

# **STUDIES ON THE FLORAL BIOLOGY AND FRUIT SET IN COCOA (*Theobroma cacao* L.)**

BY  
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## **THESIS**

Submitted in partial fulfilment of the  
requirements for the Degree of

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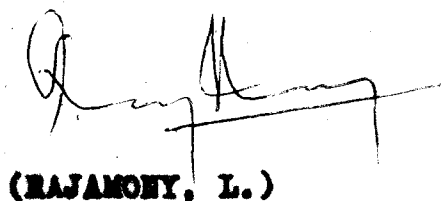
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**Dedicated to the fond memory of my mother.**

## DECLARATION

I hereby declare that this thesis entitled "Studies on the floral biology and fruit set in cocoa (Theobroma cacao L.)" is a bona fide record of research work done by me during the course of research and that the thesis had not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or society.

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# **CERTIFICATE**

Certified that this thesis entitled "Studies on the floral biology and fruit set in cocoa (Theobroma cacao L.)" is a record of research work done independently by Sri. Rajamony, L. under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to him.



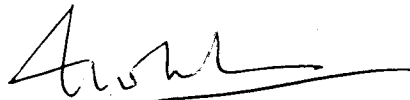
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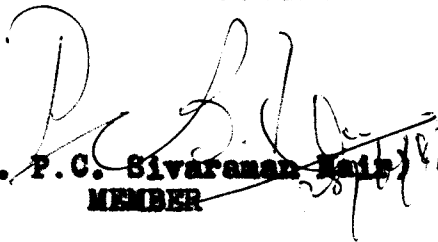
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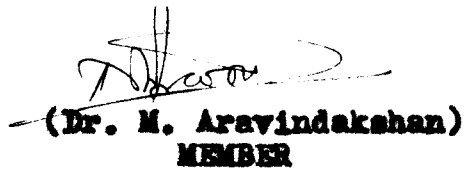
We, the undersigned members of the Advisory Committee of Sri. Rajamony, L., a candidate for the degree of Master of Science in Horticulture majoring in Plantation Crops agree that the thesis entitled "Studies on the floral biology and fruit set in cocoa (Theobroma cacao L.)" may be submitted by Sri. Rajamony, L. in partial fulfilment of the requirements for the degree.



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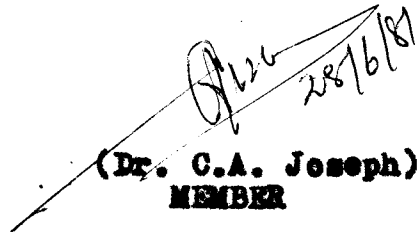
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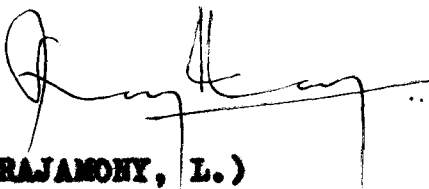
I am grateful to the members of my advisory committee, Dr. M. Aravindakshan, Professor of Horticulture, Dr.K. Kumaran, Associate Professor, and Dr. C.A. Joseph, Associate Professor for their valuable suggestions and encouragement.

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(RAJAMONY, L.)

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# *Introduction*

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## INTRODUCTION

Cocoa, Theobroma cacao L., is a recent introduction to India. It has proved itself to be an ideal intercrop for the coconut and arecanut plantations in the three South Indian states, namely, Kerala, Tamil Nadu and Karnataka. During the past five years, the area under cocoa in India increased from about 8,000 ha (in 1975-76) to more than 13,500 ha (in 1979-80). The production rose from around 200 tonnes to about 2,000 tonnes during the corresponding period. According to the National Commission on Agriculture, the production is expected to touch 20,000 tonnes mark by the turn of the Century.

Though massive expansion of the area has taken place in South India, particularly in Kerala, efforts to increase the productivity per se had been lacking. Other cocoa growing countries like Ghana, Nigeria and Malaysia have lines yielding as much as 3,000 kg of dried beans per ha per year (Wood, 1975). Increase in the productivity of plants can only be achieved by systematic and meaningful crop improvement programmes, for which basic data on various aspects of floral biology, fruit set, fruit development, etc., are pre-requisites. Though some information on these aspects are available in the literature,

data on the extent to which these parameters vary under the conditions of South India are yet to be generated.

Studies were, therefore, undertaken at the Regional Research Station, Pillicode during 1980-81 to gather information on the pattern of flowering and fruiting, aspects of floral biology as well as on fruit set and development in cocco.

# *Review Of Literature*

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## REVIEW OF LITERATURE

### Climatic requirements

Cocoa, Theobroma cacao L. is a tropical plant. The climatic features of the major cocoa growing areas, though varying to some extent, fall within the tropical range.

**Rainfall:** Adams and McKelvie (1955) stated that most of the cocoa growing areas had a short mild dry season while in Ghana, cocoa was limited to those areas which received not less than 250 mm of rain between November and March. According to Guatrecasas (1964), cocoa flourished where the rainfall ranged between 2000 mm and 8000 mm with more or less even distribution throughout the year. Pursglove (1974) remarked that the rainfall in cocoa belt varied from 1010 to 2540 mm. Further, he stated that the rainfall should be well distributed preferably with 101 mm or over per month, and with the absence of marked or intense dry season with less than 63.5 mm per month. Wood (1975) reported the mean annual rainfall in most cocoa growing countries to be between 1150 and 2500 mm. In the main cocoa growing areas of the West Africa, the rainfall varied from 1150 to 1800 mm while in Eastern Nigeria, West Cameroon and Fernando Po, rainfall amounted to 2500 to 3000 mm per annum. He concluded that total annual rainfall was a less important consideration than its distribution.

Temperature: Ernehelm (1948) stated that the lower limit temperature for cocoa was a mean monthly minimum of  $15^{\circ}\text{C}$ . The optimum temperature range for cocoa varied from  $21.1$  to  $32.2^{\circ}\text{C}$ . According to Wood (1975), a minimum range of  $18^{\circ}$  to  $21^{\circ}\text{C}$  and a maximum of  $30^{\circ}$  to  $32^{\circ}\text{C}$  limited the cocoa belt.

### Flowering

Hewison and Ababio (1929) studied the flowering pattern of cocoa in Ghana and stated that the period from March to July was the time of main flowering activity with the greatest number of flower production in April-June. They also observed that where less than a third of a tree's crop was set by the end of April, great flowering activity occurred during June. Alvin (1965) showed that the flowering was most intense during the early part of the rainy season, following the July-September drought. The scarcity of flowers from June to September was attributed to the indirect effects of low temperature. Alvin (1966) observed that the nonflowering period of cocoa in Bahia was July-September. The burst of flowering at the beginning of the wet season resulted in the main crop, after five to six months. Flowering was inhibited when the monthly mean temperature went below  $23^{\circ}\text{C}$ .

Delpinalrivero and Acunagale (1967) stated that in Cuba, abundant flowering occurred from June to September and gradually decreased thereafter. Under severe drought conditions, however, flowering decreased earlier and abruptly. Sale (1969) studied the flowering process of cecoa in relation to the temperature conditions in Trinidad, West Indies. He reported that, as compared to plants growing in regions with a day temperature of  $23.3^{\circ}\text{C}$ , plants in the regions with a day temperature of  $26.6$  to  $30.0^{\circ}\text{C}$ , had more active flowering cushions per plant and more number of flowers per cushion per week. In weather conditions suitable for growth, flowering continued until suppressed by crop development (Glendining, 1972). Alvin et al. (1972) also observed that the inhibition of flowering was due to the crop on the tree which suppressed the flowering. Couprie (1972) showed that flowering was greatest when the daily temperature variation was <sup>via</sup> least. According to Wood (1975), the number of flowers increased as temperature increased. Murray (1975) reported that a well developed cushion usually carried a large number of flowers at any one time and a full grown tree produced more than 10000 flowers in a year. In Vittal, Karnataka State, the annual flower production per meter of stem varied from 168 to 2358 (Anon., 1977).

### Pattern of cropping

Hewison and Abadio (1929) recorded that only 0.2 to 1.5 per cent of the opening flowers developed into mature fruit. Purselove (1974) as well as Cobley and Steele (1976) also estimated that only one in 500 flowers (0.2 per cent) matured to a fruit. According to Murray (1975), out of 10000 flowers produced by a full grown plant in a year only 10 to 50 (0.1 to 0.5 per cent) developed as mature fruits.

Chat (1953) reported that in most of the countries, cocoa harvest peaked twice an year - once during the rainy season and again during the dry season. In countries with pronounced wet and dry seasons, the main harvest occurred five to six months after the start of the wet season (Bridgland, 1955 and Alvin, 1967). Alvin (1974) studied the climatic and cropping pattern of cocoa in Bahia and West Africa. In Bahia, where the rainfall was fairly well distributed, the cocoa harvest season was found to be rather long, usually starting in April and extending until mid January. The April to August crop was being called as the "temporao" and the September to January crop as the "Safrá". The temporao crop was bigger than the safrá crop, depending on the rainfall pattern. January to March period

was critical (with respect to rainfall) for the temporao crop and September to November, for the safra. During the years with well distributed rainfall, the temporao and safra had almost the same volume. On the other hand in West Africa, about 80 to 90 per cent of the crop was harvested in a relatively short period between September and December (main crop) and only 10 to 20 per cent during May-July (mid-crop). Alvin (1974) also explained that the long dry season from October-November to March-April was the main factor responsible for the reduced mid crop in West Africa. Purseglove (1974) stated that the cocoa plants produced pods, throughout the year; but the main harvest usually began at the end of the wet season and continued for a period of three months. Accordingly, in West Africa, the main harvest was during October-January and in Trinidad, during February-March, followed by a minor harvest early in the rains. Wood (1975) opined that as in most other tropical crops, the cocoa harvest was not confined to one short period. There were peak harvest periods, one or two per year and in many countries, there was some cocoa to be harvested at all times of the year. He also suggested that in Ghana, on an average, 25 per cent of the crop was harvested in the peak month, November, which was about six months after the wet season began. In Malaya, where there

was no true dry season, the peak of harvest was less pronounced with 20-25 per cent of the crop in the peak which falls between November and March. Shannugavelu and Rao (1977) stated that there was two well defined cropping seasons in Ceylon - one from May to August and the other from September to January. Studies undertaken at the Kallar Fruit Station on the cropping and productivity of Criollo cocoa revealed that in the young plantations, the cropping was confined to November-June, with the major portion of the crop obtained during November-December (Kuppuswamy and Kailasam, 1970, cited by Shannugavelu and Rao, 1977). Generally in South India, cocoa is found to have two main crops in a year, in September to January and in April to June (Anon, 1978).

#### Cherelle wilt

According to Nichols (1961), cherelle wilt was a physiological thinning mechanism which regulated the size of the crop in relation to the available food reserves in the trees. Glendinning (1972) reported that any excess fruit setting was corrected by cherelle wilt. Couprie (1972) classified fruitset and cherelle wilt as the most important factors affecting yield. Murray (1975) also confirmed Nichols' view that cherelle wilt was a physiological thinning mechanism.

Heweson and Ababie (1929) reported that the losses from shedding and shrivelling of immature pods ranged from 22-84 per cent of the fruit set. In Ghana, according to Hampries (1947) and McKelvie (1955 and 1957), cherelle wilt was generally highest between April and June. The percentage of cherelle wilt was higher in large cushions, containing 10 to 30 flower buds (Naundorf and Willamil, 1949). Reyes et al. (1969) used clonal cocoa plants in their study and showed that the percentage of prematurely wilted fruits varied during the year. Losses occurred during the dry months of March-April and again during September-October. Couprie (1972) found that cherelle wilt was affected by rainfall during the 8th week before the fruit set and by the temperature during the second preceding weeks.

#### Anthesis, anther dehiscence and stigma receptivity

Wellensiek (1932) stated that in Java, the flowers opened at 4.30 PM. Around 6.30 AM, the anthers opened and the pollens were ripe. The flower was protogynous, the stigma being ripe when opened. Sampayan (1963 and 1966) reported that in the criollo and Trinitario cocoa trees in the Philippines, anthesis started at 4 PM and the sepals and petals unfolded by 6 PM. Anther dehiscence started at 6 AM of the following morning and the pollen was accessible

to insects from 8 to 8.30 AM onwards. Based on the hand pollination made at hourly intervals on the same day, he observed that the percentage of fruitset was more between 6 AM and 5 PM, reaching a maximum when pollinations were done between 10 AM and 1 PM. Delpinalrivero and Aounagale (1967) found that in Cuba, flowers normally opened at 7 PM, but remained closed or half opened almost the whole day at a temperature of about 15°C.

#### Pollen viability, germination and storage

Viability: Wellensiek (1932) reported that cocoa pollen grains removed from flowers (which had been open for more than 1½ days) failed to germinate.

Media for germination: Sucrose has been found to be quite helpful as an artificial medium for testing pollen germination serving as a nutrient during pollen tube growth (Ostopenko, 1956; Vasil, 1960; Singh, 1961 and Singh et al., 1961). Shapiro and Burdick (1961) suggested that, in general, the germination and pollen tube growth increased when boric acid and calcium were added to sucrose solution. Varas and Sonia (1962) suggested 5 to 15 per cent sucrose in the media for germinating cocoa pollen. The best germinating medium for cocoa pollen appeared to be two per cent



agar with 10 per cent sucrose. They also observed that addition of one to ten ppm Boric acid seemed to stimulate germination of cocoa pollen from 22.0 to 42.3 per cent. Jacob et al. (1969) reported that germination of cocoa pollen was possible in sacrose solution (upto 30 per cent concentration). Ravindran (1977) obtained good cocoa pollen germination and tube growth in a medium containing 15 per cent sucrose. He further stated that boron or calcium was necessary for proper pollen germination and tube growth and that these nutrients enhanced tube growth considerably.

Delpinalrevero and Acunagale (1967) stated that cocoa pollen kept at or below  $14^{\circ}\text{C}$  for two days germinated slowly. Martinson (1973) obtained best germination of cocoa pollen grains at a temperature of 28 to  $31^{\circ}\text{C}$ . Ravindran (1977) observed no pollen germination at low temperature ( $10^{\circ}\text{C}$ ) and maximum germination and tube growth at  $35^{\circ}\text{C}$ . Further increase in temperature to  $40^{\circ}\text{C}$ , affected both germination and tube growth.

Storage: According to Wellensiek (1932), cocoa pollen retained its germinating power in closed glass tubes for at least 12 hours. Varas (1962) found that pollen stored for a week in a desiccator germinated better in artificial medium. Simmons (1976) recorded that cocoa pollen could be kept at  $5^{\circ}\text{C}$  in sealed tubes over calcium chloride for about one week.

### Pollination aspects

Pollinating agents: Wellensiek (1932) reported that in Java, pollination by insects was not observed. According to him, cross pollination probably occurred only between flowers situated close together. In Trinidad, Frankliniella parvula Hood and Wasmannia auropunctata Rog. were reported to be mostly responsible for pollen transportation (Cops, 1940). The insects responsible for much of the cross pollination in coeca were identified by Posnette (1950) as Forcipomyia quasi ingrains; Aashanti and Lasiochelea litoraurea. Posnette and Entwistle (1958), Saunders (1958), Dessart (1961), Sumner (1962) as well as Gerard and Saunders (1964) have also listed Forcipomyia spp. as the major pollinating insect in coeca plantations. Cuatrecasas (1964) remarked that pollination in coeca was only by insects. Several kinds of flying and crawling insects (thrips, ants, midges and aphids) were found to be involved in pollen transportation. Texopeus and Jacob (1965) recorded that out of the 450 freshly opened flowers examined none contained Forcipomyia spp while aphids were present in most of them. Hernandez (1967) identified thrips (Frankliniella parvula); aphids (Aphis goswami); ants (Wasmannia auropunctata) and bees (Trigona laty) as the insects most commonly found associated with coeca flowers at Turialba. Entwistle (1972), Glendinning (1972) and

Soria et al. (1975) listed the flying female midges of the genus Forcipomyia as the most important pollinating agents in cocoa. Kaufmann (1975) revealed that not less than 50 species of ceratopogonidae were found at Tafo, Ghana and the most abundant one was Forcipomyia squamipennis. He also reported a new pollinator identified as a species of genus Trigona (Hymenoptera: Apidae), variously called sweet bees, stingless bees or morpane bees. The reports of Winder (1978) showed that the Dipterans associated with cocoa flowers included 70 species of ceratopogonidae, of which 75 per cent were Forcipomyia spp. and 20 species, of other depterous families.

