Himachal Journal of Agricultural Research 36 (2): 174-179, 2010

Studies on floral biology and systems of pollination on seed setting in okra (*Abelmoschus esculentus* L. Moench) under mid-hill conditions of Himachal Pradesh

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(Manuscript Received: 24.08.2010; Accepted: 10.01.2011)

Abstract

An experiment was conducted on floral biology and efficacy of different systems of pollination on seed setting in okra cv. P-8. The duration of maximum anthesis, anther dehiscence and stigma receptivity was recorded at 9.00 to 10.00 a.m. The temperature and relative humidity were found to have hastening effect on these processes. The optimum temperature and relative humidity for peak anthesis and anther dehiscence was 22.6°C and 78.90 per cent, respectively, whereas the temperatures of 22.2°C relative humidity of 79.10 per cent were optimum for maximum stigma receptivity. Temperature has pronounced influence *vis-à-vis* to relative humidity, which shows that the stigma receptivity may be prolonged under mild climate of mid hills, thus affording more time in hybridization programme for affecting crosses in okra. Maximum pollen viability was recorded on the day of anthesis in field and viability was reduced to 15.5 per cent under laboratory condition at 20-28°C room temperature after 24 hours of storage. The refrigerator conditions at 4-5°C temperature, offer good opportunity of storing pollens grains for five days with the viability of more than 25 per cent and the stored pollen grains may be utilized at favorable time to affect crossing under high rainfall area of Palampur. None of the three systems of pollination affected number of pods per plant, number of seeds per pod, 100 seed weight and seed yield per plant, indicating thus, okra is potentially a self-fertilized crop. The seed production of pureline varieties can efficiently be taken up through any of the systems of pollination. Nevertheless, the open pollination in isolation is the most economic system for seed multiplication *vis-a-vis* to controlled pollination and hand sib pollination.

Key words: Okra, *Abelmoschus esculentus* L. Moench., floral biology and pollination.

Introduction

The improvement of a crop plant, which is the ultimate objective of plant breeder is not a simple task. The information on the biological behaviour of a crop is a prerequisite in affecting any improvement in the crop. It facilitates the work of breeders engaged in crop improvement to plan their work efficiently. The pollination and seed setting studies are the pre- requisites for switching the genetic amelioration endeavour in any crop species. Knowledge of floral biology is of great importance to breeder for execution of most of the breeding programme as it helps in crossing programme. Okra, *Abelmoschus esculentus* (L.) Moench, of the family Malvaceae is one of the important summer vegetable crops of India. The floral biology has not received much attention therefore, it was thought desirable to study the different aspects of floral biology and effect of systems of pollination on seed setting.

Materials and Methods

The present investigation was carried out in okra variety P-8, which was sown on May, 2000 in spacing of 60 x 20 cm at Experimental Farm of Department of Vegetable Science and Floriculture, CSK, HPKV, Palampur. The recommended agronomical practices and plant protection measured were adopted to raise healthy crop of okra. The floral biology study was conducted on randomly selected plants in bulk population grown for study, whereas for seed setting study, each system of pollination (Controlled self pollination, Open pollination, Hand sib pollination with fresh bulk pollen) was laid out in randomized block design with three replications. The details of procedure adopted for taking observations for each character is described as under:

***Anthesis*:** To determine the time of flower opening, a number of flower buds expected to open next day were tagged at random on each day

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and time of anthesis was noted on subsequent days at hourly internal. The total numbers of flowers opened during six days of observations were obtained and percentage worked out. Meteorological data, viz., temperature and relative humidity were also recorded simultaneously with the help of thermo hygrometer at hourly internal during the period of study.

Anther dehiscence: The anther dehiscence was also recorded on the flowers, which were examined for anthesis at hourly internal, with the help of hand lens. The total number of completely dehisced anthers in selected flowers were recorded and percentage worked out. The temperature and relative humidity were also recorded.

Pollen viability and longevity: Pollen viability was tested using pollens stored at 20-28°C under room temperature and at 4-5°C temperature under controlled conditions. Pollen viability was observed in one per cent acetocarmine solution. One to two drops of acetocarmine solution were placed on the slide and the pollen grains dusted, covered with cover slip and examined under microscope. The deeply stained, normal looking pollens were counted as viable, whereas shrivelled, poorly and weekly stained as non-viable and the percentage of viable pollens was calculated.

