CROSS COMPATIBILITY BETWEEN *Sesamum indicum* L. AND *S. malabaricum*

BY

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DECLARATION

I hereby declare that this thesis entitled "Cross compatibility between *Sesamum indicum* L. and *S. malabaricum*" is a bonafide record of research work done by me during the course of research and that the thesis has 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 "Cross compatibility between *Sesamum indicum* L. and *S. malabaricum*" is a record of research work done independently by Kum. Lali, K.S. under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.

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INTRODUCTION

Sesame (*Sesamum indicum* L.) is an important oil-seed crop in India, grown in an area of about 2.4 million hectares producing 0.5 million tonnes of seed annually. It is the most widely cultivated annual oilseed crop in Kerala, occupying an area of 15000 hectares with an annual seed production of nearly 3800 tonnes. Sesame oil is an important edible oil with high quality and stability. Seed contains about 50 to 55 percent oil and about 25 per-cent protein. It is also rich in calcium, phosphorus and vitamin E.

In Kerala, traditionally sesame is cultivated in the summer rice fallows as a catch crop. Attempts are being made to increase sesame production through its cultivation as a pure crop in uplands and also as a floor crop in coconut gardens during the rabi season. Suscepti-bility to diseases and drought is the most important problem facing sesame cultivation in such new environments. Natural variation for resistance to stress conditions is rather limited in the cultivated species. *Sesamum malabaricum*, the common wild species in the state shows tolerance to drought and most of the important diseases affecting the crop. There is good scope for utilization of this species for incorporation of it's resistance genes into the cultivated species.

A number of interspecific crosses were attempted in *Sesamum*. But only a few of them produced fertile hybrids. Reproductive isolation mechanisms such as incompatibility, hybrid inviability and hybrid sterility have been reported in many of the crosses. A study of the interrelationship and cross compatibility of *S. indicum* with *S. malabaricum* is an essential primary step for obtaining success in hybridization. Preliminary studies have indicated that the two species are noncompatible. Reciprocal crosses have to be attempted and the mechanism involved in their genetic isolation has to be fully exposed. Hence this study proposes to critically analyse as follows the reasons for seed failure in crosses between *S. indicum* and *S. malabaricum*.

1. Estimation of capsule and seed set in interspecific crosses.
2. Study of the role of pollination in capsule and seed development.

3. Study of the nature and pattern of seed development following cross pollination.

An understanding of the nature and severity of the barrier to gene exchange between the two species will enable us to adopt suitable methods for overcoming the noncompatibility and achieve interspecific gene transfer and thereby widen the scope for stress resistance breeding in sesame.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

The genus *Sesamum* belongs to the family Pedaliaceae which consists of only 16 genera. It was described by Linnaeus in "Genera Plantarum" during 1754. The name "Sesamum" was taken by Hippocrates from the Arabic word "Semsin". It is a genus of annual or perennial herbs or occasionally shrubs found in the warmer regions of Africa, Asia and Australia (Joshi, 1961; Rendle, 1963).

Literature on origin, classification, interspecific and intergeneric crosses in sesame and other annual oil-seed crops with emphasis on the barriers to crossability has been reviewed and presented.

1. Origin

There are a number of reports on the time and place of origin, the ancestral species and mode of evolution of sesame. East Indies, India, Middle east and Africa have been suggested as the possible centres of origin (De Candolle, 1886; Watt, 1893; Vavilov, 1926, 1951). Hiltebrandt (1932) regards Africa with its multiplicity of wild species as the primary centre of origin of sesame and India and Japan as secondary centres of origin. Watt (1893), Vavilov (1951) and Ramanujam and Joshi (1954) have suggested a polytopic origin for sesame. According to Nair and Mehra (1970) it is possible to narrow down the probable centres of origin to Africa and Peninsular India. Perhaps sesame might have evolved either independently in both these regions or has originated in one region and moved to the other. Both Africa and India are botanically contiguous, having a number of species common to both these regions. Bedigian and Harlan (1986) based on phytochemical and morphological data together with archaeological evidence concluded that sesame originated in India.

The progenitor of sesame is still a matter of controversy. Considering the wide distribution of S. grandiflorum (S. schenckii), its identity in chromosome number with that of the cultivated sesame, and also its breeding behaviour with the latter, Nair and Mehra (1970) advanced this species as the probable progenitor of sesame. But Nair (1976) reported that there is only little experimental evidence in support of the above proposition and stressed the need for the study of the probability of *S. indicum* subsp. *malabaricum* as an ancestral species to the cultivated *S. indicum*. Bedigian et al. (1985) based on the studies on lignin content, chromosome number and reciprocal crosses supported the view that *S. indicum* subsp. *malabaricum* (*S. orientale* var. *malabaricum*) is the progenitor of *S. indicum*.

2. The species of Sesame and their classification

Linnaeus described two species of sesame: *S. indicum* and *S. orientale*. De Candolle united them and preferred the specific epithet *indicum* to *orientale* (Bruce, 1953). Genus *Sesamum* since its first publication has been revised by various workers like Endlicher, Bernhardi, De Candolle, Bentham and Hooker, and Stapf dividing it into subgenera and sections based on morphology of leaves and seeds (Bruce, 1953; Joshi, 1961).

In Hooker's "Flora of British India" and Gamble's "Flora of the Presidency of Madras" besides the cultivated species *S. indicum*, L., two wild perennial and prostrate species occurring in India *S. prostratum*, Retz. and *S. laciniatum*, klein. have been described (John et al., 1950). Abraham (1945) collected wild sesame plants in

Travancore (Kerala State) and based on cytological studies in the wild plants, cultivated sesame and their sterile hybrid, considered it to be a distinct species and provisionally named it as S. grandiflorum. A species by the same name from tropical Africa was recorded in "Index Kewensis".

There is record of S. malabaricum from Kerala in "Index Kewensis" (Joshi, 1961). John et al. (1950) described a wild sesame of Malabar and based on cytological studies and its easy crossability with the cultivated species considered it to be a variety of the cultivated sesame and designated it as S. orientale var. malabaricum. Joshi (1961) suggested that Abraham's S. grandiflorum might be the same as the wild sesame of Malabar described by John et al. (1950).

In "Index Kewensis" and its supplements, 36 species of Sesamum have been recorded. Many species are still under controversy and need further confirmation. S. alatum is mentioned in "Index Kewensis" as a synonym of S. capense. Naidu (1953) described a species S. ekambaramii from Hyderabad which was found to be the same as S. alatum by Ramanujam and Joshi (1954). Bruce (1953) reported that

S. schenckii and *S. grandiflorum* are synonymous. Similarly *S. radiatum* was considered as a synonym of *S. occidentale* by early workers. Since they differ morphologically Kedharnath (1954) was inclined to believe that they are two distinct species. Close relationship between these two species has been reported by Subramanian (1975). According to Joshi (1961), a thorough cytotaxonomic examination of the numerous species of the genus *Sesamum* may result in further reduction of the number of species recorded under this genus.

Nair (1963) has reported a new species from Punjab and named it as *S. mulayanum* which according to him comes near the African species *S. angustifolium* and *S. radiatum* in many respects. Another wild sesame from Madurai (South India) which shows close similarity with *S. orientale* var. *malabaricum* has been reported by Amirthadevarathinam and Subramanian (1976). They have designated it as *S. indicum* var. *Yanamalai*.

The basic chromosome number of the genus *Sesamum* has been reported as 7, 8, 9 and 13 by various workers (Joshi, 1961; Nair and Mehra, 1970). The somatic chromosome number of *S. indicum* was first reported by

Moringa et al. (1929) as 26 which has been confirmed by many other workers. John and Rao (1941) reported the chromosome number of S. radiatum to be 2n = 64. Ramanujam (1941) reported the chromosome number of S. prostratum as 2n = 32. Abraham (1945) determined the diploid chromosome number of S. grandiflorum as 26. Somatic chromosome number of S. laciniatum was reported by Ramanujam and Joshi (1948) as 32. Kedharnath (1950) determined the chromosome numbers of two wild species from Africa, S. alatum and S. angolense as 2n = 26 and 32 respectively. Sampath and Ramanathan (1949) reported the chromosome number of S. occidentale obtained from Ceylon as 2n = 64. The chromosome number of S. orientale var. malabaricum was determined by John et al. (1950) and the chromosome number of S. indicum var. Yanamalai by Amirthadevarathinam and Subramanian (1976) as 2n = 26.

Based on the data available on the chromosome number of the species of Sesamum three broad groups were recognised by Joshi (1961).

Group I n = 13 chromosomes - S. indicum and S. alatum

Group II n = 16 chromosomes - *S. prostratum*
S. laciniatum and
S. angolense

Group III n = 32 chromosomes - *S. radiatum* and
S. occidentale

Nair and Mehra (1970) have listed 34 species of *Sesamum* which incorporates some changes and additions to the one given in "Index kewensis". The list of species along with their chromosome numbers and distribution are furnished below.

Species name	Chromosome number (2n)	Distribution
S. indicum	26	Cultivated species
S. indicum ssp *malabaricum*	26	India
S. capense	26	Tropical Africa, India, Australia.
S. schenckii	26	Tropical Africa, India, East Indies.
S. angolense	32	Tropical Africa.
S. laciniatum	32	India
S. prostratum	32	India
S. radiatum	64	Tropical Africa, Ceylon.

Species name	Chromosome number (2n)	Distribution
S. angustifolium	Not known	Tropical Africa.
S. antirrhinoides	"	Tropical Africa.
S. auriculatum	"	Crete
S. biapiculatum	"	Congo
S. brasiliense	"	Brazil
S. caillei	"	Guinea
S. calycinum	"	Tropical Africa.
S. denterii	"	Tropical Africa.
S. ligitaloides	"	Tropical Africa.
S. hendelotii	"	Tropical Africa.
S. latifolium	"	East Africa.
S. lepidotum	"	Tropical Africa.
S. malabaricum	"	India
S. marlotii	"	Africa, East Indies, Australia.
S. microcarpum	"	Tropical Africa.
S. mombazense	"	Tropical Africa.
S. pedaloides	"	Tropical Africa.
S. repense	"	Tropical Africa.
S. rigidum	"	Tropical Africa.
S. sabulosam	"	Sudan

Species name	Chromosome number (2n)	Distribution
S. schinzianum	Not known	Africa, East Indies.
S. somalense	"	Somalia
S. talbotii	"	Nigeria
S. thonnerii	"	Tropical Africa.
S. trifoliatum	"	India
S. tryphyllum	"	Africa, East Indies.

3. Interspecific Hybridization

Interspecific hybridization is a practice in plant breeding commonly adopted for transferring one or a few desirable genes from the wild or distantly related species to the cultivated species. Problems and prospects of interspecific hybridization as a breeding procedure have been dealt with in detail by many authors like Allard (1960), Briggs and Knowles (1967), Stalker (1980) and Singh (1983).

a. Objectives

Wild species are potential sources of many useful traits such as resistance to pests, diseases and drought which are in constant demand by plant breeders. In the genus *Sesamum*, *S. prostratum* is reported to be resistant to the caterpillar pest and phyllody (Ramanujam, 1942). *S. laciniatum* has been considered as a source of many desirable genes by Aiyadurai et al. (1962). Subramanian (1972) also described it as a hardy plant, resistant to drought, pests and diseases with partly dehiscent capsules. He further reported dense setting of capsules, longer size of capsules, resistance to drought, pests and diseases in *S. alatum* and desirable characters such as high seed set, profuse branching and vigour in *S. radiatum* and *S. occidentale*. While integrating the breeding objectives and agricultural practices under Indian conditions, Desai and Goyal (1981) identified the Indian wild taxa *S. prostratum*, *S. laciniatum*, *S. grandiflorum* and *S. orientale* var. *malabaricum* as sources of resistance to phyllody, drought and many insect pests.

b. Achievements

In Sesame, interspecific crosses have been tried for the first time by Ramanujam (1942), between *S. indicum* and *S. prostratum*. A number of crosses were reported in the later years. With the 34 known species of sesame, nearly 25 cross combinations have been tried so far involving *S. indicum*, *S. schenckii*, *S. grandiflorum*, *S. alatum*, *S. capense* (all 2n = 26), *S. prostratum*, *S. laciniatum*, *S. angolense* (all 2n = 32), *S. radiatum* and *S. occidentale* (2n = 64) (Nair, 1976).

The results of interspecific crosses may range from failure to obtain any seed set to complete fertility in F_1. ~~progeny~~ Hybridization was highly successful in *S. prostratum* x *S. laciniatum* (2n = 32) and *S. radiatum* x *S. occidentale* (2n = 64). Ramanathan (1950) and Kedharnath (1954) studied *S. prostratum* x *S. laciniatum* cross and reported that the reciprocal hybrids looked alike in all respects and the characters of *S. prostratum* were dominant in the F_1. The hybrids were fully fertile and had good seed set. In the F_2 and F_3 generations no weak or sterile plants were found. In *S. radiatum* x *S. occidentale* cross also, these authors reported good viable seeds and the hybrids from the reciprocal crosses looked alike. The characters of *S. occidentale* were dominant in the hybrids which were fully fertile. In the F_2 and F_3 generations there were no weak or sterile plants. Dadlani (1958) conducted embryological studies in *S. radiatum* x *S. occidentale* cross. The different developmental stages in the reciprocal crosses between these two species were similar to those of S. occidentale. Embryo development was quicker in S. radiatum x S. occidentale cross than in S. occidentale and the reciprocal cross. In S. radiatum x S. occidentale cross, the zygote started first division by 120 hours and reached the heart shaped stage at 360 hours after pollination, while in the reciprocal cross it started division at 144 hours and was found to be a globular structure even at 360 hours. Subramanian (1972, 1975) also reported that in the S. radiatum x S. occidentale cross, the hybrids were morphologically similar and more vigorous than the parents and with dominance biased towards S. occidentale. A continuous array of variation was observed in the F_2 segregants as in the case of inter-varietal crosses.