Natural pollination, fruit set and hand pollination:

Barroga (1965) reported that the highest rate of pollination was observed from 6.00 to 11.00 AM and from 2.00 to 5.00 PM. He also recorded that no pollination occurred at noon. Toxopeus and Jacob (1965) stated that the percentage of pistils with pollen grains was slightly higher in the flowers collected between 12 noon and 1 PM which indicated that more natural pollination took place at mid-day than in the early morning. Widjanarko (1967) proved that the extent of pollination was chiefly influenced by the morning weather conditions and was low in rainy weather. Purseglove (1974) opined that the low pollination efficiency in cocoa was

compensated by the large number of flowers produced. In Ghana, 2 to 5 percentage of flowers only are pollinated and a fairly large number of these failed to set seed. Later during the season, when flowers were fewer, pollination rose to 50 to 75 per cent. Murray (1975) reported that estimates of the proportion of flowers pollinated ranged from 1 to 50 per cent, according to the season and number of flowers opening at the time. But the percentage of pollinators were higher during August (57.3) followed by in July (56.2). The lowest per cent was in June (11.2) (Kaufmann, 1975). Under natural conditions, pollen dispersal was most intense at 8 AM and 5 PM (Massaux et al. 1976). In the cocoa plants of South Karnataka (Vittal) successful pollination (stigmas having 40 or more pollen grains) was only to the tune of 8 per cent though the percentage of flowers pollinated was 28. The maximum successful pollination (11 per cent) was in March and minimum (4 per cent) in December. Though, high pollination per cent was recorded in August, there was no fruit setting (Anon, 1977). The microscopic examinations of a number of styles by Parvais et al. (1977) revealed that only a few of them carried pollen in quantities sufficient to ensure fertilizations. They therefore suggested that pollination appeared to be a limiting factor for good yields. Winder (1978) also reported that lack of pollination was responsible for low production in cocoa.

Normal fruit setting in cocoa required deposition of pollen grains on stigma in quantity (Glendining, 1972). Low rates of fruit setting in naturally pollinated flowers appeared to be due to inadequate or non-pollination. A large proportion of flowers never produced fruit even if they were fertilized. Further, large number of fertilized ovaries are shed before maturity (Amponsah, 1972 and 1977).

According to Pureglove (1974), fertilization took place seven to eight hours after pollination and it could be ascertained by the swelling of the ovary. Fruit setting was generally more during December to June. The mean annual fruit setting was only three per cent. Lower fruit setting observed during peak period of flowering, has been attributed to insufficient number of pollinating agents (Anon, 1977).

Wellensiek (1932) used a method of artificial pollination in cocoa in which the pollen flowers were picked in the morning and used for pollinating newly opened flowers at 7.30 PM the same evening. This method resulted in three per cent of ripe fruits. Based on the results of hand pollination in cocoa, Posnette (1950) and Jacob and Atanda (1975) reported that the pod yield could be improved considerably by hand pollination. Amponsah (1972 and 1977) found that the percentage of fruit set on hand pollination was very

much higher than those by the natural pollination (30 per cent and 2 per cent, respectively of the total number of flowers).

### Compatibility

Self-incompatibility in cocoa was first reported in Trinidad by Harland in 1925 and again by Pound in 1932 (Purseglove, 1974). Self-incompatibility in Theobroma cacao L. (which reduced the flower setting and number of pods/tree) has been reported to be a major barrier in realizing the yield potential of cocoa cultivars (Toxopeus and Jacob, 1970 and Jacob and Atanda, 1975).

According to Cope (1962), the site of incompatibility was the embryo sac and not the stigma and style. Incompatibility occurred only when the male gametes came to be in contact with the egg and the polar nuclei in the embryo sac. Bartley (1963) reported that out of 39 trees, eight trees were self-compatible. Criollo Forestero natural hybrids exhibited varying degrees of self-compatibility from 0 to 93 per cent (Sampayan, 1963 and 1966). In Brazil, the clone UF-221 appeared to be self-compatible while the clones UF-667 and UF-613 showed high degree of self-incompatibility (Delpinalrivero and Acunagale, 1967). Purseglove (1974) stated that in the incompatible matings, the flowers were shed three to four days after pollination. He further stated that the self incompatible Trinitario clones were cross incompatible

also; but were compatible with self compatible trees. In Trinidad, the self-incompatible and cross-incompatible tree required pollen from a self-compatible tree to ensure fruit-set; but elsewhere self-incompatible trees showed cross-compatibility (Murray, 1975). Based on the assessment of flower setting and pod harvesting in 23 W.A.C.R.I. 'C' clones, Jacob and Atanda (1975) identified two self-compatible clones, C-26 and C-73.

#### Fruit development and occurrence of cherelle wilt

The time taken by cocoa pods to reach maturity stage from fertilization has been reported to be five months (Wood, 1975), five to six months (Murray, 1975; Cobley and Steele, 1976) and four to six months (Anon, 1978).

Hewison and Ababio (1929) stated that the majority of pods in cocoa ultimately reaching maturity, attained maximum size in seventeen to eighteen weeks after fertilization. The period of development was reported to be sixteen to twentyone weeks in Nigeria (Waters and Hunter, 1929), five to six months in Grenada (Cooper, 1940) and an average of six months in Papua-New Guinea (Bridgland, 1953). Weekly observations by McKelvie (1954) on the growth rate of cocoa pods in Amazon, Amelonado and Trinilario selections showed that the growth rate was similar in these selections upto four weeks of fertilization.

Alvim et al. (1972) presented evidences and concluded that the rate of pod development increased with increase in temperature. Pods grew more slowly in cooler climate.

Hewison and Ababio (1929) found that losses of immature fruits (cherelles) were <sup>the</sup> highest during the first week of growth. Another critical period was between the fourth and seventh weeks. McKelvie (1955) studied the occurrence of cherelle wilt in relation to the fruit development and concluded that cherelle wilt occurred in two distinct waves. The first period of wilt or 'first wilt' began a few days after fertilization and lasted for 9 to 10 weeks with a peak at the 7th to 8th week. The second period of wilt or 'second wilt' started from the 8th or 9th week lasted till the 14th week with a peak at about the 12th week. After 14 weeks (corresponding to a pod of 13 to 14 cm length), there was no further wilt. He also found that the second wilt very rarely occurred in young plantations where the first wilt could account for upto 95 per cent loss. In a later study, McKelvie (1956) found that cherelles with a length of 35 to 60 mm were most sensitive to wilt. According to Nichols (1964), the fruits wilted during the first half of their life cycle ie. upto 80 days (approximately) from fertilization. Purselove (1974) stated that maximum wilt occurred at about 50 days after fertilization when the



cherelles were about 6 cm long. During the stage of further development (50 to 70 days), wilt did not occur and the fruits matured and ripened. Murray (1975) opined that the young developing fruits (during the first 2 to 3 months of their development) were subjected to chereille wilt. Cobley and Steele (1976) pointed out that wilt occurred mostly when the fruits were 7 to 8 weeks old. After three months of growth there was little risk of fruit failing to mature.

# *Materials and Methods*

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## MATERIALS AND METHODS

The studies were conducted from January to December 1980 at the Regional Research Station, Pilicode, Cannanore District on the Criollo-Forastero natural hybrid population of cocoa. Ten year old plants intercropped in an adult coconut plantation were used for the study. Cultural practices and other operations were adopted as per the package of practices recommendations of the Kerala Agricultural University. During the summer months, the plants were irrigated once a week.

The physical and chemical properties of the soil are given in Table 1. The weather data, both of the preceding year (1979) and of the experimental year (1980) are presented in Table 2 and illustrated in Fig. 1 and 2.

### Flowering pattern

Thirtyfive bearing plants were selected and marked for the study. Weekly counts were made of the number of flowers per plant. In the case of plants with very large number of flowers, precise counting was done on the main trunk. Then, the number of flowers on a single fan shoot was counted and this number was multiplied by the number of fan shoots to get the total number of flowers in the fans.

Table 1. Soil analysis particulars at the Regional Research Station, Pilicode

		Content (%)	
Particulars		0-30 cm depth	30-60 cm depth
Physical characters	Clay	27.71	31.41
	Silt	6.90	7.60
	Fine sand	20.44	18.26
	Coarse sand	44.95	42.73
Chemical characters	Moisture	1.73	1.99
	Loss on ignition	5.39	5.76
	Lime	0.04	0.048
	Potash	0.17	0.200
	Phosphoric acid	0.081	0.064
	Nitrogen	0.085	0.071
	Available $P_2O_5$	0.017	0.0069
	Available $K_2O$	0.0025	0.0018

Table 2. Weather data - 1979 and 1980

1979			Month	1980		
Maximum tempera- ture (°C)	Minimum tempera- ture (°C)	Rain fall (mm)		Maximum tempera- ture (°C)	Minimum tempera- ture (°C)	Rain fall (mm)
33.37	20.25	-	January	33.74	18.76	-
33.07	21.53	-	February	32.62	21.09	-
33.48	21.72	-	March	32.88	21.79	-
33.67	23.28	-	April	32.37	22.18	129.4
33.86	22.96	-	May	32.41	21.40	120.9
31.89	22.18	1529.2	June	30.15	21.82	740.6
28.28	21.79	1321.9	July	33.51	21.76	870.0
28.28	22.15	572.0	August	33.01	21.79	839.6
29.23	22.33	191.6	September	32.93	21.67	204.0
30.14	22.37	66.8	October	33.08	21.86	173.8
30.96	22.59	162.0	November	32.74	21.41	148.0
33.01	21.51	45.4	December	32.58	21.36	-
3888.90			Total	3226.30		

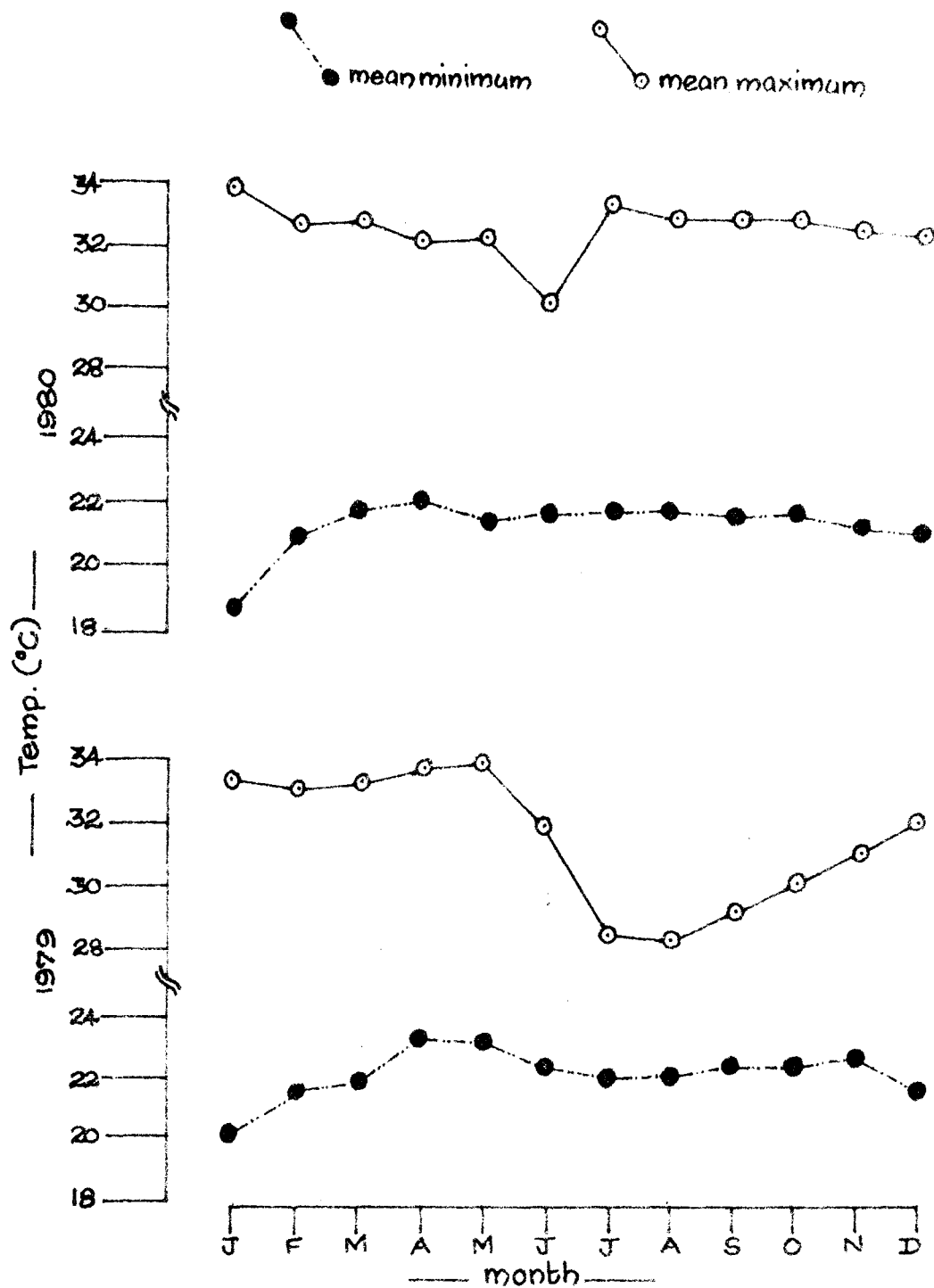


Fig.1 - Monthly maximum and minimum temperatures.  
(1979 and 1980).

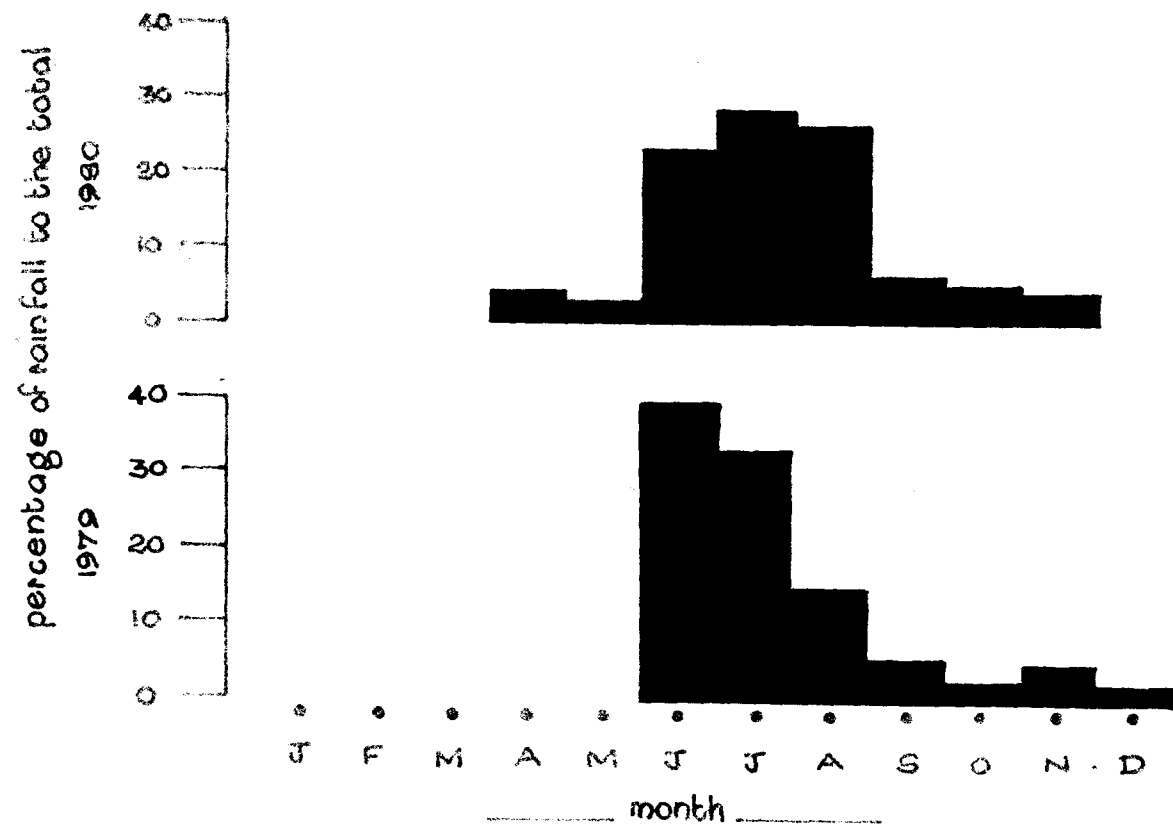


Fig. 2. - Monthly rainfall distribution. (1979 and 1980).

During counting, older flowers were differentiated and excluded by the dried appearance of the stigmatic surface, change of the petal colour from creamy white to deep yellow, the drooping character of unpollinated flowers and by the swollen ovaries of fertilized flowers, as suggested by Purseglove (1974) and Murray (1975). From the observations, the mean number of flowers per plant per day, the mean number per plant for each month and the percentage of flowers for each month (to the total number of flowers produced per plant per year) were calculated.

#### Cropping pattern and intensity of cherelle wilt

Plants tagged for observing the flowering pattern were utilized for this study also. Harvesting of ripe pods was done monthly from January to December. Diseased and squirrel/rat attacked pods were also reckoned as yield. Number of wilted young fruits were recorded during every month and also at harvest. From the data, mean number of pods per plant per month, percentage of pods in each month (to the total number of pods), mean number of wilted pods per month and the percentage of wilted pods in each month (to the total number of wilted pods) were worked out.

