-9:30	11:35	130	10:10	33.8	35	50	50
15 Sept.	9:35-9:40	12:00	145	11:00	29.5	29.0	72	86									
									Male	10:30	NR	NR	11:15	33.8	35	50	50
									3 Oct.								
									Female	10:10-10:15	12:30	135	10:40	30	32.5	51	47
									Male	10:45	NR	NR	-	32	33	48	47
									4 Oct.								
									Female	9:45-9:50	12	130	10:40	33	NR	44	NR
									Male	11:20	NR	NR	12:05	34	NR	43	NR

SO: spikelet opening; SC: spikelet closing ; MOS: maximum opening of spiklets ; NR: not recorded; *: a rainy day

applied and supplementary pollination was done at 10.30 and 11.30 am (Table 3). The results also indicated that in a favorable weather conditions the higher GA_3 dose (180g/ha) had an advantage for stigma exsertion in comparison to a lower dose (90g/ha). However, this difference was not very consistent in case of unfavourable weather conditions.

Pollen production, availability and deposition on stigma

In addition to synchrony of flowering of parental lines pollen production on male parent and its transfer to female (male sterile) line is important for increasing hybrid seed production. For this purpose supplementary pollination is practiced usually by rope pulling in which plants of male line are shaken towards female parent. Time of anthesis in the male and spiekelet opening time in the female determine the time and frequency of supplementary pollination.

The temperature at spikelet opening time ranged from 29.5-34°C in two experimental sites. Spikelet opening times in the female and male lines ranged from 9:35 to 10:15 hrs and 10:05 to 11:20 hrs, respectively (Table 2). Spikelets of male line always opened later than that in female line under similar condition. There was a clear difference in relative humidity at the two sites at the time of spikelet opening and closing. However, it had no apparent effect on spikelet opening time and duration. This indicated that spikelet opening is primarily dependent on the genetic make up of the line and not on weather conditions.

Higher RH and rainfall during flowering results in poor pollen flow from the male to female parent and thus leading to the low yield level as reported by Pandey [3]. A lower RH of 50-70 per cent and non rainy weather during flowering at site II was found more favourable for pollen dispersal and seed set. After supplementary pollination air borne pollen load was highest in the first row from west direction than the other rows in both the conditions. Number of pollens/cm^2 reduced from first row to fourth row (267 to 224 in site I and 380 to 283 in site II) (Table 4). Sharma and Virmani [7] showed a decrease in pollen load at 15 cm distance from male rows. Airborne pollen/liter dropped consistently with increasing distance from the pollen source. Namai and Kato [8] reported that the pollen load of more than 10 airborne pollens/liter was necessary for an outcrossing rate of more than 25 per cent at a wind velocity of 3 to 4

Table 3. Percent panicle and stigma exsertion in female parent (Pusa 6A) of P RH-10

Treatment	Site I		Site II	
	Panicle exsertion (%)	Stigma exsertion (%)	Panicle exsertion (%)	Stigma exsertion (%)
T1	73.9 (59.29)	38.4 (38.27)	82.3 (65.12)	58.9 (50.13)
T2	75.9 (60.62)	41.3 (39.97)	86.6 (68.60)	66.5 (54.67)
T3	77.7 (61.94)	41.3 (39.97)	86.8 (68.78)	66.5 (54.67)
T4	79.5 (63.08)	44.3 (41.74)	88.8 (70.47)	70.2 (56.94)
T5	74.8 (59.89)	37.6 (37.81)	81.7 (64.69)	58.3 (49.82)
T6	76.2 (60.78)	40.7 (39.60)	86.3 (68.32)	64.9 (53.69)
T7	62.4 (52.20)	28.4 (32.22)	80.8 (64.04)	54.4 (47.56)
Mean	73.9 (59.69)	38.8 (38.51)	84.7 (67.15)	62.8 (52.35)
CD(p=0.05)	4.9 (3.32)	4.68 (2.73)	4.37 (3.5)	5.55 (3.31)