Only very few economically useful strains have been evolved in sesame by interspecific hybridization. The variety TMV.3 is a selection from the cross between S. indicum and S. indicum var. malabaricum (John et al., 1950). Amphidiploids produced by chromosome doubling of the sterile hybrid from the cross between S. indicum and S. prostratum was back crossed with S. indicum and improved types 'R' and 'S' have been evolved. They are giving high yields in Andhra Pradesh (Anon, 1972). Paramasivan et al. (1982) reported that hybrids of S. indicum var. Yanamalai with S. indicum showed heterosis for several characters.

4. Barriers to Interspecific Hybridization

Interspecific gene transfers are hindered by the isolation barriers which the species have developed during the course of evolution and the degree of crossability depends on the severity of the barrier (Jindal and Kalia, 1972). There are several reviews on the reproductive isolation mechanisms in plants which prevent interspecific gene transfers (Dobzhansky, 1951; Stebbins, 1950, 1958; Allard, 1960; Briggs and Knowles, 1967; Stalker, 1980; Singh, 1983). Four major barriers were identified by most of them. They are,

i. Incompatibility
ii. Hybrid inviability
iii. Hybrid sterility
iv. Hybrid breakdown

i. Incompatibility

Zygote formation is prevented by failure or ineffectiveness of pollen tube growth or failure of fertilization. Bannikova and Khvedynich (1978) compared the pollen tubes formed on selfing and after interspecific and intergeneric hybridization in crop plants of Gramineae, Solanaceae and Compositae. In incompatible crosses there were deviations from the normal growth of the pollen tubes and considerable changes in the reproductive cells. Reasons attributed are inability of the pollen tube to use substances in the style necessary for its growth and development, and direct suppression of the metabolic processes in the pollen tube when its tissues interacted with those of the style.

In *Sesamum*, in some species crosses, flowers dropped after pollination and hence no fruit set was obtained. Ramanathan (1950) reported such cases in crosses *S. indicum* x *S. radiatum*, *S. indicum* x *S. occidentale*, *S. prostratum* x *S. radiatum*, *S. prostratum* x *S. occidentale*, *S. laciniatum* x *S. radiatum* and *S. laciniatum* x *S. occidentale*. Failure of fruit set was reported by Kedharnath (1954) in *S. laciniatum* x *S. radiatum*, *S. laciniatum* x *S. angolense* and *S. prostratum* x *S. angolense*. Ill developed capsules were obtained by Subramanian (1972) in *S. indicum* x *S. occidentale*, *S. alatum* x *S. occidentale*, *S. occidentale* x *S. alatum*, *S. laciniatum* x *S. occidentale*, *S. laciniatum* x *S. radiatum*, *S. alatum* x *S. radiatum* and *S. laciniatum* x *S. alatum* crosses. Whether this abnormal fruit development is due to abnormal pollen tube growth or failure of fertilization has not been established. *S. indicum* and *S. mulayanum* are incompatible in reciprocal crosses. According to Sastri and Shivanna (1976) this interspecific incompatibility is due to the failure of the pollen to germinate on the stigma of the recipient parent. In the presence of recognition pollen, pollen tubes entered the stigma, but they were inhibited after growing a few millimetres in the style. They concluded that the inhibitory factors were present not only on the stigma, but also in the style.

In linseed, Jindal and Kalia (1972) studied crosses among *Linum grandiflorum*, short styled (S), *L. grandiflorum*, long styled (L), *L. humile* and *L. decumbens*. *L. grandiflorum* (L) pollen could not germinate on the stigma of *L. humile* and *L. decumbens*. When *L. grandiflorum* (L) and (S) were used as female parents, *L. humile* as well as *L. decumbens* pollen grains germinated normally, but growth of the pollen tubes got arrested in the style and later on to a great extent the pollen tubes showed abnormal growth. The noncrossability was clearly due to the restricted pollen tube growth and the reason was attributed to the disparity in chromosome number of pollen and styles or the effect of inhibitory substances present in the style. Green (1983) crossed *L. usitatissimum* with 20 wild species of *Linum*. Pollen tubes were present in the style but no capsule set occurred in crosses to eight species, pollen grains failed to germinate in crosses to three species.

Sareen et al. (1970) studied pollen germination and pollen tube growth in crosses involving diploid, triploid and tetraploid forms (2n = 20, 30 and 40 respectively) of *Brassica campestris* var. brown Sarson and found that in the incompatible crosses most of the pollen tubes turned abnormal before entering the stylar tissue. Pollen tubes showed stunted growth in diploid x diploid, diploid x triploid, triploid x diploid, triploid x triploid and triploid x tetraploid crosses. Pollen from tetraploids showed good germination and growth in diploids and tetraploids. Growth was poor in triploids. Bochkarev (1976) reported that in interspecific hybridization between B. campestris and B. nigra, pollen tubes showed anomalous growth and were unable to penetrate the tissue of the style. The incompatibility was greater when B. nigra was used as the female parent. Khanna and Chowdhury (1981) conducted studies on pollen tube growth and seed set in some interspecific crosses involving B. campestris, B. juncea, and B. chinensis. The maximum pollen germination was found in the cross B. campestris x B. Chinensis and minimum in B. juncea x B. campestris. Maximum abnormalities in pollen tube growth were found in the cross B. juncea x B. campestris. Abnormalities were in the form of swelling of the tip of pollen tubes or their bursting in the stigmatic tissue. Most of the crosses showed very poor seed set. They concluded that incompatibility in these crosses is a prefertilization process. Banga (1985) attempted mentor pollen aided hybridization between B. hirta (2n = 24) and B. juncea (2n = 36). B. hirta when pollinated with irradiated self pollen followed by fresh B. juncea pollen and application of GA_3 (20 ppm) after 24 hours of pollination prevented bud fall and gave better results.

In the case of Arachis, Smartt and Gregory (1967) observed that species of the section 'Villosoid' (A. hypogaea and A. villosa) and 'Rhizomatous' (A. glabrata, A. marginata and A. hagenbeckii) are cross incompatible. Failure of fertilization along with post fertilization failure in the development of the embryo was reported. Mallikarjuna and Sastri (1985a) reported that application of GA to the bases of flowers following incompatible intersectional pollinations of A. hypogaea with species of the section Rhizomatous led to peg formation and in some cases to the formation of pods.

ii. Hybrid inviability

The major causes of hybrid inviability given by Stebbins (1958) are noncompatibility of the parental chromosomes, cytoplasmic-genic interactions and non-compatibility between embryo and the surrounding tissue ie. somatoplastic sterility. Somatoplastic sterility has been studied in detail in crops like Medicago sativa (Brink and Cooper, 1939, 1940), tobacco, barley and tomato (Cooper and Brink, 1940, 1944, 1945). In recent literature, there are a number of reports on hybrid inviability due to chromosome elimination from the hybrid embryo or endosperm. This has been reported in *Nicotiana* by Gupta and Gupta (1973) and barley by Finch (1983). The endosperm must be in a 2:1 (maternal: paternal) ratio for normal development. It develops abnormally in interploid crosses when the ratio deviates from 2:1. Johnston et al. (1980) proposed an Endosperm Balance Number (EBN) hypothesis to explain this phenomenon.

In the case of sesame, hybrid inviability has been reported in a number of species crosses. In the cross *S. indicum* x *S. alatum*, Kedharnath (1954) reported normal fruit set, but the seeds appeared shrivelled and nonviable. Early abortion of the young embryo has been considered as the reason. The reciprocal cross gave only very few crossed fruits. Though the seeds looked apparently healthy, they were nonviable. Subramanian (1972) obtained empty seeds in *S. indicum* x *S. alatum* cross. In the reciprocal cross, seeds were shrivelled. In *S. indicum* x *S. capense*, Amirthadevarathinam (1965) reported recovery of shrivelled seeds. In the reciprocal cross, there was no seed set. Sundaram (1968) also reported failure in getting well developed and viable seeds in *S. indicum* x *S. capense* (direct and reciprocal crosses). Kedharnath

(1954) reported that in crosses S. angolense x S. prostratum and S. angolense x S. laciniatum, few fruits containing shrivelled and non-viable seeds were obtained. In the reciprocal crosses there was no fruit set. Garu (1934) recorded failure in his attempts to cross S. indicum with S. radiatum. Dhawan (1946) conducted embryological studies in S. radiatum x S. indicum cross and observed that primary endosperm nucleus formed a normal two celled endosperm, 24 hours after pollination as in S. radiatum. But the two celled endosperm underwent rapid and irregular divisions, cells appeared shrivelled and their nuclei inconspicuous and they lay separated from each other. In the hybrid zygote, the male and female nuclei did not fuse till the 3rd day. The endosperm completely disintegrated and disappeared by the fourth or the fifth day. In S. radiatum, the first division of the zygot e took place four and a half days after pollination. In S. radiatum x S. indicum cross, the zygote had died by that time, apparently due to starvation. The zygote appeared to be normal till its death. When crossed to tetraploid S. orientale, S. radiatum gave a few viable seeds presumably of apomictic origin. Studies conducted at IARI (Anon., 1948) in the S. orientale x S. radiatum cross showed that death of hybrid embryo was due to disintegration of the endosperm. First sign of abnormality in endosperm was exhibited 48 hours after pollination when its cells appeared shrivelled. Upto four days after pollination, embryo growth was normal, but five days after pollination the embryosac cavity contained a few collapsed cells and no embryo was visible. Ramanathan (1950) reported recovery of both shrivelled and normally developed nonviable seeds in _S. radiatum_ x _S. indicum_ cross. Mazzani (1952) reported that _S. indicum_ x _S. radiatum_ cross gave fruit set but had no seeds. Reciprocal cross gave a few seeds of which some germinated when the embryo was removed and cultured in Tukey's medium. They gave rise to three plants of weak growth which resembled _S. radiatum_ in gross morphology. The chromosome number of these plants was n = 32. They were completely pollen fertile and were supposed to be the result of diploid parthenogenesis in the maternal plant. The progeny of these plants also resembled _S. radiatum_ but were more vigorous. Subramanian (1972) also reported recovery of small and shrivelled seeds in _S. indicum_ x _S. radiatum_ reciprocal crosses.

Dhawan (1946) reported that the cross _S. radiatum_ x

S. prostratum failed to give viable seeds and the seeds had a collapsed appearance. Ramanathan (1950) reported recovery of both shrivelled and normally developed non-viable seeds in this cross. Similar results were reported in *S. occidentale* x *S. indicum*, *S. occidentale* x *S. laciniatum*, *S. occidentale* x *S. prostratum* and *S. radiatum* x *S. laciniatum* crosses also by the same author. Dadlani (1958) studied the embryology of cross *S. occidentale* x *S. prostratum* and found that though the primary endosperm nucleus divided normally soon after the first division, subsequent divisions were irregular and rapid. The cells appeared disconnected and endosperm was disorganised. Chalazal haustoria though formed, failed to come in contact with the nutritive pocket, as well as with other endosperm cells. The endosperm gradually disintegrated. The zygote appeared normal. But due to lack of nutrients as a result of endosperm failure the embryos soon shrivelled up and died. Subramanian (1972) reported recovery of shrivelled seeds in the crosses, *S. occidentale* x *S. indicum* and *S. radiatum* x *S. laciniatum*. In *S. occidentale* x *S. laciniatum* cross seedlings died soon after germination.

In the case of groundnut, regular fertilization was noticed in *Arachis hypogaea* x *A. diogoi* cross, but hybrid inviability resulted in delayed fruit elongation, retarded embryo and endosperm growth, failure of embryo differentiation, hyperplasia of the integuments and eventually collapse of the embryo and endosperm. There was no evidence of irregular mitosis in the embryo or endosperm. Collapse of the endosperm occurred at the 32 or 64 nucleate stage. Hybrid embryos survived somewhat longer (as long as 44 days) in some cases, but eventually all of them aborted and the mature pods harvested were empty (Johansen and Smith, 1956; Raman et al., 1972). Pompeu (1977) reported failure in crossing *A. hypogaea* with the diploids *A. diogoi* and *A. villosulicarpa*. In both crosses fruits began to develop, but reached only 2-3 cm in length and contained aborted embryos. But Pompeu (1980) reported success in crossing *A. hypogaea* with *A. diogoi*. In the cross *A. hypogaea* x *A. glabrata*, hybrid inviability has been reported by Raman et al. (1972). Strong genetic differences between the two species have been attributed as the cause of this phenomenon.

In soybean, Hood and Allen (1980) reported the results of histological studies in the cross between

<u>Glycine max</u> and a perennial wild species <u>G. falcata</u>. A podset of 11.3% was obtained, but all of them aborted in 23 days. Progeny of <u>G. latifolia</u> x <u>G. tomentella</u> died as seedlings while those of <u>G. falcata</u> x <u>G. canescens</u> grew slowly for several months and then withered (Newell and Hymowitz, 1983). 2.5 to 7.6% pod set occurred in a cross between <u>G. max</u> and <u>G. tomentella</u>. But aneuploidy was observed in hybrid embryos and embryo growth was markedly retarded after the heart shaped stage (Sakai and Kaizuma, 1985).