#### Emergence of flower buds

Twentyfive cushions were marked on different plants



with indian ink. Two flower buds on each cushion were marked with labelled adhesive tape (on the stem) for recording the pedicel length as well as length and thickness of buds. Observations were started from the date on which the flower buds appeared on the cushion and continued upto the date when the flower bud started to open.

Measurements were taken with a special device - a small graph sheet (5 x 2 cm) fixed on same sized negative film. At the time of observations, the above device was placed below to the flower bud and the bud was viewed through a hand lens. The daily mean increase in the pedicel length as well as length and thickness of flower buds (in mm) were recorded. The number of days taken from the date of appearance to the date of anthesis was also recorded. The observations were repeated during January, April, July and October and the mean number of days worked out.

### Anthesis

Observations were made during January, April, July and October. After preliminary observations on commencement of anthesis (which revealed that it was between 12 noon and 6 PM), 100 mature flower buds were marked with small tag label, each morning. Observations were made at two-hour intervals in order to count the number of buds that started

to split. The observations were continued upto the commencement of anthesis in order to determine the time of complete opening of the flowers.

#### Anther dehiscence

Three hundred and fifty mature flower buds were tagged at the time of anthesis. These were divided into seven lots of 50 each for observation on the commencement and completion of anther dehiscence. The flowers in lots of fifty were collected every two hours from midnight to 10 AM and observed under a dissection microscope. Appearance of a longitudinal split in the pollen-sac indicated the commencement of anther dehiscence. When more than three anthers in a flower liberated pollen grains, that particular flower was taken as having completed anther dehiscence. The observations were repeated during the next day with another set of 350 flowers. Anther dehiscence was studied during January, April, July and October.

#### Stigma receptivity

Stigma receptivity was assessed based on the following methods.

- 1) "adherence of pollen grains to the stigma"  
as suggested by Heslop and Shivanna (1977).

ii) "intense visit of flowers by the insects"

as adopted by Toxopeus and Okoloko (1965) in  
cocoa.

iii) "fruitset after controlled hand pollination"

as adopted in cocoa by Sampayan (1963).

In the first method, anthers from freshly opened flowers were rubbed on to the stigmatic surface of the fresh flowers. The stigmatic surface was then viewed through a hand lens to see whether transfer of pollen has been effected or not.

In the second method, a new device was used for collecting the insects visiting the flowers. Cotton dipped in Benzene was kept in a clean specimen tube. A round piece of filter paper was placed tightly over the cotton. This tube was used for collecting the flowers from the plants along with the insects within them. Two hundred fresh flowers were collected as described above at two hour intervals between 6 AM and 6 PM, taken to the laboratory and dissected carefully to estimate the total number of insects trapped within them.

In the third method, the flowers were bagged with specially made hoods. Muslin cloth bag was covered over

conical aluminium wire frames (5 cm base and 10 cm height). The hoods so made were placed above the cushions without any contact with the centrally located flower buds and fixed to the trunk with four strips of adhesive tape (Plate 1 & 2).

Three hundred flower buds were bagged on the day of anthesis as described above. Fifty flowers were pollinated the next day between 6 AM to 6 PM at two-hour intervals.

For effecting pollination, freshly collected flowers were used after removing the thalamus, the sepals and the petals. The remaining portion, consisting of the ovary, the stamens, the staminodes and the style, was held in the natural pose with the pedicel end upwards (to avoid wastage of pollen grains). The staminodes and the stylar tip were held between the fingers. The free stamens were rubbed against the stigmatic surface of the flower to be pollinated. The percentage of fruitset was worked out based on the number of pods set after two weeks of pollination, as suggested by Jacob and Atanda (1975). The experiment was done during May and November.

#### Size and shape of pollen grains

Pollen measurements were made using the ocular-stage micrometer and a G.B. Olympus microscope (16 X eye piece, 40 X objective), as adopted by Takur and Singh (1965) in



Plate 1. Materials for bagging cocoa flowers.



Plate 2. Cocoa plant showing bagged cushion

annona fruit. Diameter of 100 pollen grains were measured and mean worked out. The size and shape of dry, wet (in distilled water) and stained (acetocarmine 0.5 per cent) pollen grains were estimated.

#### Estimation of pollen production

Pollen production per flower was estimated using a haemocytometer as suggested by Rao and Khader (1962). 0.1 ml water was pipetted out in the field of the haemocytometer. One anther from a fresh flower was crushed into the solution and cover slip placed carefully. Counting was done in each of the 1 mm squares and the mean number of pollen grains in a flower was calculated by the formula,

$$\begin{aligned} \text{Number of pollen grains in a flower (5 anthers)} \\ = X.9.5 \end{aligned}$$

where,

X = mean number of pollen grains in 1 mm square.

The pollen count was made during May and November and the mean worked out.

#### In vitro germination and growth of pollen tube

Media for germination consisted of agar, sucrose and boric acid in five concentrations as given below.

<u>Agar (%)</u>		<u>Sucrose (%)</u>		<u>100 ppm boric acid (%)</u>
0.1	+	5.0	+	0.5
0.5	+	10.0	+	1.0
1.0	+	15.0	+	2.0
2.0	+	20.0	+	5.0
5.0	+	50.0	+	10.0

Agar was dissolved in 10 ml distilled water by heating gently with continuous stirring. Sucrose was added to this little by little. When the sucrose completely dissolved, boric acid solution was pipetted into it and the mixture was poured into a petri-dish. To prevent cracking on solidification of the medium, the cover of the petri-dish was lined with a wet filter paper. The cover was removed only at the time of testing. The media were kept overnight at room temperature. The glassware and other instruments used for the study were sterilized at 15 lbs/sq. inch for two hours.

Fresh flowers were collected at 6 AM and the pollen grains were dusted onto the medium kept on a slide. Germination of pollen grains and pollen tube growth measurements were made after killing and staining with phenol aniline blue, as adopted by Ravindran (1977) in cocoa. Germination count was recorded twice (after 1 hour and 4 hours) and the pollen tube growth, once (after 4 hours).

The medium which recorded higher germination per cent and tube growth was taken as the standard one. Using this as the base, effect of each chemical was studied separately.

The effects of boron (as boric acid) and calcium (as calcium nitrate) alone and in a 1:1 mixture were tested at concentrations ranging from 50 to 300 ppm in 10 per cent sucrose solution. The use of agar was avoided because it was known to contain calcium and boron as contaminants (Moore and Jung, 1974). Effect of temperature on germination and tube growth was studied over a range of 10°C to 40°C. In all the studies pertaining to the effects of different treatments, the germination count and the tube growth were observed after four hours.

#### Pollen viability

Viability of pollen grains was found out by two methods.

i) Acetocarmine staining method as suggested by Zirkle (1937) and adopted by Singh (1961) in mango and Parameshwar (1974) in cardamom.

ii) Germination method as suggested by Stanley and Linskens (1974) and adopted in cardamom by Parameshwar (1974).



Stainability was assessed in 0.5 per cent acetocarmine. Pollen grains were germinated in a medium containing 0.5 per cent agar, 10.0 per cent sucrose and 1.0 per cent of 100 ppm boric acid (the most suitable medium identified in the in vitro study).

One hundred flowers (50 for the acetocarmine staining and 50 for the germination study) were collected in the morning. Pollen grains of each flower were put into a cavity slide with the stain/germination media and the stained/germinated pollen were counted separately. From the counts of 20 different fields, the mean percentage of viable pollen grains was worked out. The experiment was repeated four times (January, April, July and October).

Variation in the viability of pollen (after the dehiscence) was studied from 6 AM to 6 PM for a period of seven days during the month of November. The viability was calculated based on the percentage germination of pollen grains and the mean tube length at two hour intervals.

#### Pollen storage

Loss in the viability of pollen as a result of storage was estimated as described below.

Fresh flowers were collected in a petri-dish at 6 AM. The anthers were put in a tissue paper packet, wrapped with filter paper and inserted in a glass specimen tube as suggested by Patel (1938) in coconut. The tubes were sealed with paraffin wax. One lot was kept at room temperature as suggested by Wellensiek (1932) and another lot after two hours of desiccation (over calcium chloride) as suggested by Simmons (1976), was stored in a refrigerator (at 5-10°C). Anthers collected from freshly opened flowers and kept on wet filter paper inside a petri-dish at room temperature served as the control.

In the above experiment, viability, based on germination and pollen tube growth was tested daily till no germination was obtained.

#### Pollination aspects

In order to assess the extent of self pollination, 100 flower buds were bagged (as described earlier) before the anthesis. The percentage of pollinated flowers was estimated after two days of anthesis as per the method suggested by Purseglove (1974).

Insects visiting the cocoa flowers were collected as described earlier and identified. Photomicrographs of the insects were also taken.

The monthly variation in the rate of natural pollination was estimated from January to December. Three hundred flowers were collected at 4 PM on the day after the commencement of anthesis. The gynoecium was dissected out and examined in order to count the number of pollen grains on the style and the stigma. The flowers with no pollen grain on the style or on the stigma were considered as not pollinated; those with 1 to 39 pollen grains, as under pollinated and those with 40 pollen grains or more, as successfully pollinated as recommended by Anon (1977).

#### Compatibility status

Fifteen steady bearing plants were tagged and these were self pollinated and crossed in combination as shown in Table 23. For controlled self pollination, flowers of the same plant were used. The pollinated flowers were bagged as described earlier and were marked with labelled pieces of adhesive tape stuck to the trunk near the flower. The bags were removed after three days of pollination. After two weeks of pollination, fruit set was estimated and the developing pods were marked with tag labels. The number of pods carried to maturity was also recorded. Assessment of the compatibility status of the trees were made using the criteria proposed for Cola nitida by

Jacob (1970) and for cocoa by Jacob and Atanda (1975). According to these criteria, a tree is considered self/cross compatible if it records a minimum of 10 per cent fruit set and 5 per cent pod harvest.

In order to compare open pollination with hand pollination, 40 flowers were marked as described earlier on each of the 15 plants. They were allowed to set by natural pollination. The setting percentage as well as pods carried to harvest were recorded. These data were compared with the figures obtained in the cross-pollination and compatibility status study.

#### Flowering and fruiting on the main trunk and on fan shoots

Ten plants were selected for this study. On each plant, two cushions on the main trunk and two cushions on the fan branches were marked with indian ink. Total number of flowers produced per cushion, number of flowers pollinated, number of fruits set, number of cherelles wilted and number of pods carried to maturity were recorded during the entire season.

After completing the first cycle of flower production, the observations were continued on the same cushions in order to determine the pattern of subsequent flowering.

### Fruit (pod) development studies

One hundred flowers were tagged after fertilization (assessed by the swelling of the ovary and its green colour as proposed by Purseglove, 1974). These developing fruits (cherelles) were observed at weekly intervals. Length and thickness of the pedicel as well as the length and thickness of the fruits were recorded. The device developed for the flower bud emergence studies was used for recording the measurements at cherelles during the first week. Subsequently, the measurements were made using a metre scale. The number of cherelles wilted was also recorded.

Total number of days taken from fertilization to the maturity of pods were worked out and compared with the figures obtained as per the formula of Alvim et al. (1972), namely,

$$N = \frac{2500}{T-9}$$

where,

N = number of days to maturity and,

T = daily mean temperature in °C.

Fresh flowers were tagged every week for tracing the internal developments. When the pods developed from flowers tagged during the first week ripened, the tagging was stopped. Thus a series of developmental stages, from fresh flowers to ripe pods, was available. Photographs of the whole fruits and their L.S. were taken.

## *Results*

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## R E S U L T S

### Flowering pattern

Data on the flowering pattern in cocoa are presented in Table 3 and illustrated in Fig. 3.

Flower production was seen throughout the year. Mean number of flowers/plant/day varied from 7.02 in September to 132.89 in December. Highest flowering was recorded in December followed by May. It was observed that there were two peak seasons of flowering, namely, November-December accounting for 42.68 per cent and May-June accounting for 19.41 per cent. Lowest flower production was recorded in September (1.71 per cent).

### Pattern of cropping

Harvest of pods was confined to the months of April, June to August and October to December as can be seen from the data presented in Table 4 and Fig. 4.

The maximum production of pods (34.98 per cent) was observed in the month of July. June to August season accounted for 74.11 per cent of the total crop during the year and October-December season, 21.59 per cent.

Table 3. Flowering pattern

Month	No. of flowers	Mean/plant/ day <sup>®</sup>	Total for the month	Percentage to the total for the year
January		22.56	699.36	5.69
February		40.22	1166.38	9.49
March		29.36	910.16	7.40
April		17.33	519.90	4.23
May		51.49	1596.19	12.98
June		26.34	790.20	6.43
July		17.38	538.78	4.38
August		11.68	362.02	2.94
September		7.02	210.60	1.71
October		8.19	253.89	2.07
November		37.59	1127.70	9.17
December		132.89	4119.59	33.51
Total		-	12294.83	100.00

<sup>®</sup> Mean of thirtyfive plants averaged over four observations

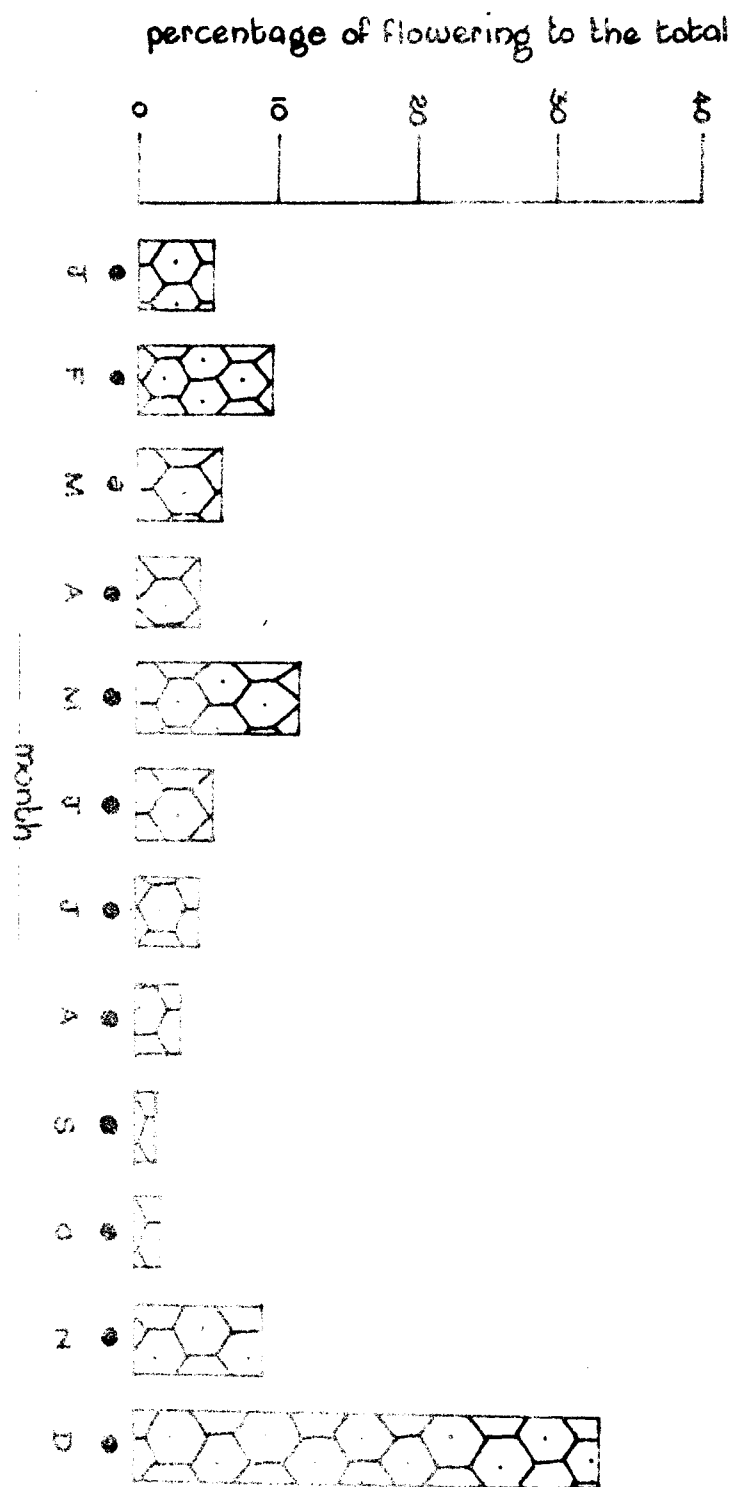


Table 4. Pattern of cropping

No. of pods	Mean/plant <sup>©</sup>	Percentage to the total for the year
Month		
January	-	-
February	-	-
March	-	-
April	1.54	4.30
May	-	-
June	10.57	29.48
July	12.54	34.98
August	3.46	9.65
September	-	-
October	2.0	5.58
November	2.20	6.14
December	3.54	9.87
Total	35.85	100.00

© Mean of thirtyfive plants

FIG. 3. FLOWERING PATTERN.