Transformed values are in parenthesis
Treatments:
T1: rope pulling at 10.30 am+ spray of GA_3 @90g per ha
T2: rope pulling at 10.30am + spray of GA_3 @180g per ha
T3: rope pulling at 10.30am +11.30am+ spray of GA_3 @90g per ha
T4: rope pulling at 10.30am +11.30am + spray of GA_3 @180g per ha
T5: rope pulling at 11.30am + spray of GA_3 @90g per ha
T6: rope pulling at 11.30am + spray of GA_3 @180g per ha
T7: Control (no GA_3 spray and no rope pulling)

km/h under tropical conditions and 1.5 or more airborne pollen grains per liter and a wind velocity of 2-3 m/second were necessary to obtain more than 50% seed set. Murlidhar [9] showed that maximum pollen availability was in first row of east (33/cm^2) and west (25/cm^2) direction, nearest to the pollen sources and the lowest in sixth row in the east (15/cm^2) and west direction (12/cm^2) which were farthest from the pollen source.

In the present study average numbers of pollen in site II (331) were higher than that in site I (240) (Table 4). It was observed that lower RH, sunny days and no rains during flowering period favoured higher dehiscence of anthers and more pollen availability for effective pollination.

The supplementary pollination treatments showed significant effects on the available pollen number. In the site I, the pollen load was higher in two times pollination as compared to a single time pollination and also between GA_3 doses. If supplementary pollination is done only once, either at 10.30 or 11.30 am higher dose of GA_3 from 90-180 g/ha did not show any significant increase in pollen availability/ stigma under both favourable and unfavourable conditions during flowering and pollination .

Number of stigma getting at least 1 pollen is very important because it leads to fertilization and seed set. Percent spikelet getting more number of pollen assures fertilization and higher seed yield. Maximum percentage of stigma with at least one pollen obtained at two times rope pulling were 87.5% and 85.2% in both the sites. Rope pulling, in general, has increased the possibility for stigma to receive pollens. It ranged from 59.6% to 87.5% compared to control (27.8% in both conditions) (Table 5). Interestingly the number of pollen/stigma and percent stigma with at least one pollen did not differ in two sites. However, higher number of pollen (up to 4-5) /stigma was observed at site II.

Namai and Kato [10] showed that spikelets set seed when at least one pollen grain was deposited and 3-4 pollen grains were almost always sufficient. Thirty minutes or more were required to allow 80% of spikelets to acquire 2 or more pollen grains after spikelet opening. The results of the present study also showed that the highest number of pollen grains observed were 1.53 and 1.91 in site I and II respectively when 180g GA_3/ha was applied and supplementary pollination was done at 10.30 and 11.30 am (Table 5) This indicated that supplementary pollination through rope pulling at least twice is essential for pollen movement to the stigma of seed parent at peak flowering of pollen parent.

Seed set and yield

The results showed that application of GA_3 @ 90 and 180g/ha had no significant difference in seed yield. Rope pulling at different times showed significant effect on seed set percentage and seed yield. In one time rope pulling seed yield ranged from 3.3-3.5 ton/ha. The pollination for two times gave the highest seed yield (4.2 ton/ha) in both the GA_3 doses (Table 6). The advantage of the second supplementary pollination in hybrid seed yield was to the extent of 0.7 to 0.9 ton/ha. The results showed that seed set ranged from 16.4 to 24.4% and 42.2 to 58.4% in the two conditions. The highest seed set (58.73%) was obtained with application

Table 4. Air borne pollen load in hybrid seed production plot

Treatment/row	Site I					Site II				
	E1	E2	E3	E4	Mean	E1	E2	E3	E4	Mean
T1	214.3	148.3	176.0	215.0	**188.4**	214.3	148.3	237.7	220.7	**272.3**
T2	207.0	161.0	221.0	242.0	**207.8**	207.0	161.0	250.3	219.0	**260.8**
T3	489.3	463.7	386.0	355.3	**423.6**	489.3	463.7	447.3	402.0	**461.6**
T4	403.7	351.7	460.0	322.7	**384.5**	403.7	351.7	505.0	444.0	**523.5**
T5	209.3	200.3	191.7	191.7	**198.3**	209.3	200.3	248.0	240.0	**278.3**
T6	253.0	180.0	163.0	183.0	**194.8**	253.0	180.0	252.0	262.0	**305.6**
T7	95.30	100.0	80.7	61.7	**84.4**	95.3	100.0	203.0	193.3	**219.2**
Mean	267.4	229.3	239.8	224.5	240.3	267.4	229.3	306.2	283.0	331.6