Nishiyama and Inomata (1966) made embryological studies in crosses between diploid <u>Brassica chinensis</u> (2n = 20) and autotetroploid <u>B. pekinensis</u> (2n = 40). It was observed that the hybrid embryos develop normally in the early developmental stages, but generally failed to develop beyond the globular stage as the degeneration of endosperm progressed. Exceptionally a few triploid plants were obtained from the 4x X 2x cross. The rate of endosperm development was accelerated when the pollen parent had a higher number of chromosomes than the ovule parent but was inhibited when the number of chromosomes was lower. Sareen etal. ~~Chowdhary~~ (1970) reported that in the cross between diploid and tetraploid B. campestris, when diploid was used as female parent, in spite of good pod set, seed set was poor. They concluded that the pollen although sufficiently successful to stimulate pod formation, was inefficient to bring about either fertilization or further growth of zygote or embryo. There was good seed set in tetraploid x diploid crosses. Ayotte et al. (1985) reported that when B. napus was crossed with B. oleracea, only half of the ovules in an ovary started developing and later started to shrivel and collapse.

Jindal and Kalia (1972) reported that in the crosses of Linum humile and L. decumbens with L. grandiflorum (short styled) as male, there was 36 and 39% capsule set respectively. Fertilization was normal one day after pollination. But 3 days after pollination the endosperm started degeneration. Growth of embryo was arrested and later the embryos degenerated. Dubey and Kumar (1973) studied the cross relationship between L. usitatissimum (n = 15) and L. grandiflorum (n = 8). It was found that post fertilization development in the cross pollinated ovaries was different from that of the self pollinated ones and abortion of ovules took place in cross pollinated ovaries. In some cases, embryo development proceeded normally upto the four celled stage, but endosperm nuclei were not observed. As a result, the growth of the embryo was arrested and the ovule started degeneration. In a second type, the development of the embryo and endosperm started normally, but after the formation of a few nuclei, the endosperm suddenly collapsed and the development of the embryo stopped at the proembryonic stage. In another type, numerous free endosperm nuclei were observed to be compactly arranged in the dense cytoplasm. Not a single endosperm nucleus was found near the developing embryo and this development of endosperm resulted in the degeneration of the embryo and finally the seed. Green (1983) tried crosses between L. usitatissimum and 20 wild Linum species. Capsule set occurred but no seeds were obtained from crosses to seven wild species.

In the case of Helianthus, hybrid inviability was reported in crosses between cultivated species (H. annuus) and several diploid perennial species (Chandler and Beard, 1983). In most cases, the endosperm was severely shrivelled or absent. Embryos from such crosses developed only upto the globular or early heart stages.

A method suggested to overcome hybrid inviability is ovary/ovule/embryo culture especially when embryo abortion takes place due to failure of development of endosperm. Now it is being tried in almost all crops. Mazzani (1952) reported embryo culture in sesame. This has also been reported in *Arachis* by Sastri et al. (1980), Bajaj et al. (1982), Sukumar and Rangasamy (1984) and Mallikarjuna and Sastri (1985b), in *Helianthus* by Chandler and Beard (1983), in *Glycine* by Newell and Hymowitz (1982) and in *Brassica* by Inomata (1983) and Mahapatra and Bajaj (1984).

iii. Hybrid sterility

Hybrid sterility has been classified as gametic and zygotic by Renner (1929), Haplontic and diplontic by Muntzing (1930) and as genic and chromosomal by Dobzhansky (1951). Hybrid sterility due to cryptic structural hybridity was first reported by Stebbins (1950).

Reciprocal crosses between *S. indicum* and *S. prostratum* were attempted for the first time by Ramanujam (1942). Pod set and seed set were good. Eleven hybrid plants obtained from the direct cross and two from the reciprocal cross grew well and flowered profusely. Reciprocal hybrids looked alike but were completely sterile.

Vegetative buds in a few hybrids were treated with colchicine. Fertility has been induced by this treatment and the treated branches produced capsules. Hybridization between these two species and the synthesis of the amphidiploid was reported by several other workers such as Raghavan and Krishnamurthy (1947), Parthasarathy and Kedharnath (1949) and Ramanathan (1950, 1955). The amphidiploid was designated as S. indicatum by Ramanujam (1944). It had a somatic compliment of 58 chromosomes and showed normal meiosis in pollen mother cells with 29 regular bivalents. It was stable and almost true breeding. Parthasarathy and Kedharnath (1949) obtained sesquidiploids or allotriploids, a di-indicum - mono-prostratum (2n = 42) and a di-prostratum - mono-indicum (2n = 45) by back crossing the amphidiploid S. indicatum to the two parental species S. indicum and S. prostratum respectively. In the progeny of the sesquidiploid di-indicum - mono-prostratum (2n = 42), nearly 80 percent of the plants had a somatic complement of 26 chromosomes like the S. indicum parent, while the remaining 20 percent of the plants were aneuploids with extra chromosomes belonging to the prostratum genome. Among the normal (2n = 26 chromosome) progeny, which greatly resembled the *S. indicum* parent in morphological characters, nearly 28 percent exhibited certain morphological traits of the *S. prostratum* parent. Joshi (1961) suggested that they were either "substitution races" or that some amount of allosyndetic pairing should have occurred during meiosis in the sesquidiploid, leading to genic exchange between the chromosomes of the two parents.

The reciprocal crosses between *S. indicatum* (2n = 58) and autotetraploid *S. indicum* could be easily made and good fruit and seed set obtained. The resulting hybrids had 2n = 55 chromosomes. The hybrids from reciprocal crosses looked alike and were more erect in habit than the sesquidiploid di-*indicum* - mono-*prostratum*. The hybrid was completely pollen sterile. When used as the pistillate parent in crosses with tetraploid *S. indicum*, diploid *S. indicum*, *S. prostratum* and *S. indicatum* there was no fruit formation (Kedharnath, 1954). Ramanathan (1955) obtained a few well filled seeds from the partially sterile hybrids (2n = 55). Five seedlings came well upto the cotyledonary stage but later on succumbed.

Hybrid sterility has been reported in reciprocal crosses of *Sesamum indicum* x *S. grandiflorum* and *S. prostratum* x *S. grandiflorum* by Abraham (1945). The hybrids in both interspecific crosses proved to be vigorous plants, intermediate in morphological characters between the two parents, but totally sterile. There was about 0.1% fertility during the "end period". Fertile amphidiploids were obtained by colchicine application on the sterile hybrids.

Ramanathan (1950), Kedharnath (1954), Aiyadurai et al. (1962, 1963), Amirthadevarathinam (1965), Subramanian (1972) and Subramanian and Chandrasekharan (1977) reported that in the direct and reciprocal crosses between *S. indicum* and *S. laciniatum*, good viable seeds were obtained. The hybrids from both the crosses looked alike in morphological features. They were vigorous and flowered profusely but were completely sterile. Hybrid vigour for height, number of branches and number of flowers was observed. The amphidiploid obtained by colchicine treatment of the sterile hybrid was named *S. laciniatale* by Aiyadurai et al. (1963).

Raman (1960) obtained sterile hybrids in the cross between *Arachis hypogaea* and *A. villosa*. Raman (1973) reported a form of genic sterility in triploid hybrids obtained from the cross A. diogoi (2n = 20) and A. glabrata (2n = 40). Hybrids were more vigorous in vegetative growth than the parents, but the flower buds did not grow out of the bracts and degenerated, leading to sterility. Seetharam et al. (1973) reported hybrid sterility in the cross between A. hypogaea and A. duranensis (diploid). Triploid sterile hybrids obtained from the cross A. hypogaea x A. villosa var. correntina was subjected to colchicine treatment and a fertile hexaploid was obtained by Pompeu (1977). Gardener and Stalker (1983) also produced amphidiploids in order to overcome hybrid sterility due to ploidy difference in the parents.

In Helianthus, hybrids obtained from the cross between diploid perennials H. gigantius and H. maximiliani and cultivated or wild annual H. annuus (diploid, 2n = 34) showed partial or complete sterility (Whelan, 1978). Hybrids from the cross between H. annuus and tetraploids (H. hirsutum and H. decapetalus, 2n = 68) had variable ploidy levels ranging from haploid to octoploid and were tuber forming but sterile (Georgieva-Todorova et al. 1980).

Hybrid sterility in Brassica was reported by

Aslamyousuf and Bechyne (1983). Reciprocal hybrids obtained from the crosses *B. campestris* x *B. carinata* and seed *B. carinata* x *B. napus*, developed normally but there was no seed set. *B. napus* x *B. carinata* hybrids set some seeds. Polyploid plants derived from the sterile hybrids were also sterile.

iv. Hybrid breakdown

In many hybrid combinations, the F_1 individuals are apparently fully fertile, but segregants in the F_2 and later generations are either weak or sterile (Stebbins, 1950, 1958). Bates and Deyoe (1973) named all the post zygotic abnormalities together as hybrid breakdown.

5. Intergeneric hybridization in Sesame

Kedharnath (1954) attempted crosses between *Ceratotheca sesamoides* (2n = 32) and various species of *Sesamum*. No viable seeds were obtained in any of the crosses, except *S. indicatum* (2n = 58) x *C. sesamoides*. In this cross about 28 percent of the seedlings showed 2n = 45 chromosomes. The plants were semierect in habit and appeared intermediate between the two parents in many of the morphological characters and were completely sterile.

Subramanian (1973) studied crossability of Martynia annua (2n = 32) with Sesamum species. Pollen from M. annua were dusted on flowers of S. laciniatum (2n = 32), S. radiatum (2n = 64), S. occidentale (2n = 64) and F_1 of S. radiatum x S. occidentale (2n = 64). Growth of the pollen tubes was normal, but the crossed flowers dropped off after 4 or 5 days of pollination.

MATERIALS AND METHODS

MATERIALS AND METHODS

The present investigation was conducted in the Department of Plant Breeding, College of Agriculture, Vellayani during the period from September 1985 to November 1986.

A. Materials

Eleven adapted varieties of sesame (*Sesamum indicum*) were collected from the germplasm maintained in the department. These varieties differed in plant, capsule and seed characters. Their major distinguishing characters are given in Table 1. Some of the varieties with diverse plant type are presented in Figures 1a to d. Seeds of the wild species *S. malabaricum* were locally collected. Its plant type is provided in Figure 2. Morphological description of *S. indicum* in comparison to *S. malabaricum* is presented in Table 2. The capsule characters of the 11 varieties and the wild species are presented in Figure 3.

B. Methods

1. Cross compatibility

Each variety of *S. indicum* was crossed separately

Table 1. Distinguishing characters of *Sesamum indicum* varieties.

Sl. No.	Name of variety	Number of capsules per axil	Number of locules per capsule	Seed colour
1	ACV.1	1	6-8	White
2	ACV.2	1	6-8	Grey
3	IC.284	1	6-8	Yellowish brown
4	S.8	1	4	Black
5	P.10-1	1	6-8	White
6	Si.1275	1	6-8	Light brown
7	Cul.16	3	4	Light brown
8	Cul.28	1	6-10	White
9	Cul.40	3	4	Light brown
10	Vinayak	1	4	Brown
11	N-62-32	1	6-8	Dark brown

Table 2. Morphological description of *Sesamum indicum* and *S. malabaricum*.

Sl. No.	Character	*S. indicum*	*S. malabaricum*
1	Habit	Erect annual herb	Erect annual herb
2	Height	Tall	Tall
3	Branching	Simple or branched	Compact or semibranching
4	Stem		
	Colour	Yellowish green	Pale green
	nature	Obtusely quadrangular with longitudinal grooves on all sides.	Cylindrical towards the base and becoming more or less quadrangular towards the tip. A longitudinal groove is present along the quadrangular region.
	Pubescence	Finely pubescent to glabrous usually more or less glandular.	Finely pubescent.

5	Leaves	Heteromorphic, opposite or alternate.	Simple, opposite below and rarely alternate above.
	petiole	Lower leaves long petioled, upper ones more shortly petioled.	Lower leaves long petioled, upper leaves with very short petioles.
	shape	Lower leaves ovate or lanceolate, 3 lobed 3 partite or 3 foliate rounded or obtuse at the base and acute at the apex, margins often dentate. Upper leaves norrow, oblong, lanceolate or linear, usually entire and narrowly cuneate at the base.	Lower leaves broad and bigger cuneate at the base and acute at the apex, cordate in shape and margins distinctly dentate. Upper leaves smaller in size.
	colour	Ranges from pale to dark green in different varieties. Usually upper surface more darker than lower surface.	Upper surface dark green with impressed veins and lower surface paler, distinctly veined.

	pubescence	All leaves thinly pubescent, more or less glandular becoming glabrous.	All leaves thinly pubescent becoming glabrous.
6	Flowers	Axillary, solitary or in groups of 3.	Axillary and solitary.
	pedicel	Short pedicelled.	Short pedicelled.
	Lateral glands	Two yellow glands are present at the base, each gland with a narrow bract. In the case of multi-flowered varieties these glands are absent.	Two dark purple glands are present at the base of flowers, each gland with a narrow bract.
	calyx	Persistant	Persistant
	corolla	Pubescent, white or light pink in colour.	Pubescent, bigger in size, pink on the exposed inner side of the lobes, light purple on ventral side and a little deep purple on the dorsal side. Just below the tip of lower lip there is a light yellow crescent with purple lines, purple dots all around inside the corolla.

	stamens	White in colour, filaments glabrous and long.	Filaments glabrous, with minute purple dots, anthers creamy white in colour.
	pistil	Ovary slightly compressed, 4 to 8 loculed, more or less rounded at the apex, style glabrous, stigmatic lobes two or more depending on the number of locules present.	Ovary slightly compressed, 4 loculed, style glabrous whitish with small purplish streaks, stigmatic lamellae bifid.
7	Capsule	Oblong, quadrangular, slightly compressed, 4 to 8 grooved, rounded at the base. Apex abrupt with short beak.	Oblong, erect, quadrangular, 4 grooved, rounded at the base, tapering towards the apex. Apex-cleft beaked.
8	Seeds	White, brown or black in colour, 3 to 3.5 mm long 1.8 to 2 mm broad. Number of seeds per capsule - 50 to 100. Mild reticulations on seed coat.	Black in colour. About 3 mm long and 1.8 mm broad. Number of seeds per capsule-90 to 130. Deep reticulation on one side of seed coat.
9	Reaction to stress conditions	Susceptible to drought, pests and diseases.	Resistant to drought and diseases like leaf spot and powdery mildew.
10	Duration	85 to 90 days.	100 to 125 days.