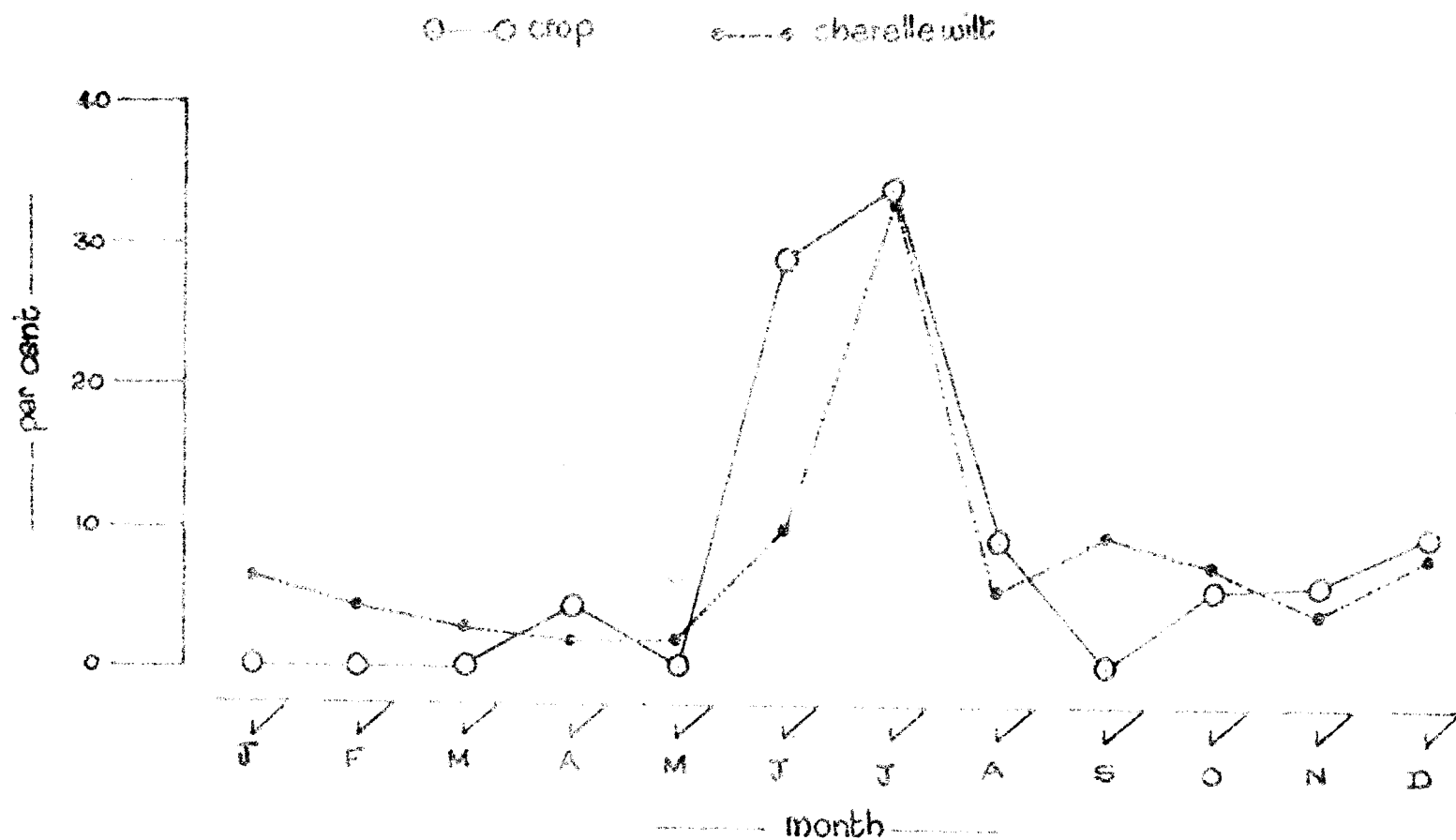


Fig. 4.— Pattern of cropping and intensity of cherelle wilt.

### Intensity of cherelle wilt

Data regarding the intensity of cherelle wilt are presented in Table 5 and illustrated in Fig. 4. Cherelle wilt was observed to occur throughout the year. The intensity was highest during July. Lowest intensity was observed in the month of April.

### Emergence of flower buds

Data regarding the rate of development of the flower buds are presented in Table 6a. Pedicels started to elongate only from the third day of the appearance. Length and thickness of the buds started to increase from the fifth day onwards. As the buds reached full maturity, the mean length of the pedicels was 18.33 mm, length of buds, 8.25 mm and thickness of buds, 4.45 mm.

Number of days taken from the appearance of flower buds on the cushion to the date of anthesis ranged from 21 to 24 (Table 6b). Data regarding the weather parameters during the period of observation are also presented in this table.

### Anthesis

Observations regarding anthesis at two hourly intervals are given in Table 7. It can be seen that, in general,

Table 5. Intensity of cherelle wilt

No. of cherelle wilt	Mean/plant <sup>®</sup>	Percentage to the total for the year
Month		
January	30.50	6.43
February	20.80	4.38
March	13.98	2.95
April	8.13	1.71
May	9.80	2.06
June	43.53	9.17
July	159.02	33.51
August	27.0	5.69
September	45.02	9.49
October	35.13	7.40
November	20.07	4.23
December	61.61	12.98
Total	474.59	100.00

<sup>®</sup> Mean of thirtyfive plants

Table 6a. Growth of flower buds from appearance to anthesis

Days after appear- ance	Mean <sup>®</sup> increase	Length of pedicel (mm)	Length of flower bud (mm)	Thickness of flower bud (mm)
1		No change	No change	No change
2		"	"	"
3		1.07	"	"
4		1.50	"	"
5		2.50	0.38	0.57
6		3.32	0.65	0.68
7		4.01	0.85	0.91
8		4.94	1.10	1.0
9		5.75	1.44	1.22
10		6.57	1.66	1.32
11		7.51	2.13	1.69
12		8.50	2.63	1.94
13		9.44	3.07	2.13
14		10.38	3.54	2.25
15		11.32	4.01	2.50
16		12.26	4.51	2.63
17		13.07	5.02	2.88
18		13.82	5.52	3.10
19		14.44	6.38	3.25
20		15.07	6.51	3.50
21 <sup>@@</sup>		15.63	6.79	3.69
22 <sup>@@</sup>		16.28	7.13	3.75
23 <sup>@@</sup>		16.84	7.51	4.0
24 <sup>@@</sup>		17.63	8.15	4.25
25 <sup>@@</sup>		18.13	8.25	4.45

<sup>®</sup> Mean of fifty flower buds.

<sup>@@</sup> The buds which showed anthesis were not measured. Hence the number was less than fifty.

Table 6b. Mean number of days from appearance to anthesis

Month	No. of buds observed	No. of days	Mean tempera- ture in °C		Total Rain fall in mm
			Maximum	Minimum	
January	50	23.50	33.74	18.76	0
April	50	23.75	32.37	22.18	129.40
July	50	21.75	33.51	21.76	870.0
October	50	22.75	33.08	21.86	173.80
Average	50	22.94	33.17	21.14	293.30

Table 7. Anthesis

Date and month	Commencement--No. of flowers at 2 hours interval <sup>@</sup>					Weather data on the day			Completion (next day)--No. of flowers at 2 hours interval <sup>@</sup>			
	10 AM to 12 noon	12 noon to 2 PM	2 PM to 4 PM	4 PM to 6 PM	6 PM to 8 PM	Maximum temperature in °C	Rain fall in mm	Minimum temperature in °C	12 mid-night to 2 AM	2 AM to 4 AM	4 AM to 6 AM	6 AM to 8 AM
10th January	0	0	82	15	3	33.33	0	18.89	0	79	18	3
10th April	0	0	91	8	1	31.11	0	21.11	2	80	16	2
10th July	52	20	26	2	0	34.44	52.4	22.22	5	85	10	0
10th October	0	0	85	11	4	33.33	0	22.22	21	67	11	1
Average	13.00	5.00	71.00	9.00	2.00	33.05	13.10	21.11	7.00	77.75	13.75	1.50

<sup>@</sup> One hundred flower buds for each interval



71.0 per cent of the flower buds started splitting between 2 and 4 PM. However, during July, more than 50 per cent of the flowers started to split between 10 AM and 12 noon. 77.75 per cent of the flower buds completed the anthesis between 2 and 4 AM, the next day.

#### Anther dehiscence

It can be seen from the data presented in Table 8 that dehiscence in 85.75 per cent of the anthers commenced between 4 and 6 AM. 85 per cent of the anthers completed their dehiscence by 8 to 10 AM.

#### Stigma receptivity

When pollen grains were dusted on the stigmatic surface of the fresh flowers, adherence was observed from 6 AM to 6 PM. The data on number of insects associated with the flowers collected at two hourly intervals (Table 9 and Fig. 5) revealed that more number of insects visited the flowers between 12 noon and 2 PM.

The results of hand pollination done at two hourly intervals from 6 AM to 6 PM are presented in Table 10 and Fig. 5. The data revealed that the flowers which were pollinated between 6 AM to 8 AM and between 4 PM to 6 PM did not set. Seventynine per cent of the flowers pollinated between 12 noon to 2 PM exhibited fruit set.

Table 8. Anther dehiscence<sup>@</sup>

Time Month	Commencement			Completion			
	12 Mid- night-2 AM	2-4 AM	4-6 AM	2-4 AM	4-6 AM	6-8 AM	8-10 AM
January	1	7	91	3	7	21	80
April	0	3	88	2	8	11	90
July	0	0	79	1	5	16	76
October	3	9	85	5	14	18	94
Average	1.0	4.75	85.75	2.75	8.50	16.50	85.0

<sup>@</sup> Per hundred flowers at two hours interval

Table 9. Stigma receptivity based on the association of insects

Time of flower collection	6-8 AM	8-10 AM	10-12 noon	12 noon-2 PM	2-4 PM	4-6 PM
Number of flowers examined	200	200	200	200	200	200
Number of insects found	17	23	58	131	46	8

Table 10. Stigma receptivity based on the percentage of fruit set at 2 hours interval

Time of polli- nation	6-8 AM		8-10 AM		10-12 noon		12 noon-2 PM		2-4 PM		4-6 PM	
No. of flowers pollinated	50		50		50		50		50		50	
	A	B	A	B	A	B	A	B	A	B	A	B
May	0	9	10	20	13	26	37	74	2	4	0	0
November	0	0	7	14	11	22	42	84	8	16	0	0
Average	0	0	8.5	17	12	24	39.5	79	5	10	0	0

A- Number of fruits set after two weeks

B -Setting per cent

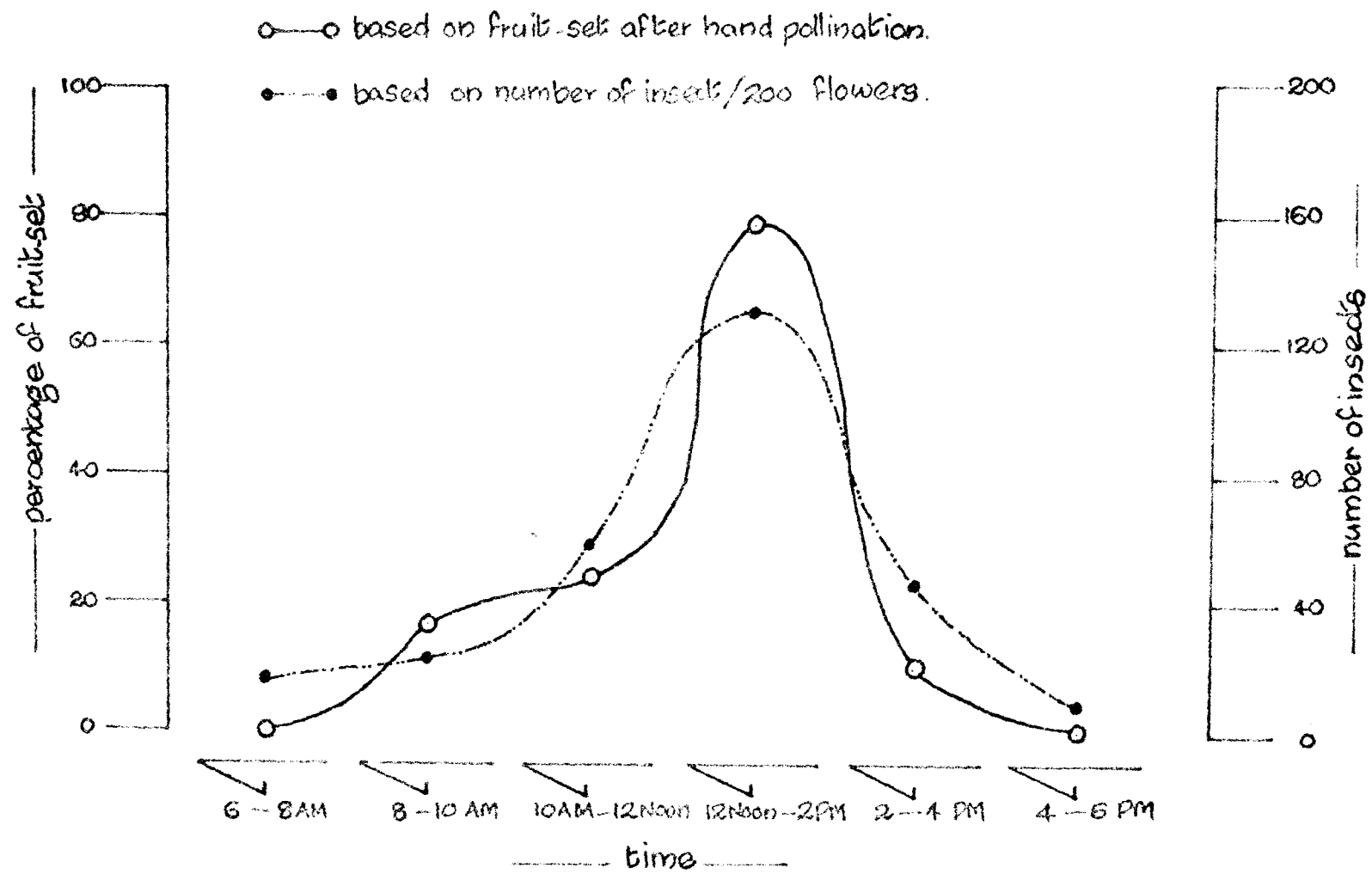


Fig. 5. — Stigma receptivity

### Size and shape of pollen grains

The pollen grains were found to be round. The palynological description are furnished in Table 11. The average diameter of pollen grains under dry, wet and stained conditions were 19.25, 21.75 and 19.35  $\mu$ , respectively.

### Estimation of pollen production

The data regarding pollen production are given in Table 12. Total number of pollen grains in a flower during the months of May and November were 6100 and 7150, respectively. The average pollen production per flower was 6625.

### In-vitro germination and growth of pollen

The pollen grains failed to germinate in water. They, however, germinated and exhibited good tube growth (Plate 3) in 5 to 20 per cent sucrose solution. Data regarding the composition of the media as well as the germination and tube growth are presented in Table 13. Highest germination and tube growth were obtained in the medium containing 0.5 per cent agar, 10.0 per cent sucrose and 1.0 per cent 100 ppm boric acid. In the medium consisting of 5.0 per cent agar, 50.0 per cent sucrose and 10.0 per cent 100 ppm boric acid, there was no germination, even after four hours.