CD (p=0.05); Row :10.1; treatment: 13.3; row x treatment: 26.7; Row :21.1; treatment: 27.9; row x treatment: 55.9
E 1, 2, 3 and 4: 1st, , 2nd, 3rd and 4th row from east side of the pollen parent; Treatments (T1 to T7): As in Table 3

Table 5. Pollen deposited on stigma and per cent stigma with at least 1 pollen

Treatment	Site I			Site II		
	Pollen/stigma		% Stigma with at least 1 pollen	Pollen/stigma		% Stigma with at least 1 pollen
	Mean	Range		Mean	Range	
T1	0.94	0-4	59.6(50.62)	1.41	0-3	76.4(60.97)
T2	1.11	0-3	68.4(55.86)	1.38	0-4	68.0(55.65)
T3	1.43	0-4	85.2(71.50)	1.73	0-5	80.4(63.83)
T4	1.53	0-5	79.7(64.17)	1.91	0-5	87.5(73.10)
T5	1.23	0-3	72.2(58.44)	1.26	0-4	73.2(58.83)
T6	1.23	0-3	61.2(51.48)	1.10	0-3	66.7(54.79)
T7	0.37	0-2	27.8(31.24)	0.28	0-2	27.8(31.24)
Mean	1.25	0-5	64.8(54.82)	1.30	0-5	68.57(56.91)
CD(p=0.05)	0.24	-	17.52(14.43)	0.28	-	14.6 (13.28)

Transformed values are in parenthesis; Treatments (T1 to T7): As in Table 3

Table 6. Per cent seed set and hybrid seed yield of PRH-10

Treatment	Site I	Site II	
	Seed set (%)	Seed set (%)	Seed yield (t/ha)
T1	18.8(25.55)	42.2(40.53)	3.330
T2	21.5 (27.60)	50.6(45.36)	3.300
T3	22.6(28.33)	54.8(47.63)	4.220
T4	24.4(29.59)	58.7(49.82)	4.220
T5	16.4(23.60)	45.7(42.55)	3.567
T6	20.8(26.90)	51.9(46.09)	3.500
T7	8.1(16.53)	35.6(36.648)	2.567
Mean	18.9(25.44)	49.6(44.09)	3.524
CD (P=0.05)	6.87(5.19)	9.24 (3.5)	0.787

Transformed values are in parenthesis; Treatments (T1 to T7): As in Table 3

of GA_3 @180 g/ha and two times rope pulling in site II where good weather condition prevailed during flowering period This indicated that higher GA_3 dose increased the available pollen for fertilization. The higher seed set at site II was also because of better synchronization of flowering between parental lines and rain free, sunny and dry weather conditions. However, percent seed set in present study was not significantly different between 180g and 90 g GA_3/ha. But it was significantly different between one time and two times rope pulling. The seed set recorded by earlier workers were from the panicle in main tiller, which got maximum benefit of GA_3 application leading to higher panicle and stigma exsertion (Personal communication). In our study it is an average value of 3 panicles per plant/hill belonging three different orders i.e., main, side and last tiller. This gives an average value of seed set in a single plant/hill. Yak [11] reported seed set in female parent up to 65 %

following Gibberellin applied (3 or 4 times instead of once or twice) in a larger amount (150 g/ha) and at a higher concentration (40-60 ppm) at early heading.