Figure 1. Varieties of *Sesamum indicum*

a. ACV.1

b. S.8

c. Cul.40

d. Vinayak

Figure 1a

Figure 1b

Figure 1c

Figure 1d

Figure 2

Sesamum malabaricum

Figure 3. Capsule characters of _Sesamum indicum_ varieties and _S. malabaricum_.


ACV.1
ACV.2
IC284
S.8
P.10-1
Si.1275
Cul.16
Cul.28
Cul.40
VINAYAK
N.62-32
SESAME -WILD

Figure 3

and reciprocally with S. malabaricum. There were thus eleven direct crosses and eleven reciprocal crosses. The crossing plot consisted of rows of S. indicum varieties and S. malabaricum sown in several batches at specific time intervals. Since S. malabaricum has a longer pre-flowering period (10-15 days than S. indicum), phased sowing was done in each batch for synchronised flowering.

The crossing technique suggested by John and Nair (1981) was followed. Mature flower buds which were due to open the next day were selected in the evening for emasculation. A longitudinal slit was made along the upper ridge of the corolla tube with a needle. The stigma then slides out of the corolla. The upper part of the corolla was cut and removed along with the stamens. Emasculated flower buds were then protected using butter paper covers. Pollination was done on the next day early morning between 5.30 and 7.30 A.M. Flowers of the male parent which have just opened were collected and pollen scooped out of the anthers using a scapula were applied on the stigmas. Pollinated flowers were again protected using butter paper covers till evening. Then they were removed and the stigmatic lobes were cut to prevent chance contamination by foreign pollen. The crossed flowers were labelled. Crossed capsules alone were retained on the plant. In the case of multiflowered varieties the central flowers only were crossed. The lateral buds in each axil were removed at the time of emasculation. The crossed capsules were harvested at maturity before dehiscence, sun dried and seeds extracted.

The observations were recorded as follows:

(i) Capsule set - Total number of flowers crossed and total number of capsules set were recorded for each cross combination and the percentage of capsule set was calculated.

(ii) Seed set - Number of normal and shrivelled seeds (medium shrivelled and highly shrivelled) were counted in each crossed capsule. Percentages of normal and shrivelled seeds were estimated from the total number of seeds from all crossed capsules in each combination.

2. Pollination and fertilization

(i) The role of pollination in capsule development was studied in all the varieties of S. indicum and the wild species. The flowers were emasculated but not pollinated. They were tagged and capsule development recorded.

(ii) The rate of pollen tube elongation and time taken for fertilization after pollination was studied in four selected varieties viz. ACV.1, S.8, Cul.40 and Vinayak (Figures 1a to d) by self pollinating the emasculated flowers and cutting away the style close to its base at specific time intervals after pollination. Styles were excised at half hour intervals from pollination upto seven hours. The selfed flowers were retained as such without style excision in the control group. Thus there were 15 treatments in total. The percentage of flowers developing into capsules was estimated. Time for which style is to be retained after pollination for fruit set gives an indication of the time required for fertilization after pollination.

(iii) Pollen tube growth

Microscopic examination of pollen tube growth in stigma and style was done in selfed and crossed flowers of *S. indicum* (variety S.8) and *S. malabaricum*. Mature flower buds were emasculated in the evening. They were selfed or crossed on the following day.

Styles were cut at the base at different time intervals (1, 3 and 5 hrs) after pollination and scalded in 1 N HCl for 2 to 3 minutes. The outer cortex of the style was gently separated using fine needles for easy penetration of the stain. The softened styles were stained in 1% cotton blue for three hours and destained in lactic acid overnight. The material was then changed to fresh lactic acid and gently pressed on a slide to spread the tissue.

3. Capsule and seed development

These studies were conducted in the four selected varieties of *S. indicum* and their direct and reciprocal crosses with *S. malabaricum* during the Kharif season ie. July to October 1986. The temperature during this time ranged from 18.5 to 32.2°c and relative humidity ranged from 60 to 100 per cent. Capsules after selfing and crossing were harvested at intervals of two days upto 20 days. Size of freshly harvested capsules (length and Breadth) and length of fresh seeds was measured. The seeds were dried and size of dry seeds (length and breadth) measured upto 30 days after pollination.

The length and breadth of capsules were measured using a graph paper. Length was measured from the base to the tip of the capsule and breadth was recorded at the broadest portion. Measurements were taken at each time interval from 10 selfed and crossed capsules.

Number of seeds were counted in 10 capsules at random and the mean was worked out for all the varieties and crosses at each time interval. During the early stages of capsule development, a hand lens was used to count the number of seeds.

Seeds were selected at random from all the 10 capsules for length measurement. A standardised occular micrometer was used for measurement. Twentyfive seeds selected at random were measured.

Measurement of dry seed size (length and breadth) was made in two varieties of S. indicum (ACV.1 and S.8), S. malabaricum and their crosses. Measurement was made using an occular micrometer following the same procedure for fresh seed length measurement.

4. Statistical procedures

Mean values of the observations recorded were determined for all the quantitative characters.

RESULTS

RESULTS

1. Cross compatibility

In *Sesamum indicum*, flower opening under Vellayani conditions was between 5.30 and 6.30 A.M. The anthers started dehiscing from 4.30 A.M. onwards. Stigmas became receptive at 5.00 A.M. and remained so upto 7.30 A.M. In the case of *S. malabaricum* also, flower opening was between 5.30 and 6.30 A.M. But anthers started dehiscing from 6.30 A.M. only. Stigma was receptive from 5.30 A.M. and remained so upto 8 A.M.

i) a. Capsule set in *Sesamum indicum* x *S. malabaricum* crosses:

Capsule and seed set in different crosses are given in Table 3. Capsule set in *S. indicum* was not much affected by this interspecific cross. Among the eleven crosses tried between different varieties of *S. indicum* and the wild species, capsule set ranged from 51% (ACV.2 x *S. malabaricum*) to 95% (Vinayak x *S. malabaricum*). Crossed capsules developed normally and they were similar to the capsules of the female parent in shape and size.

Table 3. Capsule and seed set in Sesamum indicum x S. malabaricum crosses.

Sl. No.	Crosses	Capsule set			Number of seeds			% of seed set		
		Number of flowers crossed	Number of capsules set	% of capsules set	Normal	Medium shrivelled	Highly shrivelled	Normal	Medium shrivelled	Highly shrivelled
1	ACV.1 x S.M*	71	56	78.9	0	0	Numerous	0	0	100
2	ACV.2 x S.M	65	33	50.8	0	Numerous	0	0	100	0
3	IC.284 x S.M	53	46	86.8	0	0	Numerous	0	0	100
4	S.8 x S.M	74	60	81.1	0	Numerous	0	0	100	0
5	P.10.1 x S.M	43	32	74.4	0	0	Numerous	0	0	100
6	Si.1275 x S.M	51	46	90.2	0	0	Numerous	0	0	100
7	Cul.16 x S.M	74	61	82.4	0	0	Numerous	0	0	100
8	Cul.28 x S.M	70	57	81.4	0	0	Numerous	0	0	100
9	Cul.40 x S.M	91	66	72.5	0	0	Numerous	0	0	100
10	Vinayak x S.M	97	92	94.9	0	Numerous	0	0	100	0
11	N-62-32 x S.M	65	59	90.8	0	Numerous	0	0	100	0
	Mean	68.5	55.3	80.7	0	--	--	0	36.4	63.4

*S.M - Sesamum malabaricum

b. Capsule set in *Sesamum malabaricum* x *S. indicum* crosses:

Capsule and seed set in these crosses are presented in Table 4. Data on capsule set in the direct as well as reciprocal crosses are graphically represented in Figure 4.

Percentage of capsule set was higher in reciprocal cross in general (95.5%) as compared to the direct cross (80.7%). It was more or less uniform in all the eleven reciprocal crosses attempted. In crosses *S. malabaricum* x Cul.40 as well as *S. malabaricum* x N-62-32 there were cent percent capsule set. Minimum percentage of set was recorded by the cross *S. malabaricum* x ACV.2 (85%). Capsules obtained from different crosses were similar and they resembled the capsules of the female parent (wild species) in all respects.

ii) a. Seed set in *Sesamum indicum* x *S. malabaricum* crosses:

Crossed seeds obtained from the eleven crosses along with those of the standard variety ACV.1 are presented in Figure 5a.

Normal seeds could not be recovered from any of

Table 4. Capsule and seed set in Sesamum malabaricum x S. indicum crosses.

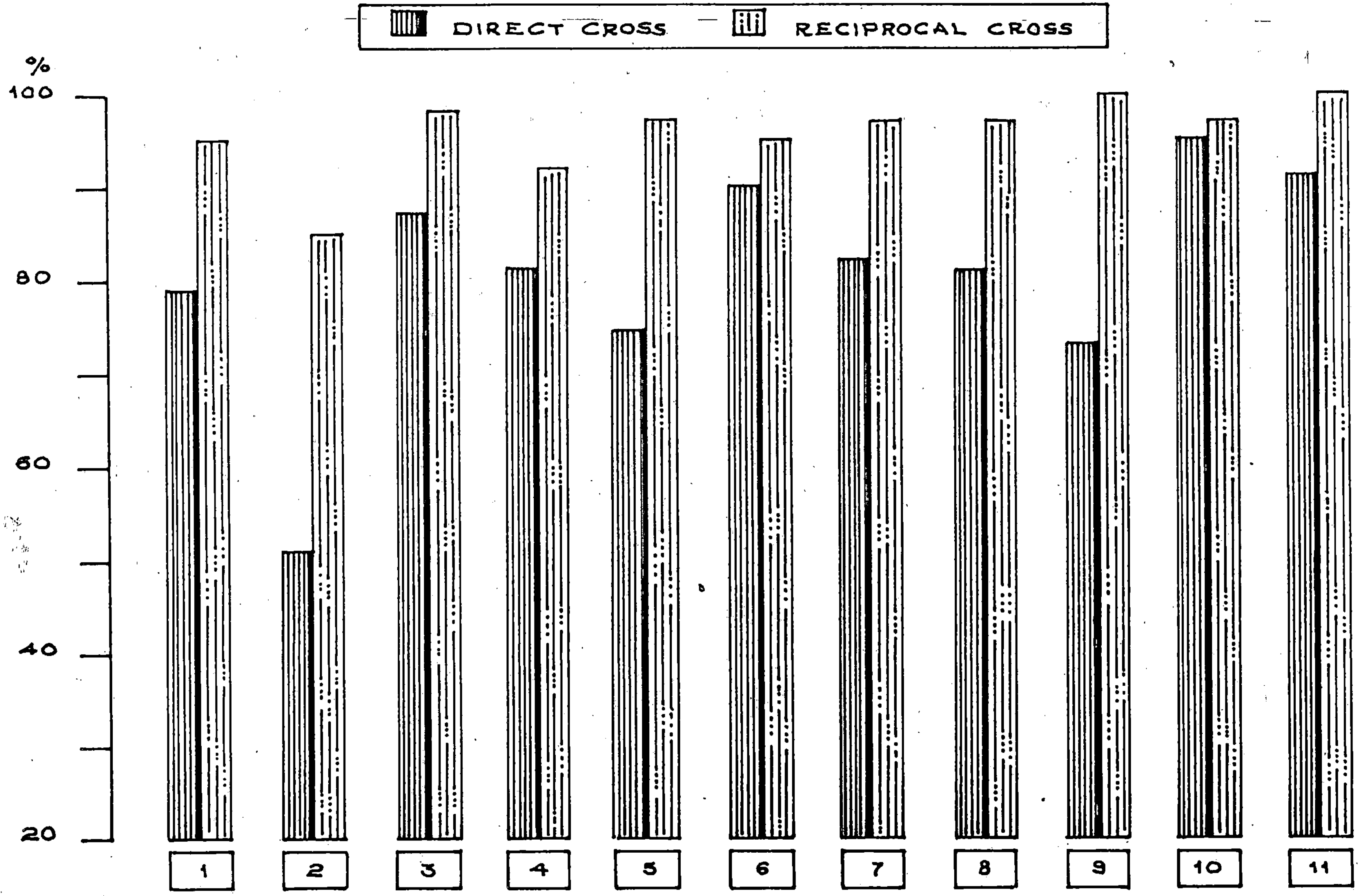
Sl. No.	Crosses	Capsule set			Number of seeds			% of seed set		
		Number of flowers crossed	Number of capsules set	% of capsules set	Normal	Medium shri-velled	Highly shri-velled	Normal	Medium shri-velled	Highly shri-velled
1	S.M* x ACV.1	37	35	94.6	0	2235	761	0	74.4	25.6
2	S.M x ACV.2	39	33	84.6	0	1699	637	0	72.6	27.4
3	S.M x IC.284	45	44	97.8	0	2332	1532	0	60.0	40.0
4	S.M x S.8	39	36	92.3	0	1800	808	0	68.8	31.2
5	S.M x P.10.1	35	34	97.1	0	2130	1375	0	61.2	38.8
6	S.M x S1.1275	37	35	94.6	0	1944	1024	0	65.9	34.1
7	S.M x Cul.16	37	36	97.3	0	1973	1339	0	59.4	40.6
8	S.M x Cul.28	33	32	96.8	0	2105	816	0	71.4	28.6
9	S.M x Cul.40	33	33	100.0	0	2245	1244	0	64.2	35.8
10	S.M x Vinayak	37	36	97.3	0	2318	1189	0	66.0	34.0
11	S.M x N-62-32	40	40	100.0	0	2557	1499	0	63.4	36.6
	Mean	37.5	35.8	95.5	0	2122	1111	0	65.6	34.4

*S.M - Sesamum malabaricum

Figure 4. Capsule set in direct and reciprocal crosses of *Sesamum indicum* and *S. malabaricum*.