Table 11. Diameter of the pollen grains ( $\mu$ )

	Dry	Wet	Stained
No. of pollen grains observed	100	100	100
Minimum	13.25	16.50	13.00
Maximum	23.65	25.75	24.13
Average	19.25	21.75	19.35

Table 12. Estimation of pollen production

Month	No. of flowers observed	Mean No. of pollen grains in	
		One anther	One flower
May	20	1220	6100
November	20	1430	7150
Average	20	1325	6625



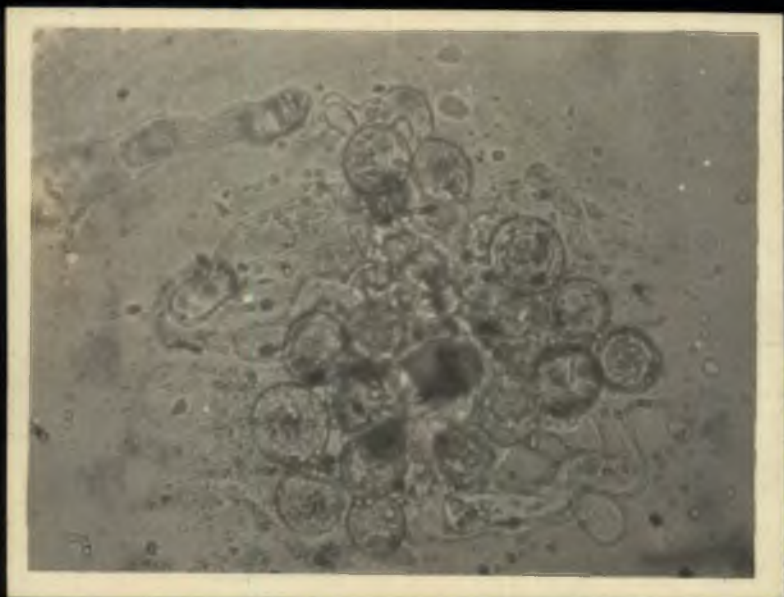


Plate 3. Germination of pollen grains in sucrose solution

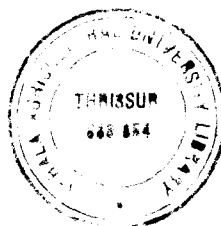


Table 13. In-vitro pollen germination and tube growth in media of different concentration of agar, sucrose and boric acid

Sl. No.	Concentration of the media (% agar, % sucrose and % 100 ppm boric acid respectively)	Germination (%)		Mean tube growth ( $\mu$ ) after 4 hours
		After 1 hour	After 4 hours	
1	0.1 + 5.0 + 0.5	8.30	20.27	32.18
2	0.5 + 10.0 + 1.0	68.74	84.31	238.56
3	1.0 + 15.0 + 2.0	30.80	48.20	70.13
4	2.0 + 20.0 + 5.0	11.31	15.75	20.35
5	5.0 + 50.0 + 10.0	0	0	-

### Effect of sucrose on pollen germination

Data on the effect of sucrose on pollen germination and tube growth are presented in Table 14 and illustrated in Fig. 6. Highest germination and tube growth were obtained in the medium with 10 per cent sucrose. There was no germination in the medium containing 30 per cent sucrose.

### Effect of Boron and calcium on pollen germination

The data presented in Table 15 and Figs. 7a, b and c indicated that calcium or boron improved the percentage of germination, when the concentration was increased from 50 ppm to 100 ppm. Further increases to 200 ppm brought about decrease in the percentage of germination. Similar effects were observed when boron and calcium in combination were applied. Similar trends were seen regarding pollen tube growth also.

### Effect of temperature on pollen germination

Data on the effects of temperature on pollen germination and tube growth are presented in Table 16 and illustrated in Fig. 8. Highest germination and tube growth were obtained at a temperature of 35°C. The germination was nil at both 10°C and 40°C.

Table 14. Effect of sucrose on pollen germination and tube growth

Sl. No.	Concentration of sucrose (%)	Germination after 4 hours (%)	Mean tube length after 4 hours ( $\mu$ )
1	5	21.30	104.51
2	10	92.56	230.12
3	15	54.21	185.00
4	20	32.18	132.23
5	30	0	-

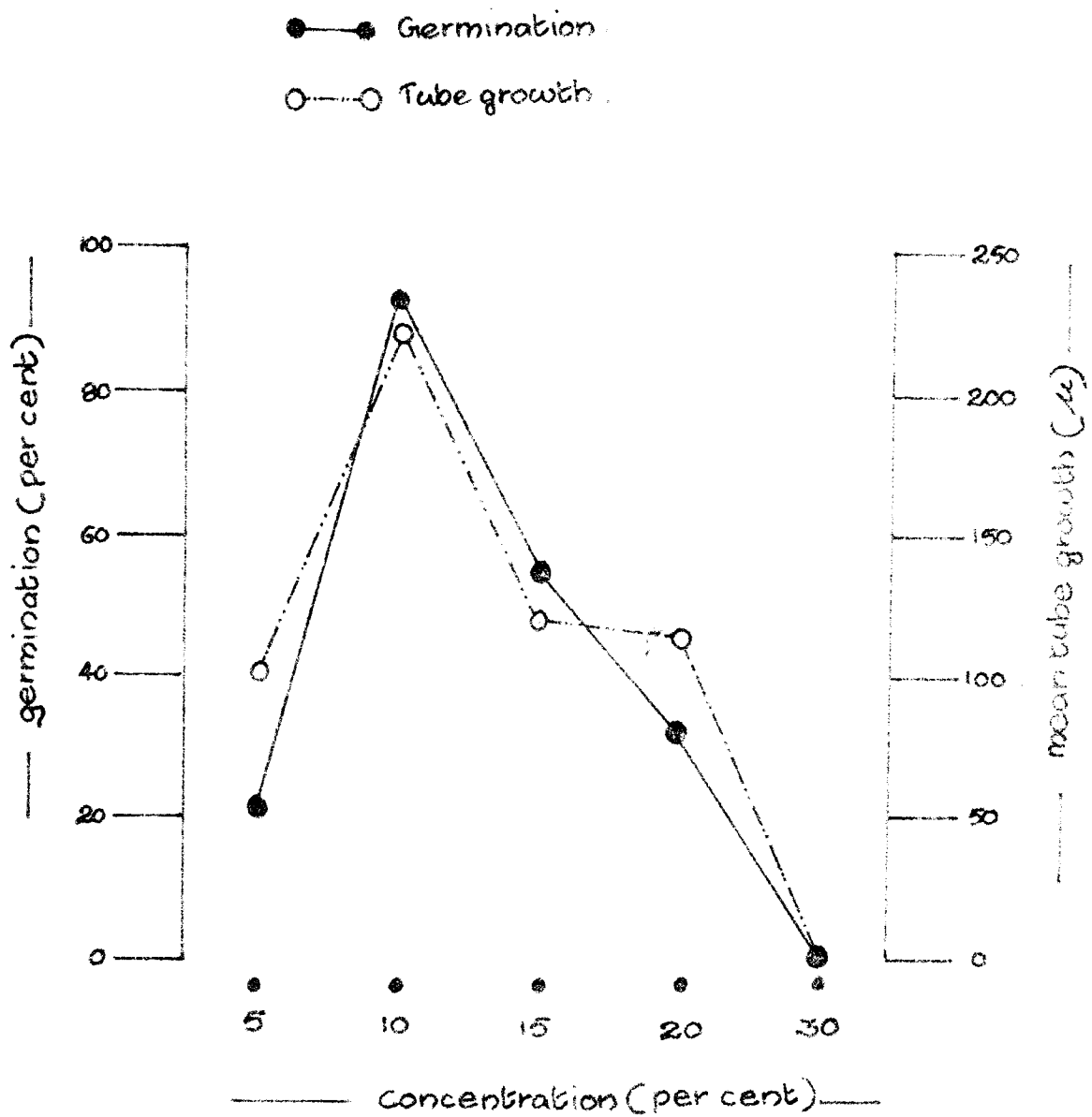
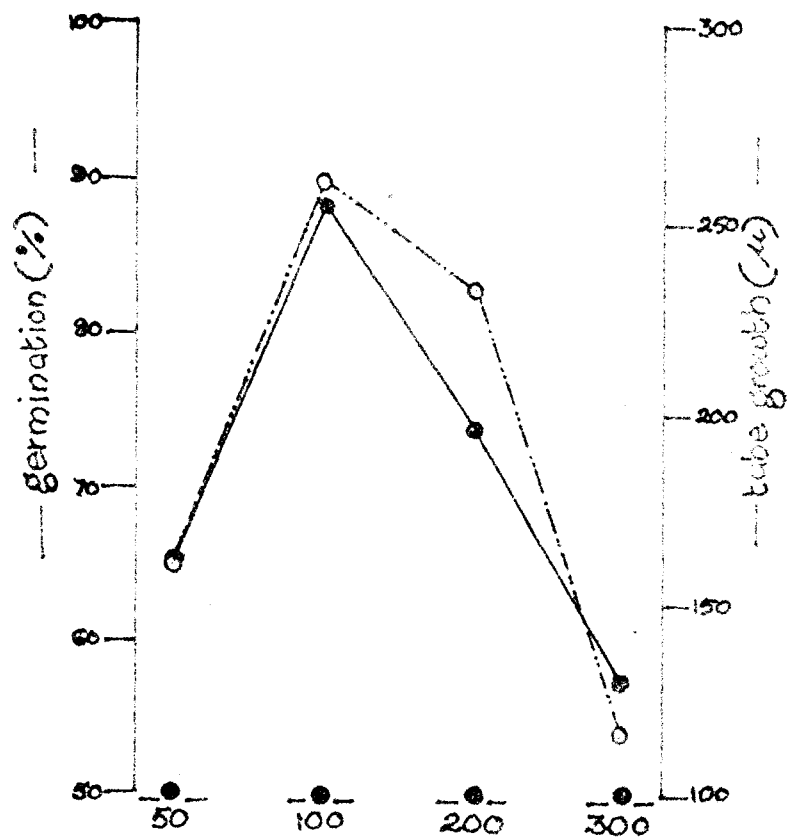


Fig.6. - Effect of sucrose on pollen germination & tube growth.

Table 15. Effect of boric acid and calcium nitrate on pollen germination and tube growth

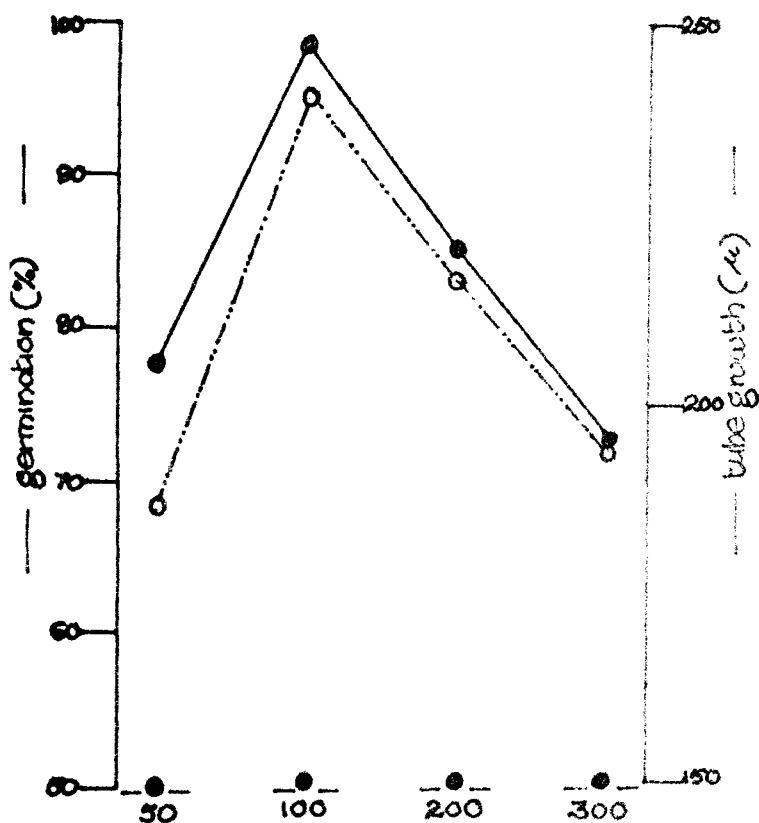
Sl.No.	Concentration (ppm)	Particulars after 4 hours	Boric acid	Calcium nitrate	Boric acid + calcium ni- trate
1	50	Germination (%)	65.20	68.40	76.70
		Mean tube length ( $\mu$ )	160.41	210.10	410.00
2	100	Germination (%)	90.35	95.55	97.82
		Mean tube growth	255.66	247.20	481.25
3	200	Germination (%)	83.15	83.21	91.10
		Mean tube growth	195.00	220.15	430.40
4	300	Germination (%)	54.82	72.50	81.65
		Mean tube growth	131.53	195.00	415.00



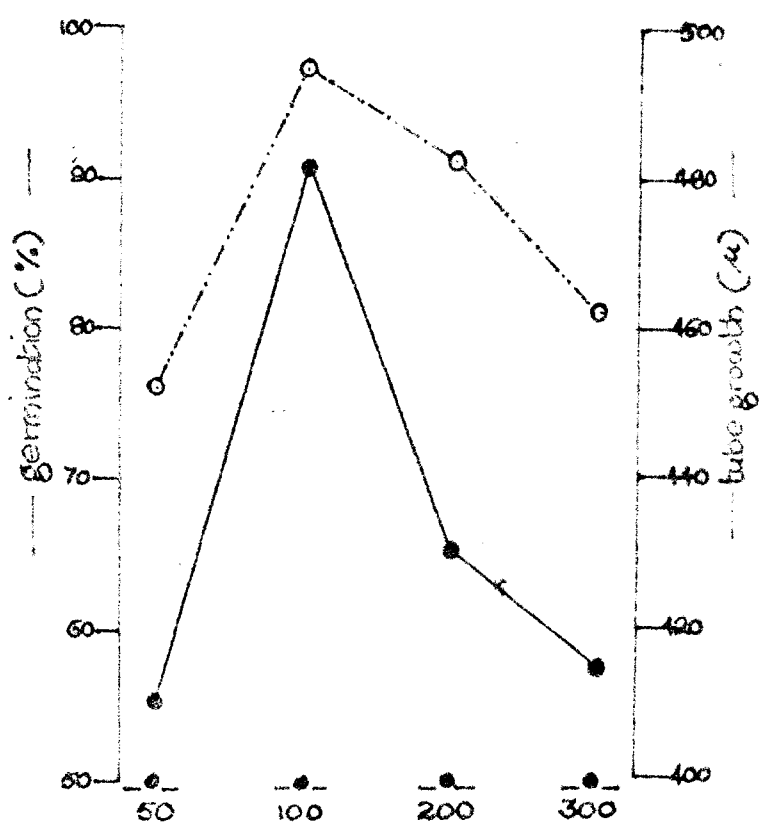
(a) boric acid (ppm)

○---○ Germination

●---● Tube growth



(b) calcium nitrate (ppm)



(c) boric acid + calcium nitrate (ppm)

Fig.7.- Effect of Boric acid & Calcium nitrate on pollen germination & tube growth

Table 16. Effect of temperature on pollen germination and tube growth

Sl.No.	Temperature (°C)	Germination after 4 hours (%)	Mean tube length after 4 hours (μ)
1	10	0	-
2	25	85.60	235.12
3	35	93.71	452.40
4	40	0	-



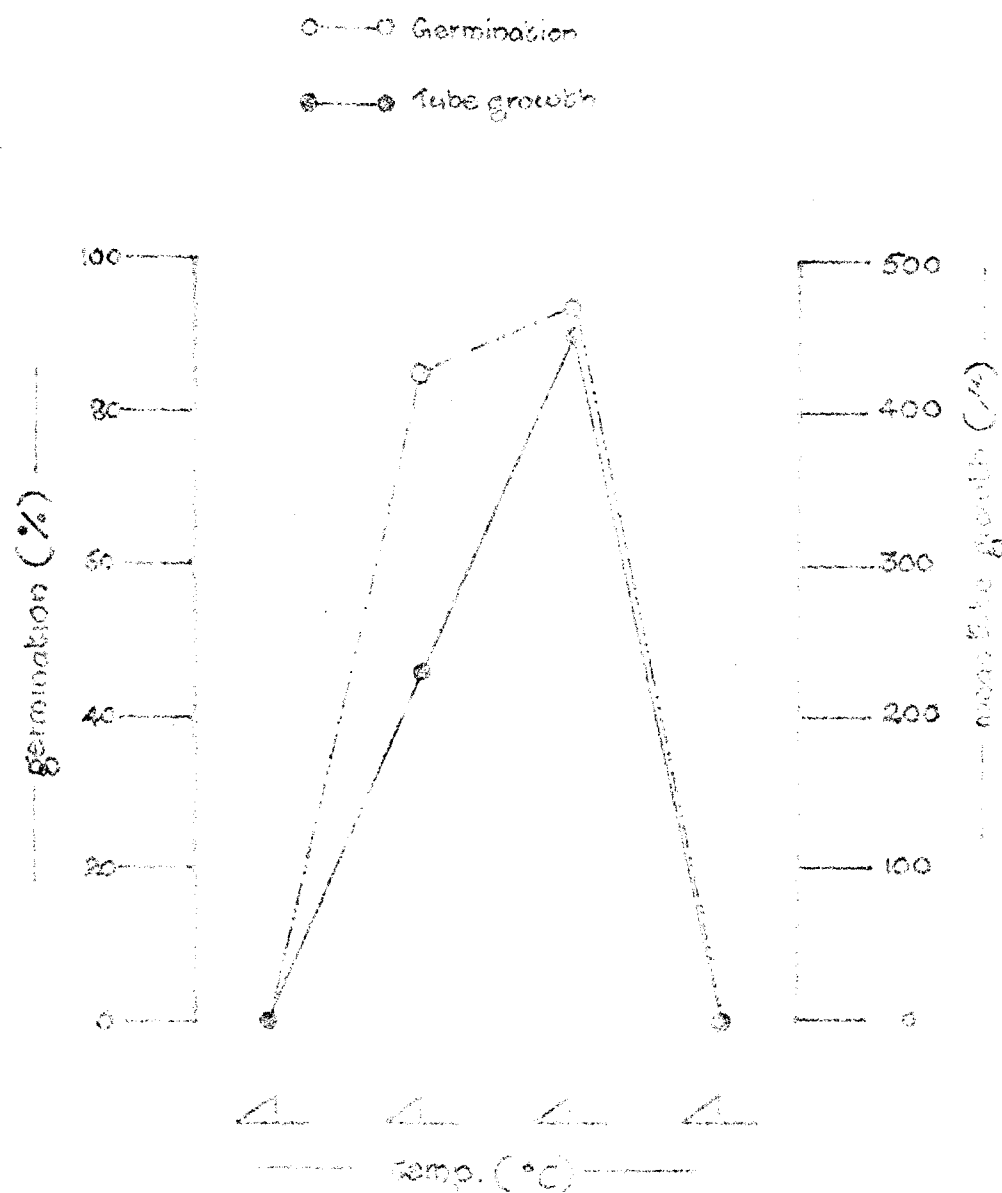


Fig. 8. Effect of temperature on pollen germination & tube growth.