Stigma receptivity: Time for the maximum stigma receptivity was recorded on the basis of seed set percentage as given below:

Seed set method: The time for maximum stigma receptivity was ascertained by artificially pollinating the emasculated buds with fresh pollen. The observations were taken for two days with periodical hand pollination of eighty flower buds daily. Every day from 600 to 1400 hrs, ten flower buds were pollinated at hourly interval and on maturity, the pods were harvested separately for each interval and seed set was counted.

Seed setting studies: In order to ascertain the efficacy of different systems of pollination on seed set in okra, three systems were adopted, *viz.*, controlled self-pollination, open pollination and hand sib pollination with fresh bulk pollen of same variety.

Controlled self pollination: The flower buds of ten plants likely to open next day were tagged and covered with butter paper bags and left as such for selfing.

Open pollination: Ten randomly taken plants were tagged and left as such for natural pollination by various pollinators.

Hand sib pollination with fresh bulk pollen: The well developed flower buds of ten randomly taken plants were emasculated a day prior to anthesis and covered with butter paper bags. They were pollinated on the following day with mixed pollen of other plants of P-8 variety and rebagged.

All these treatment were replicated three times and the observations were recorded on number of pods per plant, number of seeds per pod, hundred seed weight (g) and seed yield per plant (g). To assess the floral biology study, the correlation coefficient and regression analysis were worked out using the method described by Gupta (1983) and Goulden (1959), respectively. The data pertaining to seed setting studies were analysed according to randomized block design described by Panse and Sukhatme (1985).

Results and Discussion

Anthesis

The anthesis in okra commenced from 6.00 a.m. and continued upto 2.00 p.m. the majority of flower buds opened between 9.00 to 10.00 a.m. at the temperature of 22.6°C and the relative humidity of 78.90 per cent (Table 1.). It appears that temperature and relative humidity played an important role the anthesis of okra. The results have strengthened by fitting the data in multiple linear regression equation. Multiple linear regression equation for predicting anthesis (Y), when temperature (X_1) and relative humidity (X_2) are known:

$$Y = -241.71 + 4.39\ X_1 + 2.09\ X_2$$

The above multiple linear regression equation signifies that the one unit (1°C) increase in temperature led to increase in anthesis by 4.39 units. Similarly, the equation further indicates that with one per cent increase in relative humidity, there would be increase in anthesis by 2.09 units. This indicates that a unit change in both the independent variables resulted in increase of anthesis, however, temperature influenced anthesis to greater extent compared to relative humidity.

Table 1. Time of anthesis in okra (*Abelmoschus esculentus* L. Moench)

Time of observation (hrs)	Percentage of flowers opened	Cumulative percentage	Temperature °C		Relative humidity (%)
			Max.	Min.	
6-7 AM	2.71	2.71	18.9	16.1	80.5
7-8 AM	7.45	10.61	20.7	16.2	84.5
8-9 AM	20.77	30.63	24.1	18.1	83.2
9-10 AM	33.86	64.77	25.9	19.2	78.9
10-11 AM	23.93	88.72	28.7	20.7	67.5
11-12 AM	7.22	95.94	29.8	23.1	63.2
12-1 PM	2.71	98.65	30.1	24.2	63.0
1-2 PM	1.58	100.23	30.7	25.7	61.4

The correlation between per cent anthesis and temperature as well as relative humidity was non-significant; however, the correlation between temperature and relative humidity was significant (Table 2.).

Table 2. Correlation coefficient between anthesis, temperature and relative humidity

Factors considered	d.f. (n-2)	'r' value	Calculated value (t)	$t_{0.05}$
Anthesis (%) and temperature (°C)	6	-0.131	0.369	2.45
Anthesis (%) and relative humidity (%)	6	0.349	1.313	2.45
Temperature (°C) and relative humidity (%)	6	-0.932	-6.298*	2.45

• Significant at 5% level

The estimate of coefficient of multiple determination (R^2 x 100) was 40.59 per cent, which signifies that 40.59 per cent of variations in anthesis could be attributed to temperature and relative humidity.

The possible reason for the opening of maximum number of flowers during the interval (9.00-10.00AM) may be attributed to the interaction of temperature and relative humidity, a view supported by Sulikeri and Rao (1972), who reported that the temperature of 29.6°C combined with the relative humidity of 91.0 per cent seemed to be responsible for maximum percentage of anthesis. The finding of Singh and Tiwari (1977) also supports the results who substantiated that the time of anthesis varies with temperature and relative humidity of a place.