1. ACV.1 x *S. malabaricum*
2. ACV.2 x *S. malabaricum*
3. IC.284 x *S. malabaricum*
4. S.8 x *S. malabaricum*
5. P.10-1 x *S. malabaricum*
6. Si.1275 x *S. malabaricum*
7. Cul.16 x *S. malabaricum*
8. Cul.28 x *S. malabaricum*
9. Cul.40 x *S. malabaricum*
10. Vinayak x *S. malabaricum*
11. N-62-32 x *S. malabaricum*

FIG. 4. CAPSULE SET IN DIRECT AND RECIPROCAL CROSSES OF *Sesamum indicum* AND *S. malabaricum*



these crosses. All the seeds in each cross were either medium shrivelled or highly shrivelled (Table 3). In these seeds, the seed coats were normally developed. So the seeds almost resembled the female parent variety in seed colour. Since there were no normal seeds, the number of seeds per capsule were not counted.

b. Seed set in *Sesamum malabaricum* x *S. indicum* crosses:

In these crosses also, normal seeds were not obtained. There were medium shrivelled and highly shrivelled seeds in each cross and hence the seeds obtained from different crosses were similar in appearance (Figure 5b, Table 4). Medium shrivelled seeds presented a partially filled appearance with normal black coloured seed coat similar to the female parent, *S. malabaricum*, but they were empty and crumbled when pressed. A scaly endosperm was seen inside the thick seed coat.

Table 4 shows that percentage of medium shrivelled seeds is more than the percentage of highly shrivelled seeds in all the crosses. Percentage of medium shrivelled seeds was maximum in the cross *S. malabaricum* x ACV.1 (74%) and it was minimum in the cross *S. malabaricum* x Cul.16 (59%).

Figure 5. Seeds of *Sesamum indicum* x *S. malabaricum* crosses.

a) Direct cross

1. ACV.1 x S.M*
2. ACV.2 x S.M
3. IC.284 x S.M
4. S.8 x S.M
5. Si.1275 x S.M
6. R.10-1 x S.M
7. Cul.16 x S.M
8. Cul.28 x S.M
9. Cul.40 x S.M
10. Vinayak x S.M
11. N-62-32 x S.M
12. ACV.1 (standard)

b) Reciprocal cross

1. S.M x ACV.1
2. S.M x ACV.2
3. S.M x IC.284
4. S.M x S.8
5. S.M x Si.1275
6. S.M x P.10-1
7. S.M x Cul.16
8. S.M x Cul.28
9. S.M x Cul.40
10. S.M x Vinayak
11. S.M x N-62-32
12. ACV.1 (standard)

*S.M - *Sesamum malabaricum*

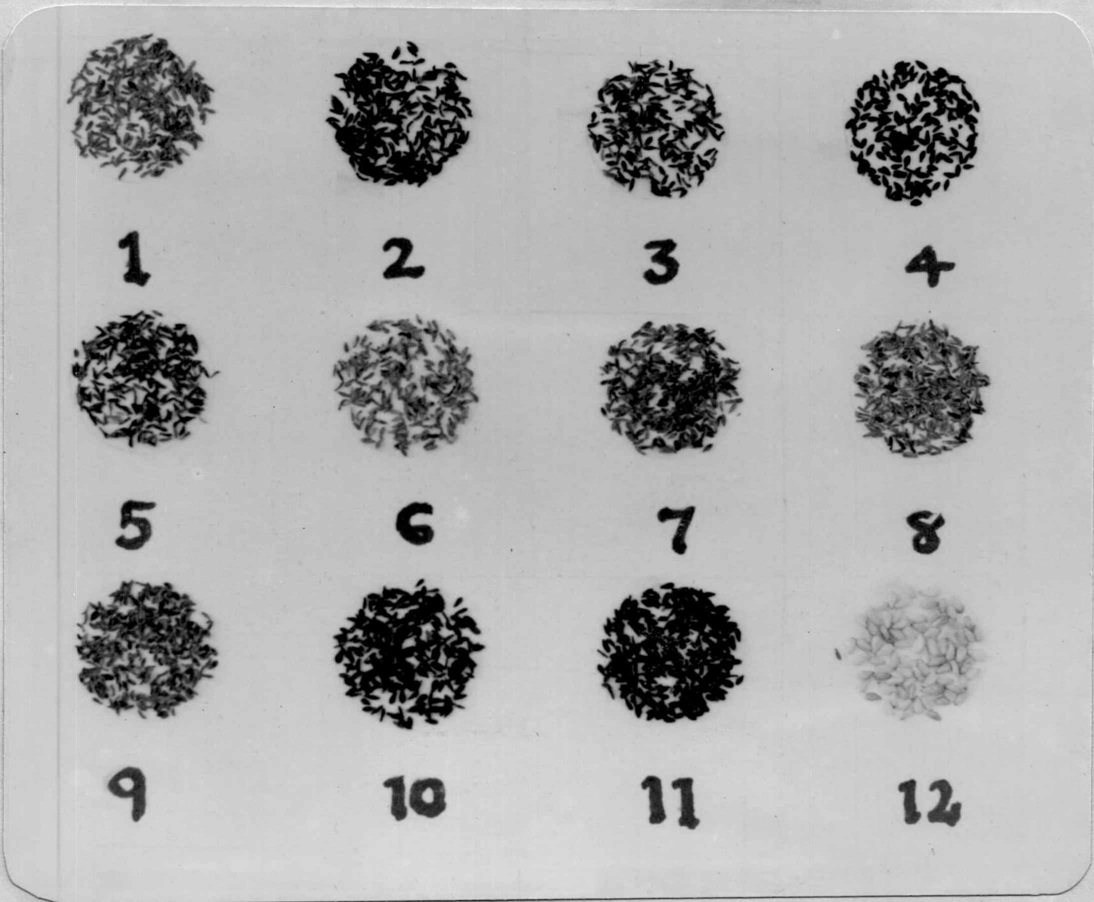


Figure 5a

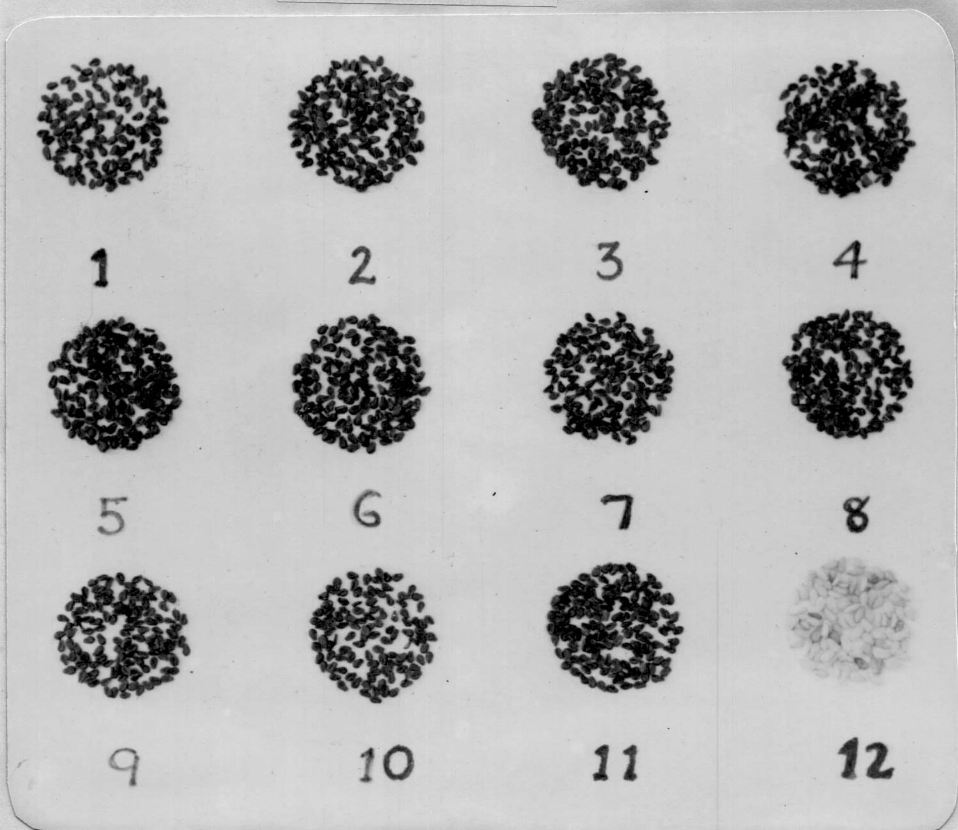


Figure 5b

Characteristics of crossed seeds of selected varieties are clear in Figure 6a to d. In ACV. 1 x S. malabaricum and Cul.40 x S. malabaricum, seeds were highly shrivelled whereas in the other two direct crosses the seeds were medium shrivelled. Seeds from the four reciprocal crosses were similar in all cases.

iii) Seed viability

None of the seeds in the 22 crosses (direct and reciprocal) was normal. Hence there was no need to conduct seed viability test.

2. Pollination and Fertilization

i) Capsule development from emasculated flowers without pollination

No capsule developed from flowers emasculated and protected, without pollination. This was true in all the eleven varieties of S. indicum as well as in the wild sp., S. malabaricum. Data relating to this study are presented in Table 5.

ii) Capsule setting after style excision at different time intervals after self pollination:

The data relating to Sesamum indicum (4 varieties)

Figure 6. Seeds of selected varieties and crosses.