### Pollen viability

The viability of pollen grains was assessed both by the acetocarmine staining method and by the germination method. The results are presented in Table 17. Viability readings in the germination method was found to be lower than those obtained in the staining method. The mean percentage viability assessed by the acetocarmine staining method and by the germination method were 97.10 and 66.25, respectively.

### Variation in the viability of pollen after anther dehiscence

The percentage germination and the mean tube growth of pollen collected at two hourly intervals from 6 AM to 6 PM are presented in Table 18.

Highest germination (96.41 per cent) was observed in the flowers collected at 8 AM. Tube growth was also highest (334.13  $\mu$ ) in the pollen grains collected at 8 AM. The data on germination and tube growth are shown in Fig. 9.

### Pollen storage

Pollen grains were stored by three methods and the data collected on their germination and tube growth have been presented in Table 19 and Figs. 10a and b.

Table 17. Pollen viability

Month	Percentage of viable pollen grains			
	Total number of flowers observed	Acetocarmine	Total number of flowers observed	Germi- nation
January	50	96.59	50	92.56
April	50	95.98	50	42.18
July	50	96.21	50	76.06
October	50	97.62	50	54.21
Average	50	97.10	50	66.25

Table 18. Variation in the viability and tube growth after anther dehiscence.

Time of flower collection	6 AM	8 AM	10 AM	12 noon	2 PM	4 PM	6 PM
No. of flowers observed	35	35	35	35	35	35	35
Mean germination after 4 hours (%) <sup>@</sup>	52.56	96.41	94.25	82.01	80.71	69.04	58.32
Mean tube length after 4 hours (u) <sup>@</sup>	110.00	334.13	327.39	221.38	191.13	169.13	138.88

<sup>@</sup> Totally from thirtyfive fields

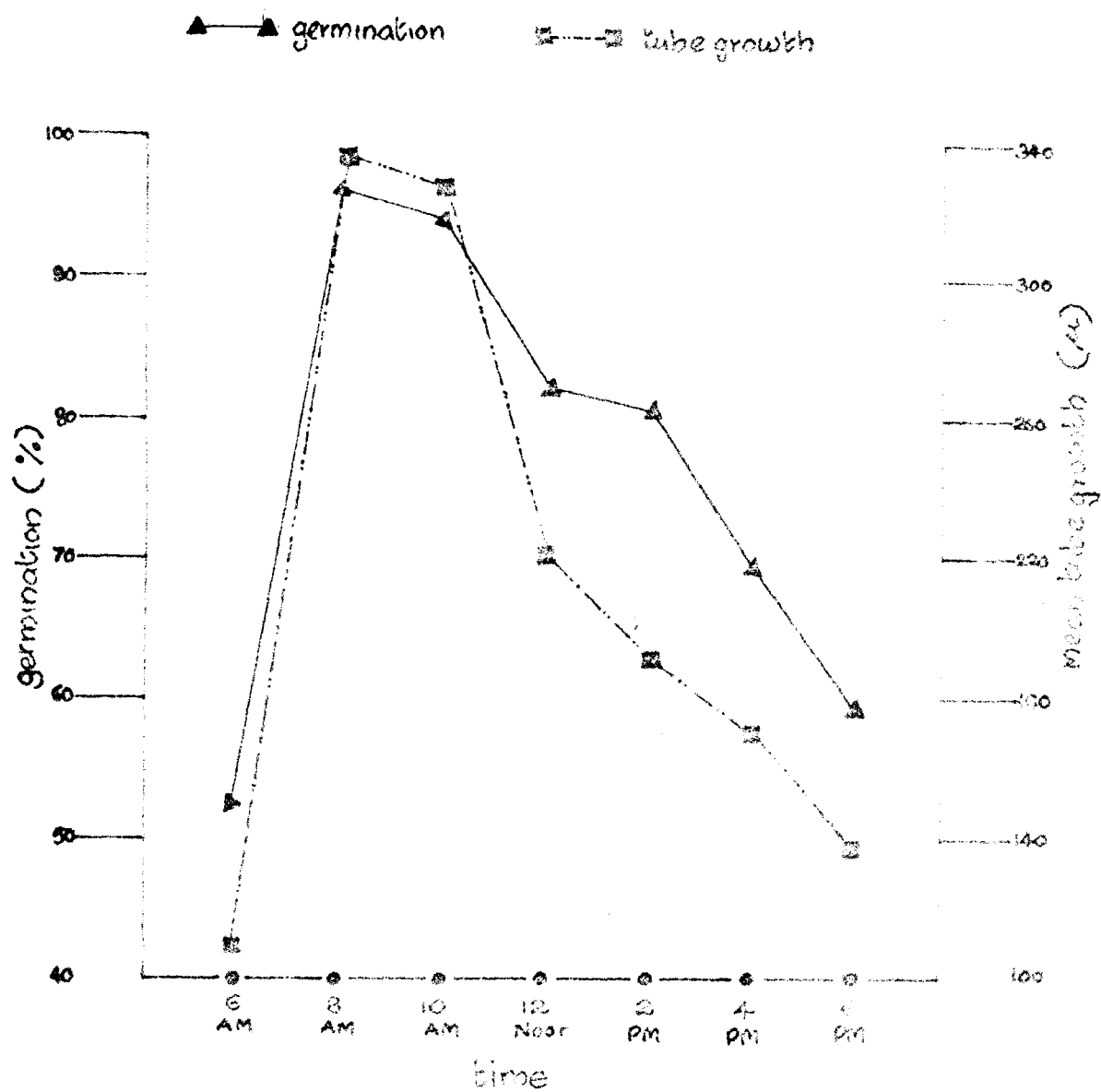


Fig. 9.—Pollen germination & tube growth after anther dehiscence.

Table 19. Pollen storage

Method <sup>++</sup> No.	Particulars <sup>+</sup>	Storage period								
		1-day	2-day	3-day	4-day	5-day	6-day	7-day	8-day	9-day
I	Germination (%)	86.70	60.42	51.75	21.62	2.56	0	-	-	-
	Mean tube growth ( $\mu$ )	220.31	164.06	101.85	60.12	21.33	0	-	-	-
II	Germination (%)	92.16	87.04	84.53	78.70	72.55	48.22	13.35	6.71	0
	Mean tube growth ( $\mu$ )	310.39	286.10	240.67	206.11	162.54	112.13	48.20	25.62	0
III	Germination (%)	41.30	7.26	0	-	-	-	-	-	-
	Mean tube growth ( $\mu$ )	92.04	21.57	0	-	-	-	-	-	-

+ Mean from twenty fields

++ I Anthers in tissue paper cover, wrapped with filter paper, inserted in glass tube, sealed and stored at room temperature after two hours of desiccation.

II Anthers in tissue paper cover, wrapped with filter paper, inserted in glass tube, sealed and stored at 5 to 10°C after two hours of desiccation.

III Kept on wet filter paper inside a petri-dish at room temperature (control).

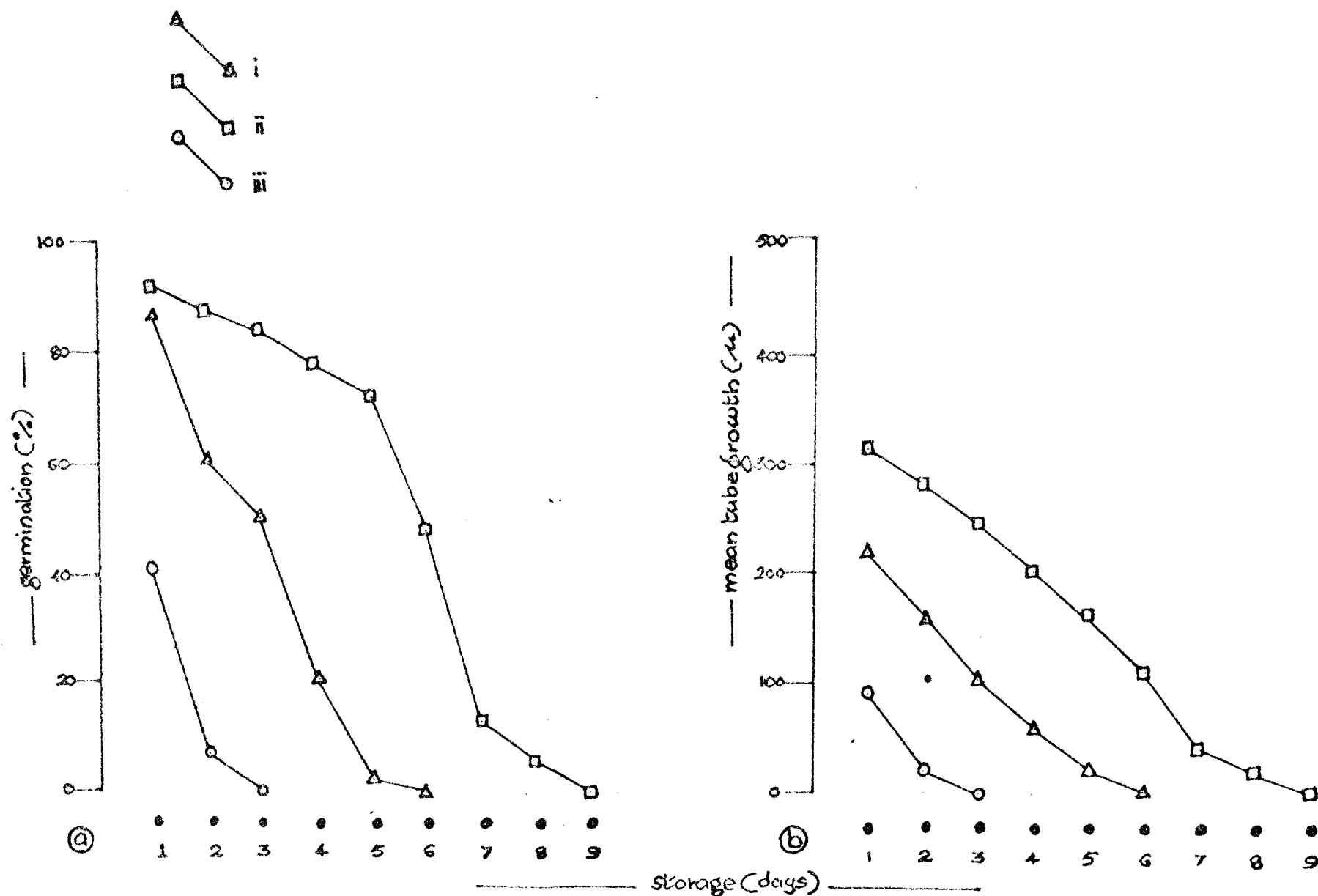


Fig. 10. Pollen germination and tube growth Vs storage methods.

In the first method, the germination after one day of storage was 86.70 per cent with a tube growth of 220.31  $\mu$ . Germination and tube growth gradually reduced. After storage for six days germination was found to be nil.

In the second method, after a day's storage, the germination and tube growth were 92.16 per cent and 310.39  $\mu$ , respectively. After nine days of storage, germination was reduced to zero.

In the third method, after one day of storage, the germination and tube growth were 41.30 per cent and 21.57  $\mu$ , respectively. After three days of storage the germination was nil.

### Pollination aspects

Extent of self pollination: Flowers which were bagged did not set fruit. They were shed 24 to 48 hours after the anthesis. It was also seen that no pollen grains were present on the style or stigma of drooped flowers.

Natural pollination rate: The rate of natural pollination from January to December are presented in Table 20 and illustrated in Fig. 11. Number of pollinated flowers were more in the months of January, February and March. The percentage of successful pollination was found to be higher



Table 20. Natural pollination rate for the year (1980)

Month	Groups	Non-pollinated (%)	Pollination		
			Total Pollinated (%)	Under pollinated (%)	Successfully pollinated (%)
January		59.87	40.13	28.93	11.20
February		60.75	39.25	26.44	12.81
March		59.00	41.00	32.00	9.00
April		64.72	35.28	28.18	7.10
May		68.95	31.05	25.70	5.35
June		67.47	32.53	27.41	5.12
July		68.72	31.28	23.03	8.25
August		68.33	31.67	25.27	6.40
September		74.70	25.30	20.58	4.72
October		76.00	24.00	20.95	3.05
November		73.70	26.30	21.80	4.50
December		73.88	26.12	18.37	7.75

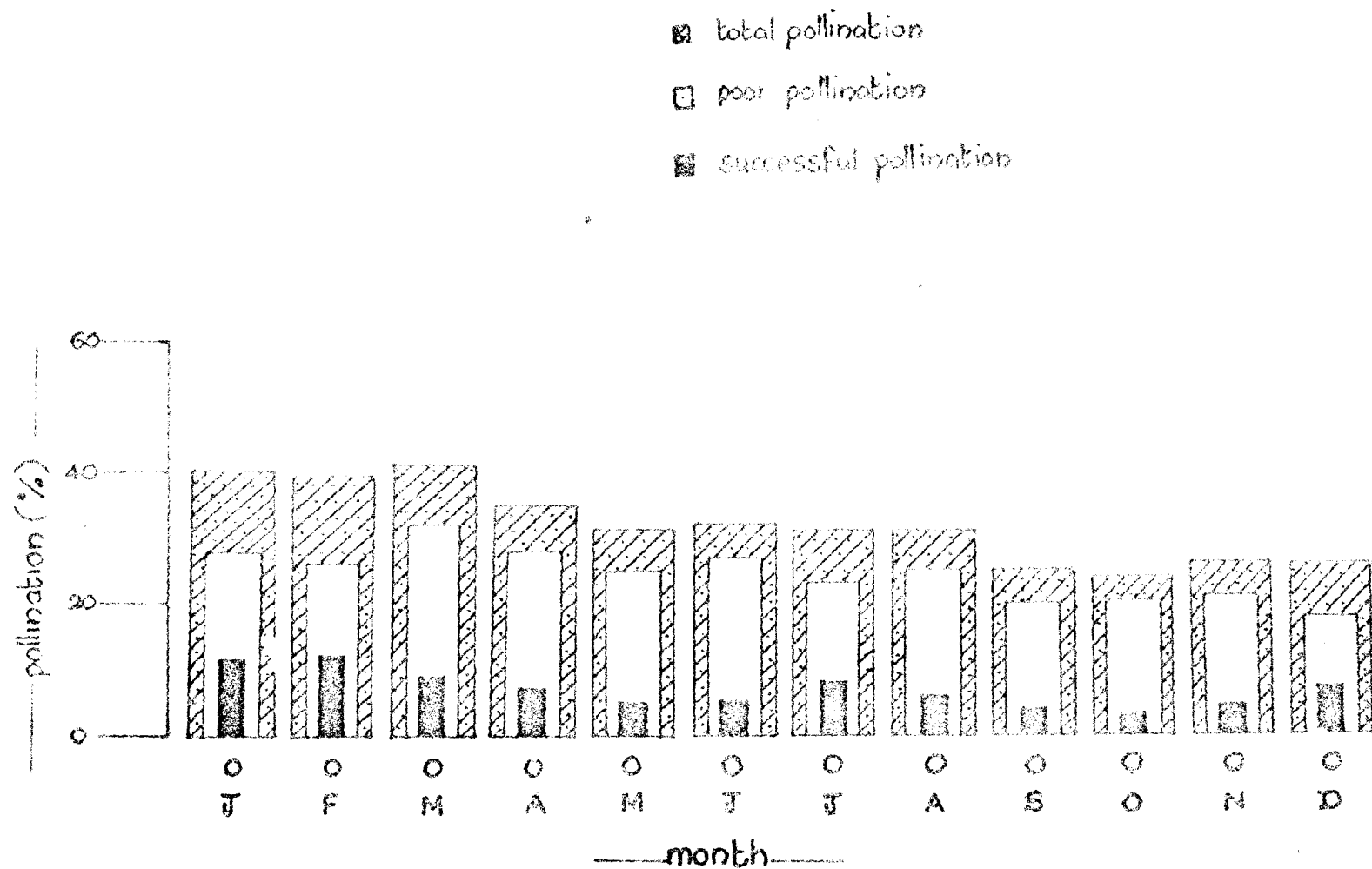


Fig. 11. MONTHLY POLLINATION RATE.

in January (11.20) and February (12.81). Number of under-pollinated flowers were more in March.