Anther dehiscence

Anther dehiscence attained its maximum at the time period of 9.00 to 10.00 a.m., when the temperature and relative humidity were 22.6°C and 78.90 per cent, respectively (Table 3.). Multiple linear regression equation to predict the anther dehiscence (Y), when the two independent variables, temperature (X_1) and relative humidity (X_2) are known:

$Y = -212.58 + 3.76\ X_1 + 1.89\ X_2$

Table 3. Anther dehiscence in okra (*Abelmoschus esculentus* L. Moench)

Sr. No.	Time of observation (hrs)	Percentage of flowers dehisced	Cumulative percentage	Temperature °C		Relative humidity (%)
				Max.	Min.	
1.	6-7 AM	4.67	4.67	18.9	16.1	80.5
2.	7-8 AM	9.19	13.86	20.7	16.2	84.5
3.	8-9 AM	20.84	34.70	24.1	18.1	83.2
4.	9-10 AM	31.57	66.27	25.9	19.2	78.9
5.	10-11 AM	20.61	86.68	28.7	20.7	67.5
6.	11-12 AM	7.55	94.43	29.8	23.1	63.2
7.	12-1 PM	4.19	98.62	30.1	24.2	63.0
8.	1-2 PM	1.36	99.98	30.7	25.7	61.4

The above multiple linear regression signifies that a unit change in both the independent variables resulted in an increase in anther dehiscence. This indicates that 1°C increase in temperature led to increase in anther dehiscence by 3.76 units. Similarly, one unit increase in relative humidity resulted in the increase of anther dehiscence by 1.89 units, however, the temperature influenced anther dehiscence to greater extent *vis-a-vis* to relative humidity.

The correlation between per cent anther dehiscence and temperature as well relative humidity was non-significant (Table 4).

The estimate of coefficient of multiple determination was 43.53 per cent, which indicates that about 43.53 per cent of changes in anther dehiscence could be attributed to the impact of these two variables.

Table 4. Correlation coefficient between anther dehiscence, temperature and relative humidity

Factors considered	d.f. (n-2)	'r' value	Calculated value (t)	$t_{0.05}$
Anther dehiscence (%) and temperature (°C)	6	-0.207	-0.518	2.45
Anther dehiscence (%) and relative humidity (%)	6	0.431	1.170	2.45
Temperature (°C) and relative humidity (%)	6	-0.932	-6.298*	2.45

• Significant at 5% level

The temperature seemed to have played an important role in the dehiscence of anthers. These findings corroborate with results of Srivastava (1964) and Ramu (1976) who reported that the peak anther dehiscence is dependent on increased temperature and low humidity. Purewal and Randhawa (1947) also reported the occurrence of maximum dehiscence on bright sunny days, when the temperature and relative humidity was high and low, respectively. Schmid and Alpert (1977) put forth a hypothesis which states that anther dehiscence in many flowers results from water loss and the withdrawal of other tissues, particularly nectarines, not by transpiration. They further suggested that the exact time of anthesis dehiscence is subject to weather conditions, age of the flower, altitude and the location of experimental field etc.

Pollen viability and longevity

Maximum pollen viability of 95.3 to 96.7 per cent was noticed in fresh pollens collected at anthesis, pollen fertility under laboratory conditions decreased consequently with passage of time and was only 15.5 per cent after 24 hours of observation, thereby indicating short duration of pollen viability under laboratory conditions (Table 5.). Under refrigerator conditions, pollens remained viable for comparatively long period of time as the rate of decline in pollen viability was less and even after storage of five days, more than 25 per cent pollens were found viable. These investigations are in line with the earlier findings of Rao and Ramu (1979) and Ramu (1976), who reported that pollens retained viability upto 3 days at 5°C temperature and 90 per cent relative humidity.