a) ACV.1 x _S_. _malabaricum_

1 - ACV.1

12 - _S_. _malabaricum_

1 x 12 - Direct cross

12 x 1 - Reciprocal cross

b) S.8 x _S_. _malabaricum_

4 - S.8

12 - _S_. _malabaricum_

4 x 12 - Direct cross

12 x 4 - Reciprocal cross

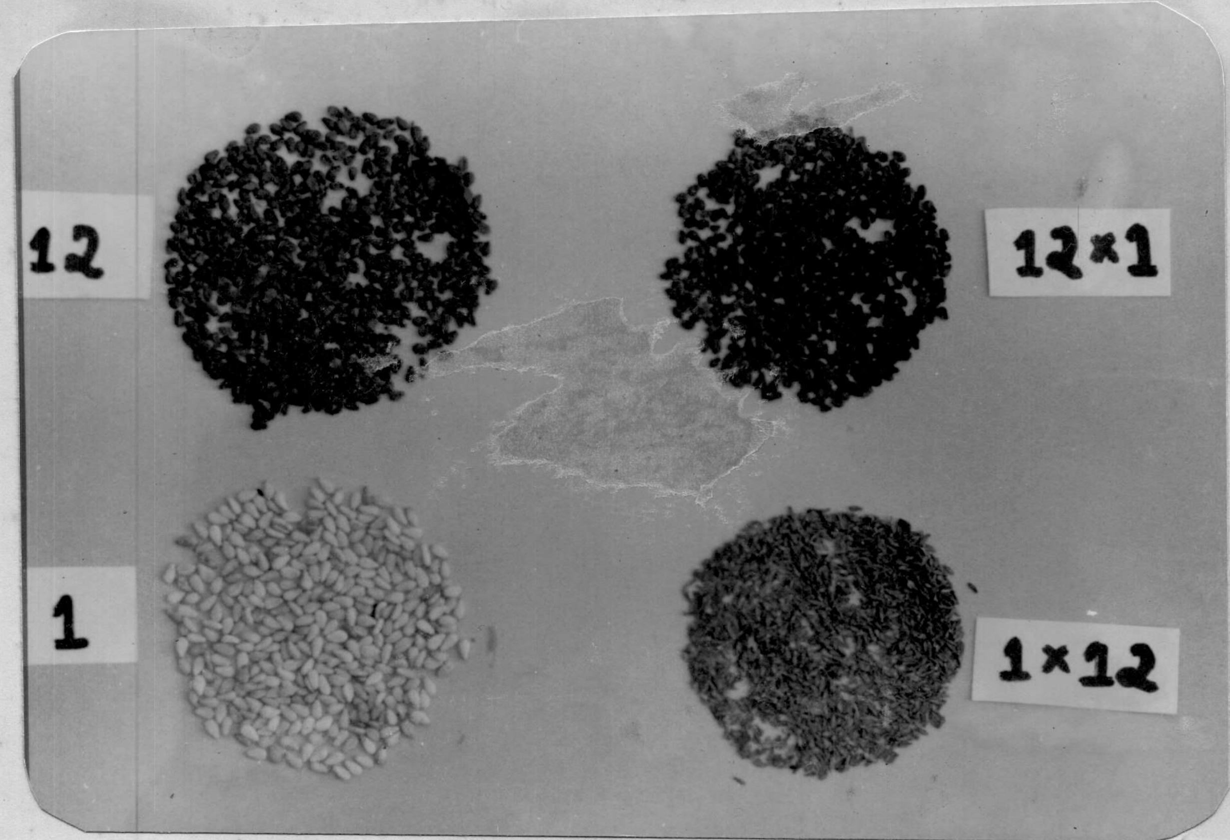


Figure 6a

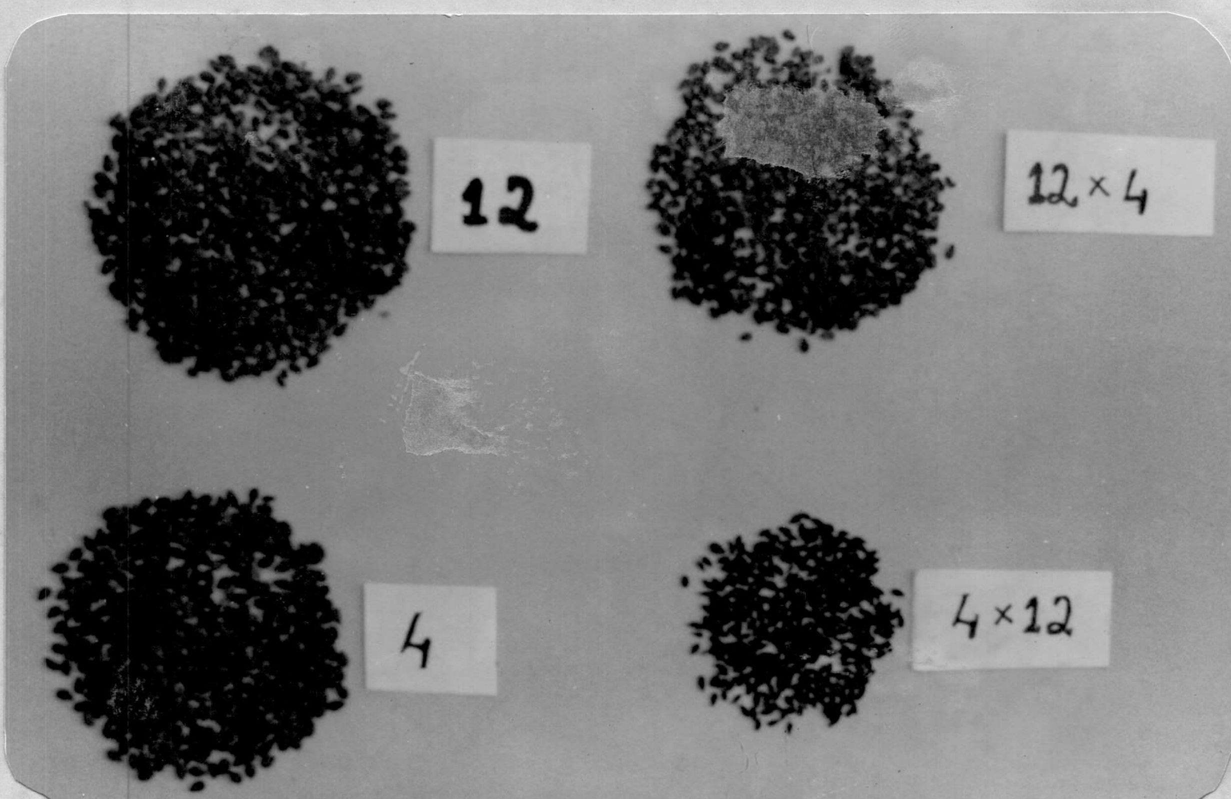


Figure 6b

Figure 6 (contd.)

c) Cul.40 x S. malabaricum

9 - Cul.40

12 - S. malabaricum

9 x 12 - Direct cross

12 x 9 - Reciprocal cross

d) Vinayak x S. malabaricum

10 - Vinayak

12 - S. malabaricum

10 x 12 - Direct cross

12 x 10 - Reciprocal cross

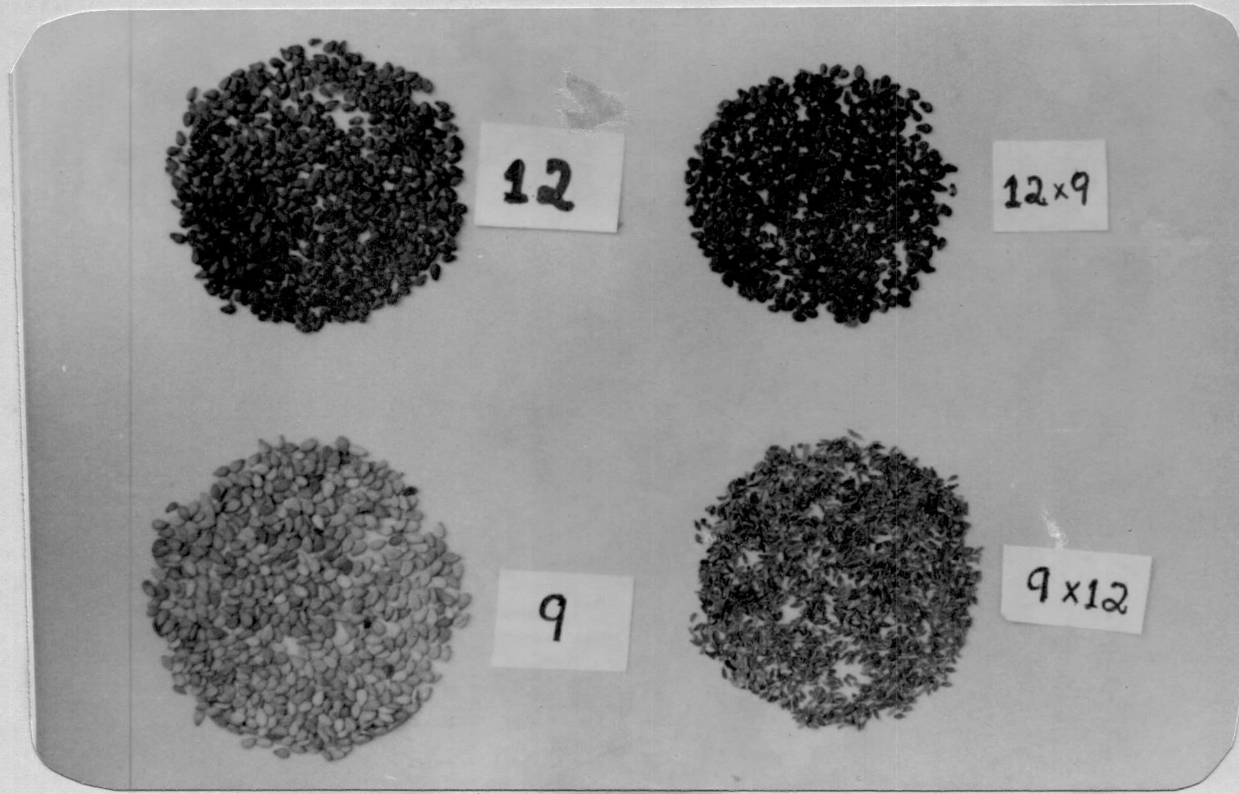


Figure 6c

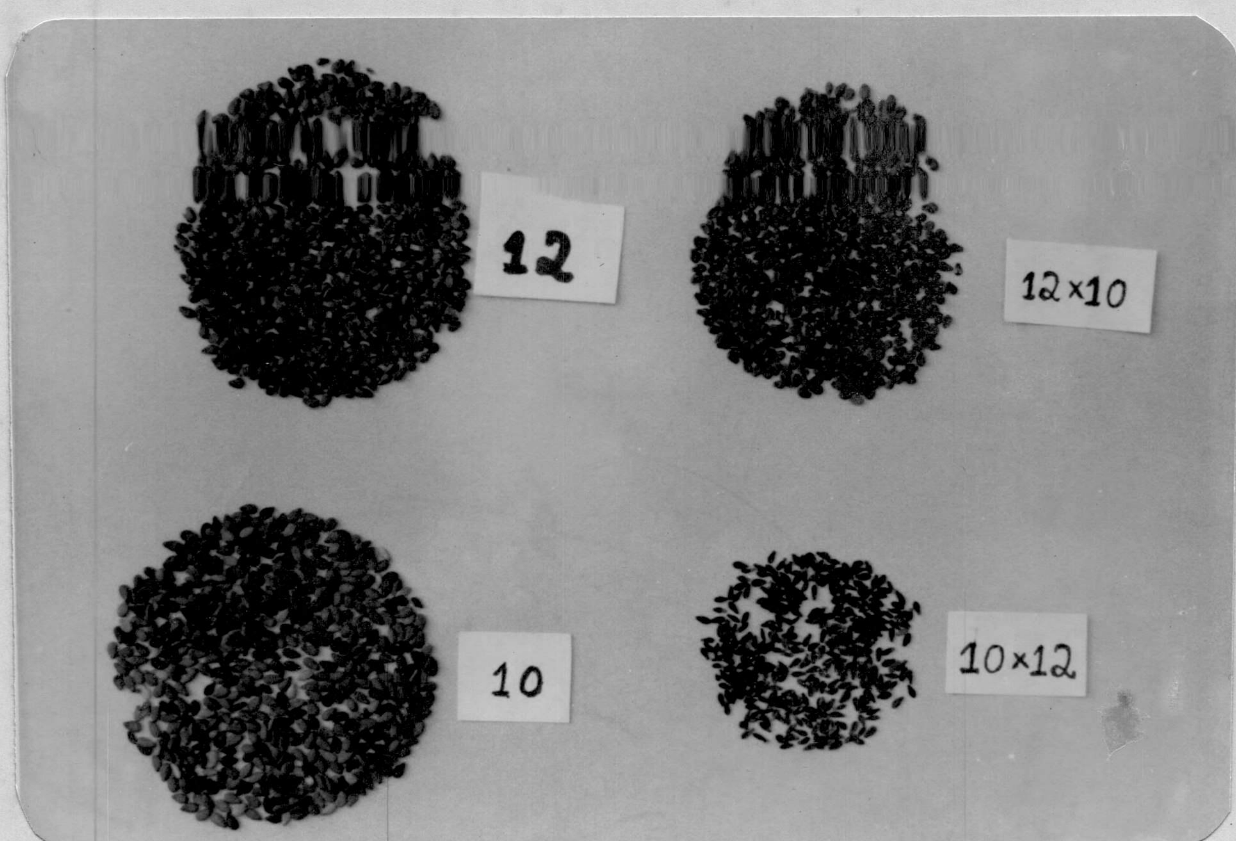


Figure 6d

Table 5. Capsule development in flowers emasculated but not pollinated.

Sl. No.	Variety	Number of plants	Number of flowers emasculated	Number of capsules set
1	ACV.1	8	15	0
2	ACV.2	6	12	0
3	IC.284	8	13	0
4	S.8	4	9	0
5	P10.1	10	22	0
6	Si.1275	5	12	0
7	Cul.16	7	14	0
8	Cul.28	10	24	0
9	Cul.40	5	12	0
10	Vinayak	9	17	0
11	N-62-32	5	15	0
12	S.M*	16	29	0

*S.M - *Sesamum malabaricum*

and *S. malabaricum* are presented in Table 6. Minimum duration for which style is to be retained after pollination for capsule development was four hours in *S. indicum* varieties and five hours in *S. malabaricum*. Style excision after four hour of pollination resulted in 60% capsule set in ACV.1, 33% capsule set in S.8, 25% in Cul.40 and 45% capsule set in Vinayak. Thereafter the percentages increased steadily and attained the peak values sooner or later in different varieties.