Insects associated with cocoa flower: Seven insects belonging to the order Diptera and five Formicid species (order Hymenoptera) were identified as common floral visitors (Table 21). Microphotographs of these insects have been displayed in Plates 4 to 9.

Compatibility status

Data regarding the number of flowers pollinated, percentage set and pod harvest resulting from self pollination are presented in Table 22 and those resulting from cross pollination in Table 23.

Out of the 15 plants self-pollinated, only four were found to be self-compatible (Table 22). The plant J 7/3 ranked first with regard to fruit set (67.69 per cent) and second for pod harvest (29.23 per cent). The plant L 9/3 ranked first in pod harvest (6.0 per cent) and second in percentage fruit set (67.31). The plant J 11/2 was third, both in fruit set and pod harvest.

The fifteen plants, when cross pollinated among themselves, recorded good percentage of fruit set and pod harvest. The plants which were identified as self-compatible,

Table 21. Insects associated with cocoa flowers

Sl.No.	Name	Family	Order
1	Unidentified Psychodid	Psychodidae	DIPTERA
2	Unidentified Cecidomid-1	Cecidomidae	"
3	Unidentified Cecidomid-2	"	"
4	<u>Forcipomyia</u> sp.	Ceratopogonidae	"
5	Unidentified Chironomid	Chironomidae	"
6	Unidentified Drosophilid-1	Drosophilidae	"
7	Unidentified Drosophilid-2	"	"
8	Unidentified Myrmecin	Formicidae	HYMENOPTERA
9	<u>Cremastogaster</u> sp.	"	"
10	<u>Plageolepis longipes</u>	"	"
11	<u>Prenolepis</u> sp.	"	"
12	<u>Camponotus compressus</u>	"	"



Plate 4. Insects associated with cocoa flowers-  
 Left: Unidentified psychodid x 320  
 Right: Unidentified chironomid x 320



Plate 5. Insects associated with cocoa flower -  
 Top: Farsia sp. x 320  
 Bottom: Unidentified cecidomid x 320



Plate 6. Insects associated with cocoa flower -  
unidentified Drosophilid  $\times 320$



Plate 7. Insects associated with cocoa flower -  
Left: Cremastogaster sp.  $\times 200$   
Right: Unidentified Myrmecia  $\times 200$



Plate 8. Insects associated with cocoa flower -  
 Left: Plagioclepis longipes x 200  
 Right: Frenoclepis sp. x 200



Plate 9. Insects associated with cocoa flower -  
Camponotus compressus x 200

Table 22. Self pollination and compatibility

Sl. No.	Plant No.	Number of flowers pollinated	Number of fruit set	Percentage of fruit set	Number of pods harvested	Percentage of pod harvest	Rank		Compatibility status <sup>@</sup>
							Fruit set	pod harvest	
1	J 2/3	62	2	3.23	0	-	9	-	si
2	J 2/5	50	0	-	-	-	-	-	si
3	J 4/2	50	0	-	-	-	-	-	si
4	J 4/6	65	1	1.54	0	-	11	-	si
5	J 7/3	65	44	67.69	19	29.23	1	2	sc
6	J 7/4	51	2	3.92	0	-	8	-	si
7	J 8/5	73	5	6.85	0	-	6	-	si
8	J 9/8	71	0	-	-	-	-	-	si
9	J 11/2	58	32	55.17	14	21.14	3	3	sc
10	J 11/3	55	0	-	-	-	-	-	si
11	L 84/1	60	3	5.00	0	-	7	-	si
12	L 33/3	56	1	1.79	0	-	10	-	si
13	L 9/3	52	35	67.31	21	60.00	2	1	sc
14	L 52/4	50	23	46.00	10	20.00	4	4	sc
15	L 52/7	68	7	10.29	1	1.47	5	5	si

<sup>@</sup> si - self-incompatible  
 sc - self-compatible



Table 23. Cross pollination and compatibility

Sl.No.	Female plant	Male plant	No.of flowers polli-nated	No.of fruit set	Fruit set (%)	No. of pods har-vested	Pod har-vest (%)	Rank		Compatibi- lity status
								Fruit set	Pod harvest	
1	J 2/3	L 52/7	55	33	60.00	7	12.73	6	14	cc
2	J 2/5	L 52/4	51	21	41.18	10	19.61	10	11	cc
3	J 4/2	L 9/3	63	50	79.37	15	23.81	1	8	cc
4	J 4/6	L 33/3	72	48	66.67	31	43.06	3	1	cc
5	J 7/3	L 84/1	50	23	46.00	11	22.00	9	9	cc
6	J 7/4	J 11/3	50	32	64.00	20	40.00	5	3	cc
7	J 8/5	J 11/2	56	20	35.71	6	10.71	14	15	cc
8	J 9/8	J 2/3	65	24	36.92	16	24.62	12	6	cc
9	J 11/2	J 2/5	71	26	36.62	20	28.17	13	4	cc
10	J 11/3	J 4/2	51	19	37.25	7	13.73	11	13	cc
11	L 84/1	J 4/6	58	33	56.90	12	20.69	7	10	cc
12	L 33/3	J 7/3	63	41	65.08	27	42.86	4	2	cc
13	L 9/3	J 7/4	61	41	67.21	16	26.23	2	5	cc
14	L 52/4	J 8/5	51	24	47.06	9	17.65	8	12	cc
15	L 52/7	J 9/8	50	16	32.00	12	24.00	15	7	cc

& cc - cross compatible

showed satisfactory fruit set and pod yield in cross pollination. The average fruit set ranged from 32.0 (L 52/7) to 79.37 (J 4/2) per cent. The percentage of pod harvest ranged from 10.71 (J 8/5) to 43.06 (J 4/6).

#### Natural vs hand pollination

The difference in fruit set and pod yield under natural pollination and on hand pollination are summarised in Table 24.

Out of the 600 flowers under natural pollination, only 71 set fruits. Ultimately only five pods were carried to maturity. Thus, the percentage of fruit set and pods harvest were 11.81 and 0.83, respectively. However, on hand pollination, 451 flowers out of the 867 set fruits and 219 of them were carried to maturity. Thus, the percentage of fruit set and pod harvest worked out to 52.02 and 25.76, respectively.

#### Flowering and fruiting behaviour on the main trunk and fan branches

Data on the number of flowers produced per cushion, flowers pollinated, fruit set, cherelles wilted and pods harvested are presented in Table 25.

The mean number of flowers produced per cushion on the main trunk was 37.0. Of these, 37.29 per cent flowers

Table 24. Natural vs Hand pollination

	Natural	Hand
No. of flowers labelled	600	867
No. of fruit set	71	451
Percentage of fruit set	11.81	52.02
No. of pods harvested	5	219
Percentage of pods harvested	0.83	25.26

Table 25. Flowering and fruiting behaviour on the main trunk and on fan branches

Plant part	Total number of cushions observed	Mean number of flowers/ cushion at a stretch	Number pollina- ted	Number set	Number wilted	Number harvested
Main trunk	20 <sup>+</sup>	37	13.80	5	3.55	1.45
Per cent	-	-	37.29	13.51	9.59	3.91
Fan branches	20 <sup>+</sup>	18.90	6.70	1.20	1.15	0.05
Per cent	-	-	35.44	6.34	6.08	0.26

+ In ten trees @ two cushions per main trunk and two cushions per fan branch.

were seen pollinated. The percentage of fruit set was 13.51. 9.59 per cent of the total number of flowers were lost due to cherelle wilt and only 3.91 per cent reached the harvestable stage.

In the case of fan shoots, the mean flower production per cushion was only 18.90. 35.44 per cent of these flowers were found to be pollinated. The fruit set worked out to 6.34 per cent. The loss due to cherelle wilt was 6.08 per cent and only 0.26 per cent of the total flowers reached the harvest.

#### Rhythm of flower production in cushions

As can be seen from the data presented in Table 26, the interval between flower production on the cushions ranged from 33.81 days to 165.30 days. Those cushions that supported the developing pods upto the harvestable stage flowered less frequently than those which exhibited no flower set or complete wilting of cherelles.

#### Fruit (pod) development

The weekly developmental stages of the pod (from fruit set to ripening) are depicted in plates 10, 11 and 12.

Data on length and thickness of the fruit-stalk, length and thickness of the pods, thickness of the shell (rind) as

Table 26. Type of flower production in the cushion based on its Rhythm - mean table

Type	Number of flowers produced at a stretch <sup>@</sup>	Number of fruit set	Number of fruit wilted	Number of pods har- vested	Duration between flower produc- tion (days)
I	9.76	0	-	-	33.81
II	22.37	5.62	5.62	0	65.31
III	15.12	2.41	1.24	1.17	165.30
IV	6.0	1.76	0	1.76	161.73

<sup>@</sup> Mean of forty cushions



Plate 10. Stages of fruit development upto seventh  
day after fertilisation

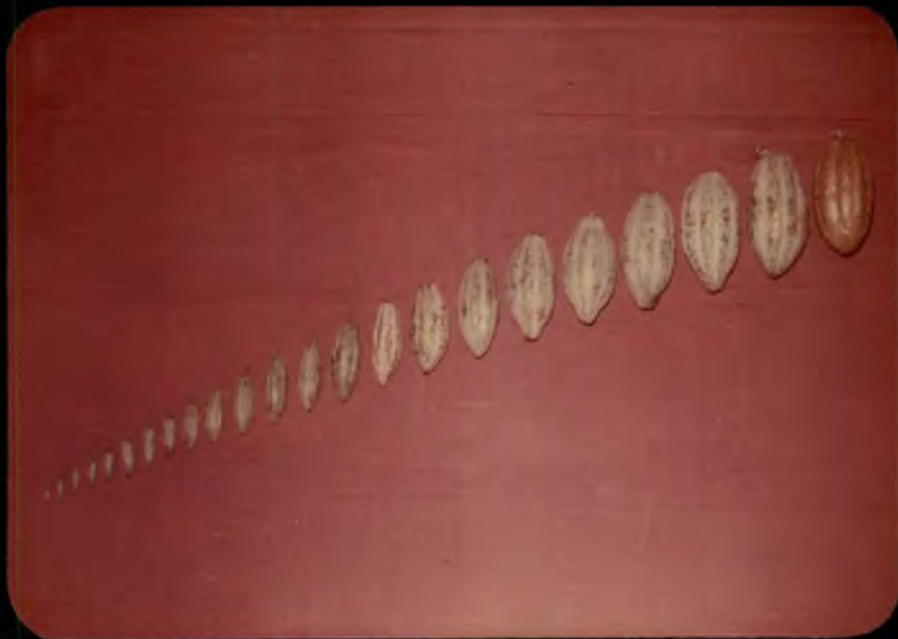


Plate 11. Stages of fruit development - one week after fertilisation to ripening

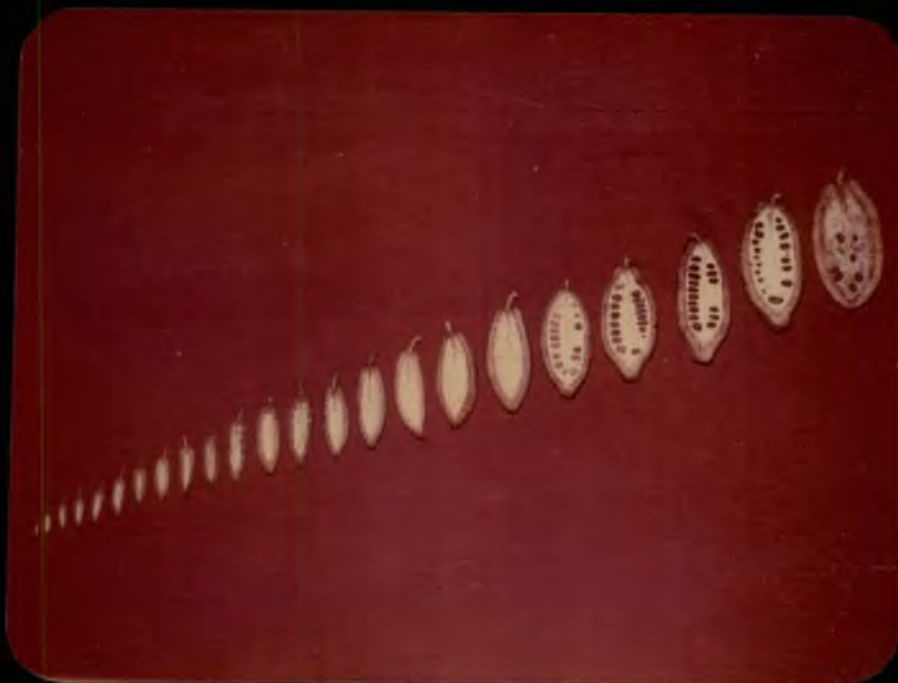


Plate 12. Stages of fruit development - longitudinal section



well as percentage of cherelles wilted were collected at weekly intervals (Table 27a). The mean length of the fruit-stalk, after one week of growth, was 15.06 mm and after 21 weeks, (25.63 mm). The mean thickness increased gradually from 1.51 mm to 10.33 mm during the corresponding period. There was no further increase in the length and thickness of the fruit-stalk during the 22nd week. The mean length and thickness of the pods increased gradually from 20.38 mm and 7.81 mm (after one week) to 160.10 mm and 85.43 mm (after 21 weeks), respectively. During the 22nd week, there was no further increase in the length of the pod while the thickness exhibited a slight reduction. The thickness of the shell gradually increased from 2.31 mm during the first week to 12.52 during the 20th week. Thereafter, the shell thickness showed a slight reduction.

Examination of the longitudinal sections of the developing pods showed that the differentiation was very slow upto four weeks. After six or seven weeks, seed formation was visible. By the 15th week, both the shell and the fruit-stalk started to harden. This was followed by the clear appearance of the cotyledons. The seed coats started to harden by the 17th week. By the 18th week, the cotyledons took up light pink colour followed by the hardening of the seed. The colour of the cotyledons changed to dark pink. By the

Table 27a. Stages of fruit/pod development at weekly intervals  
(mean of 100 pods)

Number of weeks after fertilization	Fruit-stalk length (mm)	Fruit-stalk thickness (mm)	Fruit/pod length (mm)	Fruit/pod thickness (mm)	Shell thickness (mm)	Percentage of cherelle wilt (percentage to the total)
1	15.06	1.51	20.38	7.81	2.31	6.25
2	15.71	2.20	22.71	8.60	2.53	6.25
3	16.20	2.62	24.52	10.63	3.65	8.75
4	17.15	2.85	30.98	11.91	4.20	13.75
5	17.70	3.14	38.25	12.03	4.56	15.0
6	18.54	3.27	45.57	14.71	4.82	18.75
7	20.68	3.69	50.03	16.53	5.12	12.50
8	20.93	4.00	55.68	18.94	5.35	8.75
9	22.07	4.27	65.14	20.75	5.81	8.75
10	22.38	4.72	75.78	25.21	6.52	1.25
11 @	22.91	5.13	85.51	30.54	7.03	-
12 @	23.81	5.48	90.40	32.75	8.10	-
13 @	24.31	5.75	95.35	33.07	8.31	-
14 @	24.68	6.20	100.81	35.31	8.67	-
15 @	25.00	6.61	110.62	36.85	8.91	-
16 @	25.17	7.23	120.03	45.62	10.05	-
17 @	25.25	7.41	130.91	50.01	10.41	-
18 @	25.32	7.63	135.46	55.43	10.80	-
19 @	25.40	7.81	140.82	65.98	12.15	-
20 @	25.55	8.52	150.31	75.30	12.52	-
21 @	25.63	10.30	160.10	85.43	12.52	-
22 @	25.63	10.30	160.10	85.12	11.83	-

@ Mean of 60 pods

Table 27b. Number of days for pod maturity

Number of days from ferti- lization			Daily mean temperature in °C	Calculated days by Alvin's formula
Minimum	Maximum	Average		
127	141	138.17	27.04	138.58

22nd week, the pods were ripe.

Data on the occurrence of cherelle wilt during various stages of fruit development are also presented in Table 27a. One-week old cherelles recorded 6.25 per cent wilt. The percentage of cherelle wilt gradually increased and reached a peak of 18.75 per cent at the sixth week. Afterwards, the percentage of wilt reduced gradually. Beyond the 10th week, there was no loss due to cherelle wilt.

Cocoa pods took 127 to 141 days (mean 138.17 days) for reaching the ripening stage (Table 27b). The figure worked out by the formula of Alvim et al. (1972) came to 138.58 days.