Jagadeeswari *et al.* [12] reported a yield level 2.7-3.1 t/ha of CMS Line, IR 58025A seed with application of GA_3 @ 90-135g/ha. An average yield of 2.7t/ha of IR58025A line with GA_3 @ 135-180 g/ha in 1:2:1 ratio applied on 1st, 3rd and 5th day after panicle emergence was reported by Jose *et al.* [13]. The results of the present study also showed that use of GA_3 @ 90g/ha and 180g/ha with two times pollination gave the highest hybrid seed yield (4.2 ton/ha). The higher seed yield on the Pusa 6A was due to higher panicle, stigma exsertion and percent seed set in a highly favourable weather condition, beside a perfect synchronization of flowering of parental lines. The high yield could also be attributed to the inherent genetic superiority in Pusa 6A compared to IR 58025A.

Effect of environmental factors on seed yield

Environmental factors influencing natural outcrossing in rice include temperature, relative humidity, light intensity, wind velocity and its direction. Mao *et al.* [14] reported that the seed set percent and seed yield of CMS lines are negatively correlated to relative humidity. They further suggested that higher seed yield could be obtained, if the seed and pollen parent flower when the relative humidity is 50-60 per cent, maximum/minimum temperature is 28-30/21-22°C and wind velocity is above 2.5 m/sec. In China, the favourable weather conditions identified for higher outcrossing rate were a daily temperature of 24-28°C, relative humidity of 70-80%, diurnal difference in temperature of 8-10°C and sunny days with a breeze as observed by Xu and Li [15]. They also reported that the spikelet opening percentage is reduced when temperature is high, relative humidity is low or when temperature is low and RH is high. Pollen viability and stigma receptivity also reduced under such conditions.

In the present study, seed set percent in site I was low which resulted to low seed yield where as both seed set and seed yield in site II were high. In site II mean wind velocity was 4.1 km/hr, while in site I it was recorded 8.5 k/hr (Table 7). Mean R.H was 82 per cent in site I, whereas in site II it was 62 per cent. Therefore, seed yield in site I was low due to high wind velocity, rainfall, and high R.H in addition to non-synchronization of flowering of parental lines resulting in poor pollen dispersal from male to female parent. But in site II, no rain, 62 per cent R.H., ample sunshine hours (9.5hrs/day), and a wind velocity of 4.1km/hrs were conducive for flowering and pollination. An adverse effect of high R.H. during flowering on seed yield was reported by Pandey [8]. He obtained 0.2-0.5 t/ha higher yield in *kharif,* 1997 (66% mean R.H. during flowering) compared to that of *kharif,* 1999 (80% mean R.H during flowering). Jagadeeswari *et al.* [12] also reported that seed yield during *kharif,* 2000 (mean R.H 71 % and mean wind speed 4.6 km/hr) was higher by 500-600kg/ha than that in *kharif* 1999 with higher mean R.H (80%) and calm wind during flowering in the morning hours which did not favour pollen dispersal from pollinator to seed parent. The seed set on seed parent line depends upon prevailing weather condition besides synchronization of flowering and period of anthesis [16].

Table7. Meteorological observation during flowering period

Weather parameters	Site I	Site II
Date of transplanting	14 July, 2005	27 July, 2005
Date of flowering (A line)	11 Sept., 2005	29 Sept., 2005
(R line)	6 Sept., 2005	30 Sept. & 2 Oct., 2005
Mean max. temperature (°C)	31.9	33.5
Mean min. temperature (°C)	23.7	20.0
Mean temperature (°C)	27.8	26.8
Temperature (°C): range	20.6-35	18.6-34.5
Mean relative Humidity (%)	81.9	62.3
Relative humidity (%):range	50-100	34-91
Total Rainfall (mm)	159.2	0
No. of rainy days (>20mm)	3	0
Mean sunshine (hours/day)	3.5	9.5
Mean wind velocity (km/hr)	8.5	4.1

Seed germination

Rice seeds are good storer, but hybrid rice seeds have poor storability due to the opening of glumes [17, 18]. Duan and Ma [19] observed that higher concentration of GA_3 applied to the mother plant @150 g/ha or more had adverse effect on germination and storability of seeds. Studies on storability of CMS lines seed carried out at Delhi reported that germination remain above 90% even after 6 months of storage [5, 13].