Table 5. Pollen viability and longevity in okra under laboratory and refrigerator conditions

Age (hrs)	Laboratory conditions			Refrigerator conditions		
	No. of pollen grains examined	No. of viable pollen grains	Pollen viability (%)	No. of pollen grains examined	No. of viable pollen grains	Pollen viability (%)
Fresh	127	121	95.30	182	176	96.7
4	105	54	51.4	120	91	75.8
12	122	52	42.6	129	78	60.5
14	141	54	38.3	119	60	50.4
24	116	18	15.5	111	45	40.2
5days	113	Nil	-	115	29	25.2
10days	109	Nil	-	123	4	3.25
30days	105	Nil	-	125	Nil	-

Stigma receptivity

The present investigation reveals that the maximum stigma receptivity of 90 per cent was noticed between 9.00 -10.00 a.m., when the temperature and relative humidity were 22.2°C and 79.10 per cent, respectively, giving maximum seed set (Table 6.). These observations corroborate with the findings of Ramu (1976). Singh and Tiwari (1977) and Sukhija *et al.* (1981) who observed maximum stigma receptivity on the day of anthesis. Basavaraj (2006) also observed higher seed set between 9 a.m. to 12 noon in Dharwad conditions relating to prevalence of active stigma receptivity and peak viability.

Table 6. Diurnal trends in stigma receptivity in okra (*Abelmoschus esculentus* L. Moench)

Sr. No.	Time of observation (hrs)	Number of flowers pollinated	Flowers set seed (%)	Seed set (%)	Temperature °C		Relative humidity (%)
					Max.	Min.	
1.	6-7 AM	20	12	60	18.7	15.2	79.2
2.	7-8 AM	20	14	70	21.1	16.1	84.32
3..	8-9 AM	20	15	75	23.6	18.1	84.1
4.	9-10 AM	20	18	90	25.4	19.0	79.1
5.	10-11 AM	20	15	75	28.8	19.2	68.2
6.	11-12 AM	20	13	65	28.9	22.2	65.1
7.	12-1 PM	20	11	55	29.9	23.8	64.1
8.	1-2 PM	20	10	50	30.2	25.2	61.2

Multiple linear regression equation for predicting stigma receptivity in relation to percentage seed set (Y), temperature (X_1) and relative humidity (X_2) is given below:

$Y = -116.28 + 2.52 X_1 = 1.73 X_2$

The fitted multiple linear regression equation signifies that one unit increase in temperature led to an increase in stigma receptivity in relation to seed set by 2.52 units. Similarly, the equation further indicates that one per cent increase in relative humidity would enhance the stigma receptivity by 1.73 units.

Correlation between per cent seed set and temperature as well as relative humidity was non-significant (Table 7.).

Table 7. Correlation coefficients between per cent seed set, temperature and relative humidity

Factors considered	d.f. (n-2)	'r' value	Calculated value (t)	$t_{0.05}$
Per cent seed set and temperature (°C)	6	-0.142	-0.351	2.45
Per cent seed set and relative humidity (%)	6	0.592	1.799	2.45
Temperature (°C) and relative humidity (%)	6	-0.765	-2.910	2.45

• Significant at 5% level

The coefficient of multiple determination was 46.63 per cent, indicating that about 46.63 per cent of changes in stigma receptivity could be attributed to temperature and relative humidity.

Temperature influences the stigma receptivity to greater extent vis-à-vis to relative humidity. This implies that in hills, where climate is mild, the stigma receptivity may be prolonged, thus affording more time for affecting crosses, while in hot climate ,pollination in early hours of the day may be effective.

Seed setting studies

No significant differences were found for seed setting under all the system of pollination for number of pods per plant, seed yield per plant, hundred seed weight and number of seeds per pod (Table 8.). The absence of significant differences in these characters under the three systems of pollination could be due to the fact that okra is essentially a self-fertilized crop. Purewal and Randhawa (1947) also reported okra as potentially a self fertilized crop and found no significant differences in seed set under self pollination, open pollination and hand cross pollination. Mishra *et al.* ((1993) also reported that systems of pollination did not have any effect on seed weight of okra. On contrary, Abdul-Majeed *et al.* (2003) reported higher percentage of seeds per pod, seed weight and pod length in insect-pollinated plants as compared to self-pollination.

Table 8. Effect of different systems of pollination on some quantitative characters

Characters	Systems of pollination			
	Controlled pollination	Open pollination	Hand sib-pollination	CD (P=0.05)
No. of pods/plant	10.50	10.73	9.23	NS
No. of seeds/pods	56.03	55.77	54.93	NS
100 seed weight(g)	5.88	5.67	5.46	NS
Seed yield/ plant	24.30	24.37	23.23	NS

These results predict that the maintenance of pure line varieties by controlled self pollination or hand sib pollination may be as good as the multiplication of seeds in isolation under open pollination.

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