## *Discussion*

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## DISCUSSION

The results of the studies conducted at the Regional Research Station, Pilicode on the various aspects of floral biology, fruit set and fruit development in cocoa are discussed hereunder:

### Flowering pattern

The present studies revealed that under North Kerala conditions, cocoa flowered throughout the year. As in the other tropical cocoa growing countries, two peaks in flowering were observed, namely, May-June and November-December, the latter being the major season. Flower production in South Karnataka has also been reported to be more in December than in the other seasons (Anon, 1977). However, in Ghana (Newison and Ababio, 1929) and in Cuba Delpinalrivero and Acunagale, 1967), major flowering season has been reported to be April-June and June-September, respectively. The low rate of flower production during the May-June season may be due to the competition for food between the differentiating flower buds and the new vegetative flush. Larger flower production observed during November-December season may be on account of the extra food reserves resulting from the May-June flush.

It was observed in the present studies that flower production was minimum during September. Alvim (1966) also found July-September period to be the non-flowering period in Bahia. The lowest flower production was observed in South Karnataka during August-September season (Anon, 1977).

According to Murray (1975), a cocoa plant can produce more than 10,000 flowers annually. The annual flower production observed in the present studies was 12,294. The higher temperature conditions of North Kerala must have been responsible for this heavy flower production. Wood (1975) reported that flowering may be more under higher temperature conditions.

#### Pattern of cropping

As can be expected from the observed double peaked flowering pattern, there were two harvest peaks also - one during June-August and the other during October-December. Chat (1953), Alvim (1974), Purseglove (1974), Wood (1975) and Shanmugavelu and Rao (1977) reported that the pattern of cropping in cocoa is double peaked. In the present studies, a heavy crop (about 75% of the total) was harvested during the June-August period and a light one (a little over

20 per cent) in October-December season. Owing to the intense flowering observed in November-January season, higher pod production can be expected during June to August, under Kerala conditions. The low pod production observed in the October-December season may be due to two factors, the comparatively low flower production during May-June as well as the depletion of food reserves by the newly formed leaves and the fruits developing from the November-December flowering.

#### Intensity of cherelle wilt

Cherelle wilt is considered to be one of the factors affecting the yield of cocoa (Couprie, 1972). The wilting of cherelles recorded throughout the year in the present studies support the above statement. Cherelle wilting has been stated to be a physiological thinning mechanism of the plant by which the size of the crop is regulated with the available food reserves in the tree (Nichols, 1961 and Murray, 1975). In the present studies, higher intensity of cherelle wilt was observed during June-July, the season of higher pod production. The competition for the nutrients between the developed and the newly developing fruits (pods) may have been responsible for the wilt of the latter.



### Flower bud development

Results obtained in the present studies revealed the flower bud development in cocoa to be a slow process. It took about 22 days for the buds which appeared on the cushions to reach maturity or anthesis stage. Compared to the bud portion, the pedicel showed a higher rate of growth initially, thus giving a proportionately long pedicel for the small buds.

### Anthesis

The present studies indicated that under North Kerala condition, anthesis of the flowers commenced between 2 and 4 PM. Wellensiek (1932) observed that anthesis commenced around 4.30 PM in Java. Sampayan (1963 and 1966) found the anthesis to be slightly earlier in the Philippines, (at 4 PM). However, Delpinalrivero and Acunagale (1967) reported that in Cuba, the anthesis commenced around 7 PM. This variation may be probably due to the variations in the diurnal variations in temperature and/or relative humidity in these places. An analysis of the geographical location of these countries indicated the possibility of latitude effects linked with environmental conditions on the time of anthesis. The present studies also revealed that during the rainy days, the flower buds commenced opening early in the day, around 10.00 AM to 12.00 noon.

Regarding the completion of the anthesis, Sampayan (1963 and 1966) reported that it was around 6 PM on the same day in the Philippines. The present studies showed that anthesis in cocoa was a slow process, commencing between 2 and 4 PM and completing between 2 and 4 AM, the next day.

#### Anther dehiscence

Observations of the present studies revealed that in majority of the flowers (85.75 per cent), anther dehiscence commenced between 4 and 6 AM and completed between 8 and 10 AM. Findings of Wellensiek (1932) that anther dehiscence commenced at 6.30 AM and of Sampayan (1963 and 1966) that the dehiscence commenced at 6 AM and completed at 8.30 AM support the present observations.

#### Stigma receptivity

It may be recalled that the anthesis was found to be completed between 2 and 4 AM. Based on the adherence of pollen grains to the stigma, the stigma was found to be receptive between 6 AM and 6 PM. Visit by insects was found to be intense between 12 noon and 2 PM. Data on the percentage of fruit-set on hand pollination also revealed that in most of the flowers, the receptivity was very high

between 12 noon and 2 PM. Sampayan (1963 and 1966) observed high receptivity of stigma between 10 AM and 1 PM in the Philippines based on the results of hand pollination.

Wellensiek (1932) reported that the flowers were protogynous and the stigma was receptive when they opened (4.30 PM). However, the present studies as well as the reports of Sampayan (1963 and 1966) revealed that the stigma receptivity was between 10 AM and 2 PM, the day after the commencement of anthesis. Thus, the protogynous nature of cocoa flower has not been confirmed.

#### Palynological aspects

The study revealed that the pollen grains of cocoa were round and had a diameter between 14.25 to 24.51  $\mu$ . Flowers produced during November had more pollen/flower than the those produced during May. It may be recalled that November was identified as the peak flowering season for cocoa in Kerala. The higher pollen production seen during November would have effected better pollination of the flowers, resulting in a heavier crop during June-August season.

Cocoa pollen did not germinate in water. Good pollen germination and tube growth was obtained in a medium containing 0.5% agar, 10% sucrose and 1% of 100 ppm boric acid.

Ostopenko (1956), Vasil (1960), Singh (1961) and Singh et al. (1961) reported that sucrose was helpful as an artificial medium for germination of pollen grains as it served as a nutrient during pollen tube growth. Varas and Soria (1962), Jacob et al. (1969), Ravindran (1977) and Martinson (1973) reported good results by using sucrose concentration ranging from 5 per cent to 15 per cent and 2.0 per cent agar.

Both boron and calcium enhanced the germination and pollen tube growth in the present studies. In general, the observations are in agreement with the findings of Shapiro and Burdick (1961), Varas and Soria (1962) and Ravindran (1977). In the present studies, 100 ppm of both boric acid and calcium nitrate (1:1) gave good pollen germination and tube growth.

As far as the effects of temperature on pollen germination and tube growth were concerned, the present studies revealed that there was no germination at 10°C and 40°C. Best germination and tube growth were observed at 35°C. Ravindran (1977) also had reported better germination at 35°C.

The estimate of viability of pollen grains by the germination method was found to be lower than that by aceto-carmin method. These observations confirmed the view that

the acetocarmine method was less accurate in determining the viability of pollen grains, as recorded in fruits and vegetables by Bajpai and Lal (1958). Further, all the pollen grains which appeared as viable on acetocarmine staining may not germinate in artificial medium (Parameswar, 1974). In the present studies, cocoa pollen from freshly opened flowers exhibited good germination between 6 AM and 6 PM. However, pollen germination was found to be high between 8-10 AM.

Out of the three methods of pollen storage tried, keeping pollen in tissue paper packets, sealing in glass tubes and storing the tubes in refrigerator after two hours of desiccation (over calcium chloride) was found to be superior to the other methods. Even after five days of storage by this method, 72.55 per cent of germination and 112.13  $\mu$  pollen tube growth were obtained. Keeping pollen grains in sealed glass tubes (Wellensiek, 1932), desiccator (Varas, 1962) and under refrigeration (Simmons, 1976) have been reported to extend the viability.

#### Pollination aspects

In the present studies, bagged flowers did not set any fruits and they were shed within 24 to 48 hours after the anthesis. Purselove (1974) and Murray (1975) reported that unpollinated flowers abscise within 24 hours. This

observation and the absence of pollen grains on the stigma or style of the abscised flowers led to the conclusion that in order to achieve pollination, involvement of an external agent was necessary.

The present studies revealed that several insects were associated with the pollination of flowers in cocoa. Even-though, Wellensiek (1932) could not observe pollination by insects in Java, Dessart (1961), Cuatrecasas (1964), Purseglove (1974) and Murray (1975) recorded involvement of insects in pollination of cocoa.

Seven insects of the families Psychodidae, Cecidomidae, Ceratopogonidae, Chironomidae and Drosophilidae (Order Diptera) were identified in the present studies as the floral visitors. In addition to the above, five ant species of the family Formicidae (order Hymenoptera) were also found to be associated with pollination in cocoa. Cope (1940) and Kaufmann (1975) have recorded the involvement of ants in the pollination of cocoa.

Results obtained in the present studies revealed the natural pollination rate in cocoa to be low. Several researchers like Purseglove (1974), Murray (1975), Parvais et al. (1977) and Winder (1978) have also mentioned pollination as a limiting factor in cocoa production.

The present studies revealed a higher percentage of successful pollination in February and January. In Karnataka, higher percentage of successful pollination was obtained during the month of March (Anon, 1977). This variation may be probably due to the change in the population density of the pollinating insects. The present studies indicated a relationship between the degree of successful pollination and the intensity of cropping. The large pod harvest observed during June to August may be the result of higher rate of successful pollination of flowers during January and February. It may also be assumed that the lowest rate of successful pollination observed during October resulted in the absence of pod harvest during February-March.

#### Compatibility status

Cope (1962) stated that though it was possible to classify cocoa clones broadly into compatible and incompatible groups, there was still considerable variation within each group. In the present studies, out of the 15 plants self-pollinated, only four (J 7/3, J 11/2, L 9/3 and L 52/4) were found to be self compatible. Such low proportion of self compatible trees in cocoa population has been reported by Bartley (1963) who observed that out of 39 trees he studied, only eight trees were self-compatible. Sampayan (1963 and 1966) reported wide variation (0 to 93 per cent)

in the occurrence of self compatible trees in Criollo-Forastero natural hybrids such as the one included in the present studies. It was also observed, in the present studies, that all the 15 plants (four self compatible and eleven self incompatible) were cross compatible.

#### Natural vs hand pollination

Pollination in cocoa has been reported to be a limiting factor for obtaining good yields (Purseglove, 1974; Anon, 1977 and Winder, 1978). Glendining (1972) stated that normal fruit setting in cocoa required deposition of pollen grains on stigma in sufficient quantity. Low rates of fruit setting and pod harvest obtained under natural pollination may be due to inadequate or non-pollination. Results of the hand pollination studies (52.02 per cent set and 25.76 per cent pod harvest) indicated that fruit set and pod yield can be improved considerably by assisted pollination. The economics of hand pollination in cocoa is yet to be worked out. Jacob and Okoloko (1971) had, however, indicated that hand pollination is a practical proposition in Kola (Cola nitida Vent.) production. In the light of this observation, further work on assisted pollination in cocoa is worthwhile.



### Pod production from main trunk and fans

The present studies revealed certain new information regarding the flowering and fruit-setting behaviour on the main trunk and fan branches. Flower production, successful pollinations, fruit set and pod harvest were low on the fan branches as compared to those on the main trunk. The production of larger number of flowers on the main trunk, the availability of food reserves in sufficient quantities and the resultant reduced level of cherelle wilt may have given advantage to the main trunk as compared to the fan branches.

In the present studies, it was observed that the interval between flower production per cushion ranged considerably (33.81 to 165.30 days). Those cushions which supported the pods upto the harvestable stage flowered less frequently than those which exhibited no flower set or complete wilting of cherelles. Glendining (1972) observed that flowering in cocoa continued until suppressed by crop development. Alvim et al. (1972) also attributed inhibition of flowering in cushions to the pods developing on them.

The present investigations revealed that fruit development in cocoa is gradual. During the 22nd week, the week during which the pods ripened, there was no increase

in the length and thickness of the fruit-stalk. A slight reduction in the thickness of shell was observed during the period. This must have caused the observed reduction in the thickness of pods. It was interesting to observe that beyond the 10th week of fruit development, there was no loss due to onerelle wilt. In the present studies, cocoa pods, on the average, took 138.17 days for reaching the ripening stage. This figure worked out as per the formula of Alvim et al. (1972) came to 138.53 days, thus being in close agreement with the observations made in the studies.

## *Summary*

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## S U M M A R Y

The present investigations were carried out at the Regional Research Station, Pilicode, Cannanore district during 1980-81 to study (i) the pattern of flowering and cropping (ii) the floral biology and (iii) the fruit set and fruit development. The experimental material consisted of ten-year old cocoa plants intercropped in coconut plantations. The salient findings are summarised below:

1. The flower production was found to be throughout the year with two peak seasons, namely, May-June and November-December.

2. The maximum pod harvest was obtained during June-August season. A second peak was obtained during October to December.

3. Cherelle wilt was observed to occur throughout the year, maximum being in July. The percentage of cherelle wilt during the first week of pod development was 6.25. It reached a peak of 18.75 per cent at the sixth week. There was no loss due to cherelle wilt beyond the 10th week.

4. Newly emerged flower buds took 21 to 24 days to reach maturity and anthesis. Anthesis commenced

between 2 and 4 PM and completed between 2 and 4 AM, the next day.

5. Anther dehiscence commenced between 4 and 6 AM and completed by 8 to 10 AM.

6. Stigma receptivity was found to be high between 12 noon and 2 PM.

7. The pollen grains were found to be round and had diameter ranging 14.25 to 24.51  $\mu$ . The number of pollen grains per flower showed monthly variation (6100 to 7150), the average being 6625.

8. A medium containing 0.5 per cent agar, 10.0 per cent sucrose and 1.0 per cent of 100 ppm boric acid was found to be the best for germinating pollen in vitro. Calcium or boron (100 ppm) improved the percentage germination of pollen grains. Highest germination and tube growth were recorded at a temperature of 35°C. The highest percentage of germination and tube growth were observed when fresh pollen was collected at 8 AM.

9. Keeping pollen grains in tissue paper packets wrapped with filter paper, sealing in glass tubes and storing the tubes in refrigerator (after two hours of desiccation over calcium chloride) was found to be effective

in extending the viability of pollen grains upto five days.

10. No fruit set was observed on bagging the flowers.

11. The percentage of successful pollination was found to be higher in January and February than during the other months.

12. Seven insects belonging to the order Diptera and five Formicid species belonging to the order Hymenoptera were identified as floral visitors.

13. Out of the fifteen plants studied, four were self compatible. All the 15 plants were found to be cross-compatible.

14. The percentage of fruit set and pod harvest under natural pollination were 11.81 and 0.83, respectively. On hand pollination, these rose to 52.02 and 25.76 per cent, respectively indicating scope for assisted pollination in cocoa.

15. The percentage of fruit set on the main trunk was 13.51. Only 3.91 per cent of the flowers reached the harvestable stage. In the case of fan shoots, out of the total flower produced, 6.34 per cent set fruits. Only

0.26 per cent of the total flowers were carried to maturity.

16. The interval between flower production in the cushions ranged from 33.81 days to 165.30 days. Those cushions that supported the developing pods upto the harvestable stage flowered less frequently than those which exhibited no flower set or complete wilting of cherelles.

17. The development of cocoa pods was found to be a very gradual process. The pods took 127 to 141 days (mean 138.17 days) for reaching the ripening stage.

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\* Originals not seen.



# STUDIES ON THE FLORAL BIOLOGY AND FRUIT SET IN COCOA (*Theobroma cacao* L.)

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## ABSTRACT OF A THESIS

Submitted in partial fulfilment of the  
requirements for the Degree of

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## A B S T R A C T

Studies were conducted at the Regional Research Station, Pillicode during 1980-81 to gather information on the pattern of flowering and fruiting, aspects of floral biology, fruit set, fruit development etc. in cocoa.

Though flowering was seen throughout the year, two peak seasons (May-June and November-December) could be identified. A double peaked pattern was also observed with regard to pod harvest, June-August being the major peak. Cherelle wilt occurred throughout the year, the maximum being in July. Cherelles did not wilt after the tenth week of development.

Data on the commencement and completion of anthesis and anther dehiscence were collected. The stigma receptivity was found to be high between 12 noon to 2 PM. A medium for germinating pollen grains in vitro was identified. Keeping pollen grains in tissue paper packets under dry and comparatively cool conditions extended the viability upto five days. Seven Dipterous insects and five Formicid species were identified as floral visitors.

The fifteen plants included in the studies were found to be cross-compatible; but only four of them were self-compatible.

Hand pollination increased the percentage of fruitset and pod harvest, indicating scope for assisted pollination in cocoa.

Variation was observed between the main trunk and the fan shoots with regard to the percentage of fruit set, number of cherelles wilted and the percentage of cherelles carried to maturity.

The cushions that supported developing pods upto the harvestable stage flowered less frequently than those which exhibited no set or complete wilting of cherelles.

The development of cocoa pods was found to be a very gradual process. The pods took, on the average, about 140 days to reach the ripening stage.