In the present study initial germination of hybrid seed after harvest ranged between 84.3 to 94.3%. After 6 months of storage of seeds the germination ranged

Table 8. Germination of freshly harvested and stored hybrid seed produced at two conditions/sites

Treatment	Germination (%)			
	After harvest		After 6 months of storage	
	Site I	Site II	Site I	Site II
GA_3 spray @ 90g /ha	87.8	94.3	88.7	97.7
GA_3 spray @180g /ha	87.3	92.0	88.3	96.7
Control	84.3	92.7	85.3	97.3
Mean	86.5	93.0	87.4	97.2
CD (p=0.05)	5.6		4.0	

from 85.3 to 97.7% (Table 8). Seed lots produced under favourable conditions showed significantly higher percent germination than that produced under unfavourable conditions both after harvest and after 6 months of storage. In the same production condition GA_3 doses did not show significant effect on germination percentage up to 6 months storage. The results showed that all seed lots retained germination above 85% up to 6 months after harvest. Thus, production conditions during the period of flowering and fertilization have a significant role on seed germination and vigour.

Seed health

Seed discoloration is a serious problem in hybrid rice caused by several fungi, many of which are seed borne. Zhou *et al.* [18] observed that the seed of the CMS line are borne in open glumes to allow out crossing and also the chances of external infection. In China and north western India, CMS lines in hybrid rice seed production plots show higher incidence of seed borne diseases such as paddy bunt, false smut etc. compared to those in pollen parent plots. This can cause a serious outbreak of the disease on commercial crops of hybrid rice. Huynh [20] reported that the most predominant seed borne fungi encountered consistently with the hybrid seed lots were *Bipolaris oryzae, Altrenaria padwickii, Curvularia lunata*. Hybrid rice seed produced in good weather condition had lower seed borne fungi infection compared to those produced under unfavorable conditions. Huynh [20] recommended treatment of hybrid rice seed with Vitavax 200 to control the incidence of these fungi.

In the present study seven species of fungi i.e., *Alternaria alternata, Curvularia lunata, Cladosporium ssp*, *Bipolaris oryzae*, *Trichoconiella padwickii*, *Fusarium moiniliforme*, *Fusarium semitectum* were identified. Higher incidence of fungi was recorded after harvest and 6 months of storage in case of seeds produced under unfavorable condition. Maximum incidence of seed borne fungi *Alternaria* ssp. and *Curvularia* ssp. were recorded.

In conclusion the most favourable ecological conditions identified for maximizing hybrid rice seed production were temperature ranging from 18.6-34.5^{0}C, relative humidity ranging from 34-91%, mean sunshine as 9.5 hr/day and mean wind velocity as 4.1 km/hr during the flowering period. The results indicated that in hybrid seed production of PRH-10, application of GA_3 @ 90g/ha and two time supplementary pollinations at 10:30 hrs and 11:30 hrs in a favourable conditions would give the highest hybrid seed yield (4.2 tons/ha). No adverse effect of GA_3 application was noted on germination of hybrid seed lots but higher germination and vigor was recorded in seeds produced under favourable condition than under unfavourable condition. The germination of hybrid seeds remained above 85% after 6 months of storage. Incidence of seed borne fungi was higher in seed lots with higher dose of GA_3 application than in seeds produced under unfavourable growing conditions.