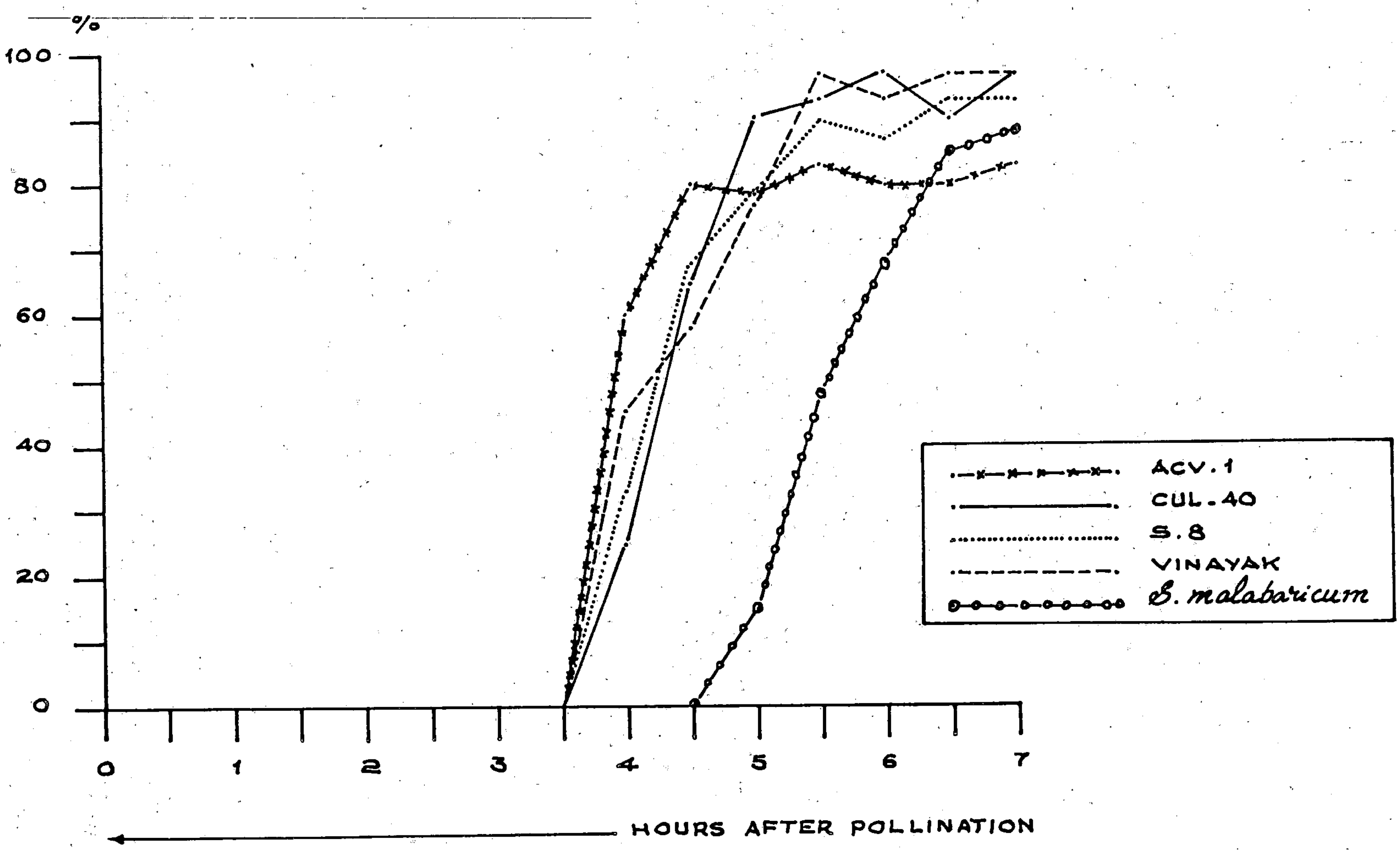
In the case of *S. malabaricum*, style excision after five hours of pollination resulted in only 15% fruit set. It increased with every half hour increase in the duration of style retention. Style excision after 7 hours of pollination resulted in 83% fruit set in ACV.1, 93% fruit set in S.8 as well as the wild species and 97% fruit set in Cul.40 and Vinayak. In the control group, where stigma excision was not done, percentage of fruit set was 70 in ACV.1, 78 in S.8, 80 in Vinayak and 100 in Cul.40 as well as in the wild species. Increase in the percentage of fruit set with increase in the duration of style retention is graphically presented in Figure 7.

Table 6. Capsule setting with style excision at different time intervals after self pollination.

Sl. No.	Time after pollination (hours)	Number of flowers pollinated					Number of capsules set					Percentage of capsule set				
		ACV.1	S.8	Cul.40	Vinayak	S.M*	ACV.1	S.8	Cul. 40	Vinayak	S.M	ACV.1	S.8	Cul. 40	Vinayak	S.M
1	0.5	10	10	10	10	10	0	0	0	0	0	0	0	0	0	0
2	1.0	10	10	10	9	10	0	0	0	0	0	0	0	0	0	0
3	1.5	10	9	10	10	10	0	0	0	0	0	0	0	0	0	0
4	2.0	9	10	10	10	10	0	0	0	0	0	0	0	0	0	0
5	2.5	10	10	9	8	10	0	0	0	0	0	0	0	0	0	0
6	3.0	38	40	40	39	40	0	0	0	0	0	0	0	0	0	0
7	3.5	39	40	40	39	40	0	0	0	0	0	0	0	0	0	0
8	4.0	40	40	40	40	40	24	13	10	18	0	60	33	25	45	0
9	4.5	39	39	40	36	40	31	26	26	21	0	80	67	65	58	0
10	5.0	38	40	40	35	40	30	30	36	27	6	79	79	90	77	15
11	5.5	30	30	40	30	40	25	27	37	29	19	83	90	93	97	48
12	6.0	30	30	30	30	40	24	26	29	28	27	80	87	97	93	68
13	6.5	30	30	30	30	40	24	28	27	29	36	80	93	90	97	90
14	7.0	30	30	30	30	40	25	28	29	29	37	83	93	97	97	93
15	Normal flowers	10	9	10	10	7	7	7	10	8	10	70	78	100	80	100

*S.M - Sesamum malabaricum

FIG. 7. CAPSULE SET AFTER STYLE EXCISION



iii) Microscopic study of pollen tube growth

Pollen tubes which were stained dark blue were clearly visible in the light blue surrounding medium.

Styles studied one hour after pollination showed that in selfed flowers of S.8, the pollen tubes have traversed about 30 to 40% of the stylar length. In the styles of flowers crossed with S. malabaricum, pollen tubes were just entering into the styles at that time. Three to three and a half hours after pollination in selfed S.8 flowers, pollen tubes were seen just above the top of the ovary. While some of the tubes have reached the ovary, some others were traversing the lower part of the style. In the case of crossed S.8 flowers, the pollen tubes have by then reached only 50 to 60% of the length of the styles. Pollen tubes have taken about five hours to reach the ovary in crossed S. indicum (S.8) flowers. From this it is clear that pollen tube growth is comparatively slower in crossed flowers than in selfed flowers of S. indicum.

In S. malabaricum the rate of pollen tube growth was slightly higher in crossed flowers than in selfed flowers. One hour after pollination in selfed flowers,

Figure 8. Pollen germination and pollen tube growth in pistil.

a. Pollen germination in _Sesamum indicum_ x _S. malabaricum_ cross.

b. Pollen tube growth in the style in _Sesamum indicum_ x _S. malabaricum_ cross.

Figure 8a

8 x 4.1

Figure 8b

8 x 4.1

pollen tubes were seen just entering into the styles, while in crossed flowers pollen tubes have traversed 10 to 20% of the stylar length. Three hours after pollination in selfed and crossed flowers pollen tubes were observed to traverse 50% and 60% of the stylar length respectively. Pollen tubes were seen entering the ovary four to four and a half hours after pollination in both selfed as well as crossed flowers.

Pollen germination and pollen tube growth in crossed flowers of *S. indicum* are shown in Figure 8a and b. Pollen tubes show normal growth in crossed flowers as in selfed flowers.

3. Capsule and seed development.

i) Length of capsules

Mean length of capsules in direct and reciprocal crosses of *S. indicum* x *S. malabaricum* at two days intervals from pollination to 20 days thereafter are presented in Tables 7 and 8. Development of crossed capsules was at a slower rate as compared to the open pollinated ones. Capsules attained full length at about 10 days after pollination.

Table 7. Length of capsules (cm) under natural and cross pollinations in <u>Sesamum indicum</u> varieties.

Sl. No.	Number of days after pollination.	ACV.1	ACV.1 x S.M*	S.8	S.8 x S.M	Cul.40	Cul.40 x S.M	Vinayak	Vinayak x S.M
1	2	0.84	0.82	0.69	0.51	0.75	0.71	0.72	0.66
2	4	1.23	1.19	1.21	1.02	1.50	1.42	1.55	1.14
3	6	1.88	1.64	1.88	1.66	2.40	1.96	2.19	1.99
4	8	2.25	2.29	2.22	1.67	2.95	2.55	2.90	2.43
5	10	2.59	2.28	2.36	2.27	2.97	2.54	3.23	3.12
6	12	2.56	2.30	2.51	2.06	3.05	2.33	3.13	3.01
7	14	2.49	2.34	2.39	2.06	3.01	2.33	3.21	2.97
8	16	2.65	2.15	2.34	2.17	3.00	2.83	3.49	2.84
9	18	2.58	2.20	2.52	2.16	2.98	2.66	3.39	3.06
10	20	2.57	2.42	2.52	2.21	3.02	2.42	3.48	3.23

*S.M - <u>Sesamum malabaricum</u>

Table 8. Length of capsules (cm) under natural and cross pollinations in *Sesamum malabaricum*.

Sl. No.	Number of days after pollinations.	S.M*	S.M x ACV.1	S.M x S.8	S.M x Cul.40	S.M x Vinayak
1	2	0.72	0.56	0.60	0.59	0.71
2	4	1.53	1.38	1.24	1.24	1.47
3	6	2.30	1.90	2.00	2.10	2.20
4	8	2.71	2.15	2.57	2.29	2.61
5	10	2.89	2.43	2.39	2.78	2.64
6	12	2.82	2.48	2.51	2.59	2.56
7	14	2.79	2.52	2.46	2.46	2.72
8	16	2.80	2.34	2.59	2.51	2.61
9	18	2.76	2.48	2.66	2.50	2.72
10	20	2.86	2.44	2.67	2.52	2.84

*S.M - *Sesamum malabaricum*

There was slight reduction in the length of crossed capsules at all the developmental stages, the percentage of reduction varying in different crosses at a particular stage and within the same cross at different stages of growth (Table 7). In ACV.1 crosses, there was 2%, 12% and 6% reduction after 2, 10 and 20 days respectively after pollination, as compared to the open pollinated capsules. In S.8 crossed, the reduction was 27%, 4% and 12% respectively at the 3 stages. In Cul.40 crossed it was 5%, 15% and 20% and in Vinayak crossed it was 7%, 3% and 15% respectively. Capsule length in the above crosses in relation to number of days after pollination is diagramatically presented in Figure 9a to d.

In the reciprocal crosses also, there was slight reduction in the length of capsules at all stages of development (Table 8). Capsule length in these crosses is graphically presented in Figure 10. Here also capsules attained full size within 10 days after pollination. After 2 days of pollination the percentage of reduction in capsule length was maximum in S. malabaricum x ACV.1 (23%) and minimum in S. malabaricum x Vinayak (1.4%). In S. malabaricum x S.8 and S. malabaricum x Cul.40 it was