References

1. **Viraktamath B. C. and Ramesha M. S.** 2000. Hybrid Rice Seed Production: Preliminary Consideration. *In*: Lectures presented in Summer School on advances in Hybrid Rice Seed Production Technology, IARI, New Delhi 110012, Singhal N.C. and Vari A.K. (eds). pp. 83-90.
2. **International Seed Testing Association**. 2004. International Rules for Seed Testing. Switzerland.
3. **Pandey S.** 2000. Studies on synchroniazation of flowering, seed production and storability of parental lines of hybrid rice. Ph.D. thesis submitted to Post Graduate School, Indian Agricultural Research Institute, New Delhi, India.
4. **Yuan L. P.** 1977. The execution and theory of developing hybrid rice. Zhejiang Nongye Kexue (Chinese Agricultural Science) **1**: 27-31.
5. **Pandey S., Sharma S. P. and Dadlani M.** 2003. Effect of gibberellic acid application on floral and morphological traits, seed yield and storability of parental lines in hybrid rice (*Oryza sativa*). Indian Journal of Agricultural Sciences, **73**: 376-380.
6. **Jose S.** 2003. Improvement of seed yield of CMS parent in rice by management of GA_3 application. M. Sc. thesis submitted to Post Graduate School, Indian Agricultural Research Institute, New Delhi, India.
7. **Sharma H. L. and Virmani S. S.** 1994. Optimum pollen load and male-female row ratio for hybrid rice seed production. *In*: Hybrid rice technology: new

developments and future prospects. Los Banos, Laguna (Philippines). IRRI, Virmani S.S. (ed.) p. 294.

8. **Namai H. and Kato H.** 1988. Improving pollination characteristics of *japonica* rice. *In*: Hybrid rice: Proceedings of international symposium, Changsha, China, 6-10 October, 1986, pp. 165-173.
9. **Murlidhar J. S.** 2002. Studies on maximization of seed production in female line of rice hybrid. M. Sc. thesis submitted to Post Graduate School, Indian Agriculture Research Institute, New Delhi, India.
10. **Namai H. and Kato H.** 1987. The number of pollen grains deposited upon a pistil assuring seed setting of male sterile seed parent in rice (*Oryza sativa* L.). Japanese J. Breed., **37**: 98-102.
11. **Yak S.** 1998. Techniques to get high yield in hybrid rice seed production in China. *In*: International Symposium on Hybrid Rice, pp. 6-10.
12. **Jagadeeswari P., Sharma S. P. and Dadlani M.** 2004. Effect of different chemicals on traits favouring out crossing and optimization of GA_3 for seed production of cytoplasmic male sterile line in hybrid rice. Seed Sci. & Technol., **32**: 473-483.
13. **Jose S., Chakrabarty S. K., Sharma S. P. and Dadlani M.** 2006. Management of GA_3 application to enhance seed production in the CMS parent of rice (*Oryza sativa* L.) hybrids. Indian Journal of Agriculture Science, **76**: 122-124.
14. **Mao C. X., Virmani S. S. and Kumar I.** 1998. Technological innovations to lower the cost of hybrid rice seed production. *In*: Advances in Hybrid Rice Technology, Virmani S.S., Siddiq E. A. and Muralidharan K. (eds.). Proceeding of 3rd Internatonal Symposium on hybrid rice, 14-16th November, 1996, Hyderabad, India, pp. 111-128.
15. **Xu S. and Li B.** 1988. Managing hybrid rice seed production. *In*: Hybrid Rice: Proceeding of International Symposium in Hybrid Rice, Changsha (China), IRRI, Manila, Philippines, pp. 157-164.
16. **Krishnaswamy V. and Ramaswamy K. R.** 1979. Influence of time of sowing and climatic factors on the flowering duration of the parental lines of CSH-5 hybrid sorghum. Madras Agricultural Journal, **66**: 512-519.
17. **Huang S. Z. and Fu J. R.** 1987. The deterioration and physiological and biochemical changes in hybrid rice seed. Acta Phytophysiologica Sinica, **13**: 229-235.
18. **Zhou Z. Y., Tang S. D., Tang G. D. and Hu J. Y.** 1989. Effects of infection and imperfect closed-glume on germination of hybrid rice seed. International Rice Research Newsletter. **14**: 4.
19. **Duan X. M. and Ma H. S.** 1992. Effects of gibberellic acid application on seed yield and quality of hybrid rice. Seed Sci. & Technol., **20**: 209-214.
20. **Huynh Van Nghiep.** 2004. Incidence of *Bipolaris Oryzae* (Breda De Hann) shoem and its management through seed treatment in rice (*Oryza sativa* L.). M.Sc thesis submitted to Post Graduate School, Indian Agricultural Research Institute, New Delhi, India.