FIG.9. LENGTH OF CAPSULES AT DIFFERENT STAGES OF DEVELOPMENT IN DIRECT CROSSES

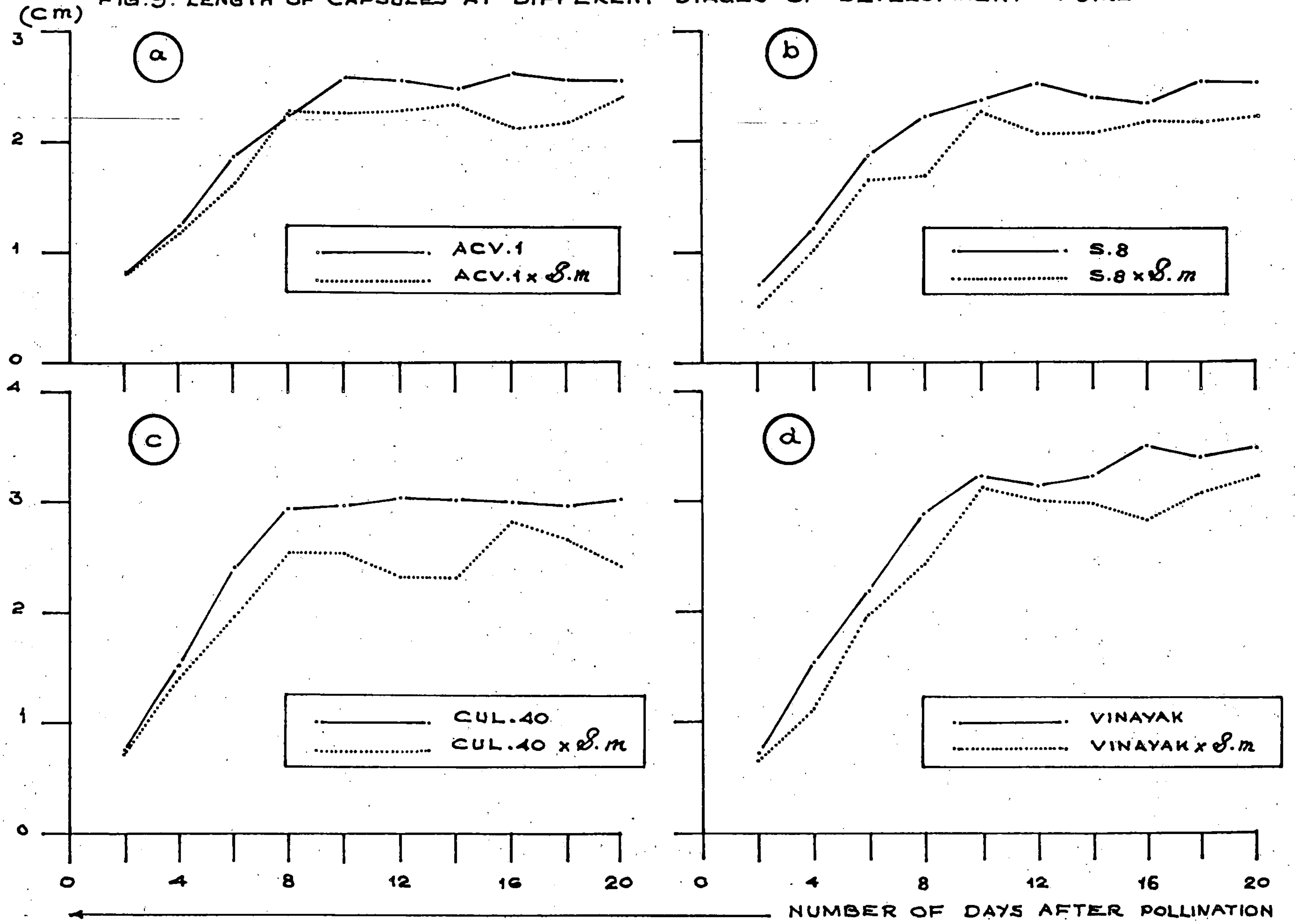
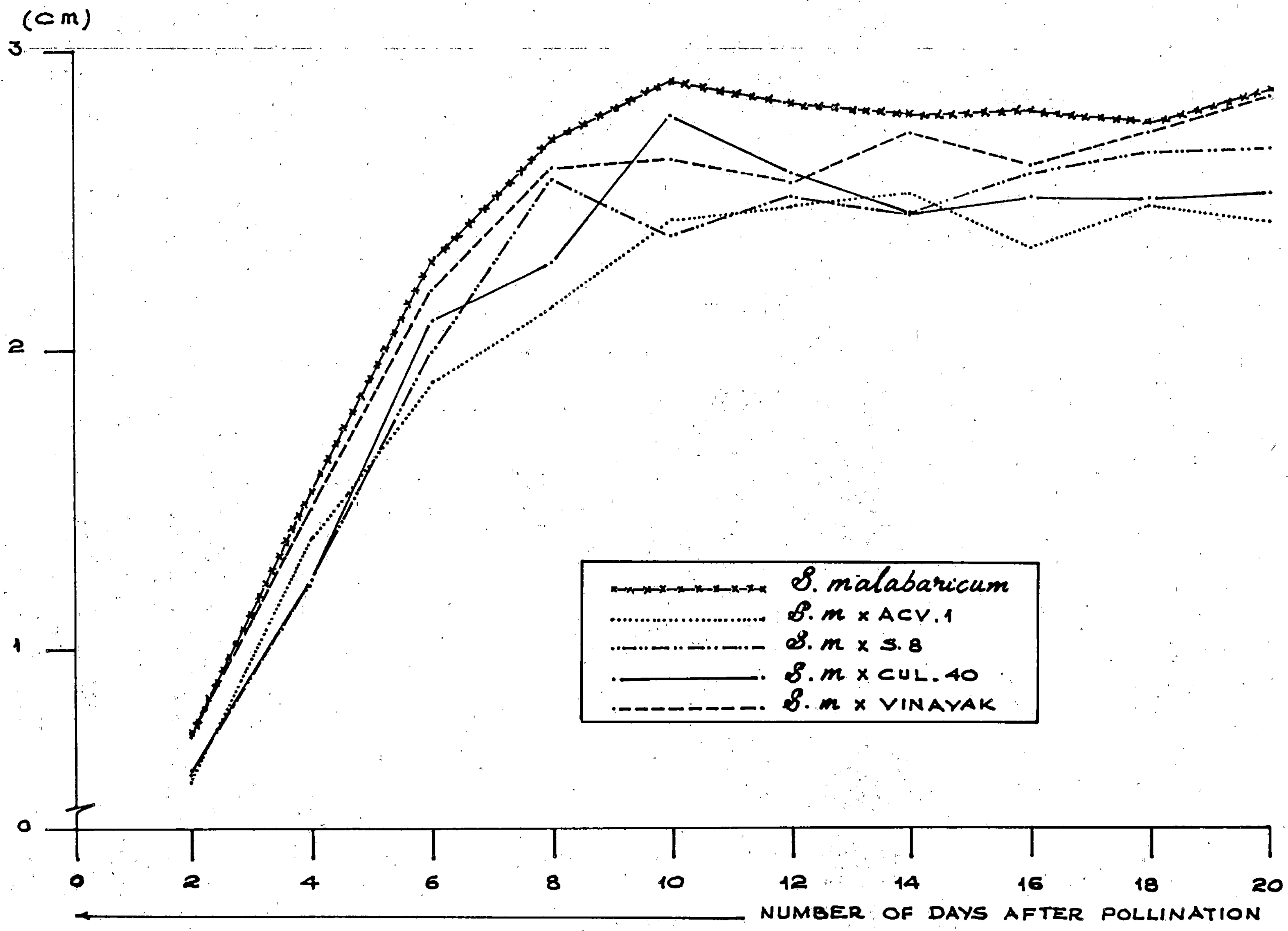


FIG. 10. LENGTH OF CAPSULES AT DIFFERENT STAGES OF DEVELOPMENT IN RECIPROCAL CROSSES



17% and 18% respectively. Ten days after pollination, S. malabaricum x S.8 showed maximum percentage of reduction (17%) and in S. malabaricum x Cul.40 the reduction was minimum (3.8%). In S. malabaricum x ACV.1 and in S. malabaricum x Vinayak it was 16% and 9% respectively. After 20 days of pollination, the percentages of reduction were 15, 7, 12 and 0.7 in S. malabaricum x ACV.1, S. malabaricum x S.8, S. malabaricum x Cul.40 and S. malabaricum x Vinayak respectively.

ii) Breadth of capsules

Similar to length, the mean breadth of capsules also showed slight reduction following crossing at all the developmental stages. In Cul.40 x S. malabaricum and Vinayak x S. malabaricum at 2 days after pollination, the breadth of capsules was equal to that of open pollinated ones. In ACV.1 x S. malabaricum and S.8 x S. malabaricum the reduction was 9% and 20% respectively. After 10 days from pollination, the mean breadth of capsules showed 7%, 6%, 6% and 12% reductions in ACV.1 x S. malabaricum, S.8 x S. malabaricum, Vinayak x S. malabaricum and Cul.40 x S. malabaricum respectively. After 20 days of pollination there was only 1% reduction in

ACV.1 x *S. malabaricum*. The percentages of reduction in the other three crosses were 9, 15 and 6 respectively. Mean breadth of capsules in direct crosses at different stages of growth are given in Table 9 and the data are graphically presented in Figure 11a to d.

In reciprocal crosses, the differences in percentages of reduction in mean breadth of capsules were very small. Mean breadth of capsules after 2 days of pollination in the wild species (open pollinated) and *S. malabaricum* x Vinayak was 0.22 cm. The means were 0.18 cm in *S. malabaricum* x ACV.1 and 0.20 cm in *S. malabaricum* x S.8 and *S. malabaricum* x Cul.40. After 10 days of pollination, the breadth was 0.80 cm in open pollinated capsules and in the cross *S. malabaricum* x Vinayak. In *S. malabaricum* x ACV.1, *S. malabaricum* x S.8 and *S. malabaricum* x Cul.40 it was 0.74 cm, 0.70 cm and 0.72 cm respectively. After 20 days of pollination, the mean breadth was 0.78 cm in open pollinated and in *S. malabaricum* x Vinayak cross. *S. malabaricum* x ACV.1, *S. malabaricum* x S.8 and *S. malabaricum* x Cul.40 recorded 0.75 cm, 0.72 cm and 0.68 cm respectively. Mean breadth of capsules in reciprocal crosses at different stages of development are presented

Table 9. Breadth of capsules (cm) under natural and cross pollinations in <u>Sesamum indicum</u> varieties.

Sl. No.	Number of days after pollination.	ACV.1	ACV. 1 x S.M*	S.8	S.8 x S.M	Cul.40	Cul.40 x S.M	Vinayak	Vinayak x S.M
1	2	0.32	0.29	0.20	0.16	0.20	0.20	0.18	0.18
2	4	0.50	0.47	0.32	0.30	0.37	0.38	0.34	0.25
3	6	0.79	0.75	0.56	0.53	0.61	0.57	0.57	0.51
4	8	0.94	0.94	0.65	0.60	0.79	0.71	0.73	0.61
5	10	1.05	0.98	0.72	0.68	0.83	0.73	0.87	0.82
6	12	1.07	1.02	0.79	0.61	0.83	0.70	0.89	0.79
7	14	1.12	1.05	0.71	0.60	0.86	0.68	0.89	0.82
8	16	1.11	0.99	0.71	0.62	0.81	0.79	0.88	0.76
9	18	1.10	0.96	0.74	0.67	0.84	0.74	0.87	0.80
10	20	1.06	1.05	0.75	0.68	0.83	0.71	0.84	0.79

*S.M - <u>Sesamum malabaricum</u>

Table 10. Breadth of capsules (cm) under natural and cross pollinations in *Sesamum malabaricum*.

Sl. No.	Number of days after pollination.	S.M*	S.M x ACV.1	S.M x S.8	S.M x Cul.40	S.M x Vinayak
1	2	0.22	0.18	0.20	0.20	0.22
2	4	0.40	0.40	0.35	0.34	0.41
3	6	0.62	0.55	0.54	0.51	0.65
4	8	0.75	0.71	0.71	0.64	0.80
5	10	0.80	0.74	0.70	0.72	0.80
6	12	0.78	0.77	0.72	0.70	0.80
7	14	0.80	0.82	0.70	0.70	0.70
8	16	0.79	0.76	0.73	0.69	0.76
9	18	0.75	0.75	0.73	0.67	0.75
10	20	0.78	0.75	0.72	0.68	0.78

*S.M - *Sesamum malabaricum*

FIG. 11. BREADTH OF CAPSULES AT DIFFERENT STAGES OF DEVELOPMENT IN DIRECT CROSSES

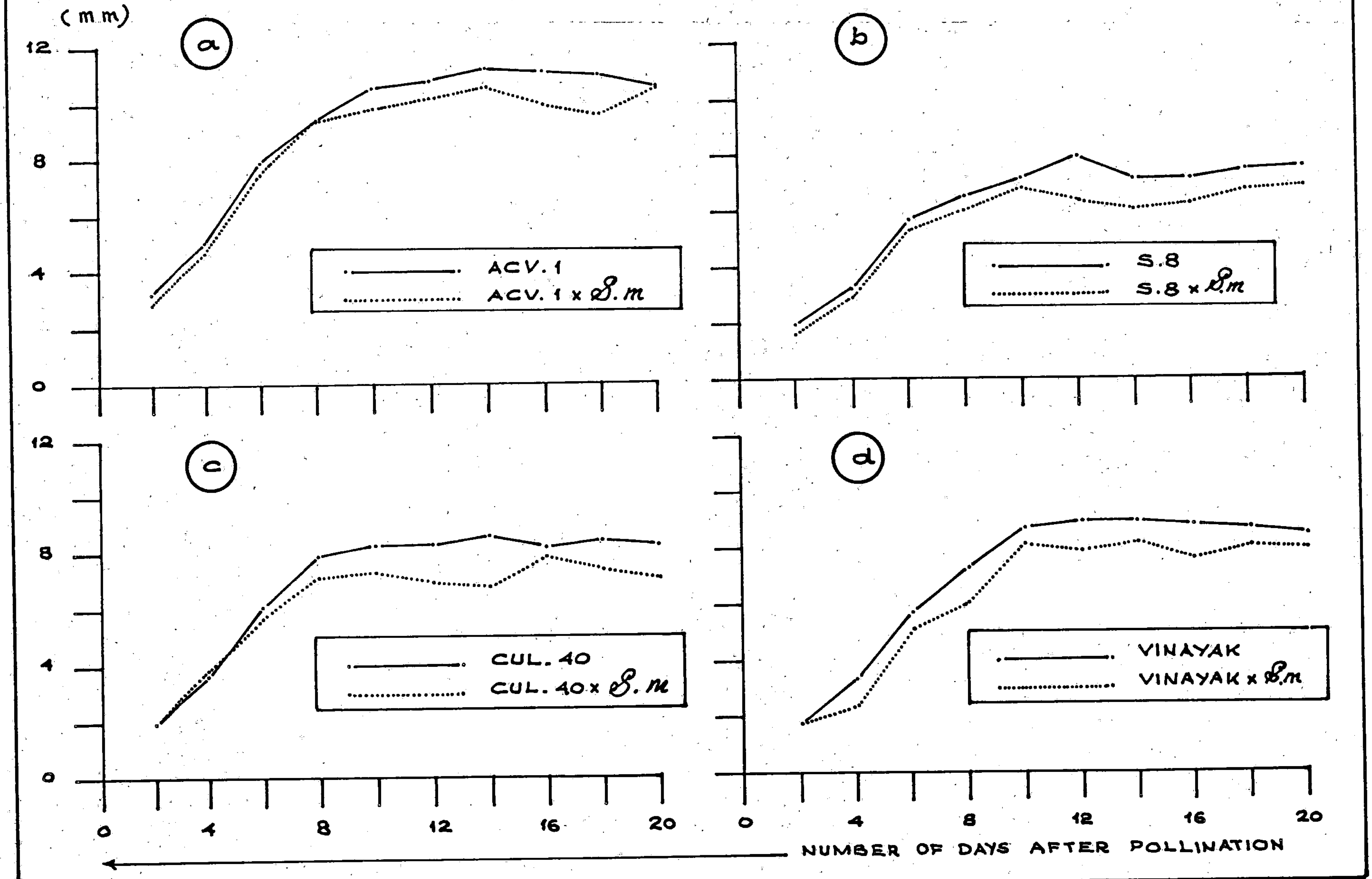


FIG. 12. BREADTH OF CAPSULES AT DIFFERENT STAGES OF DEVELOPMENT IN RECIPROCAL CROSSES

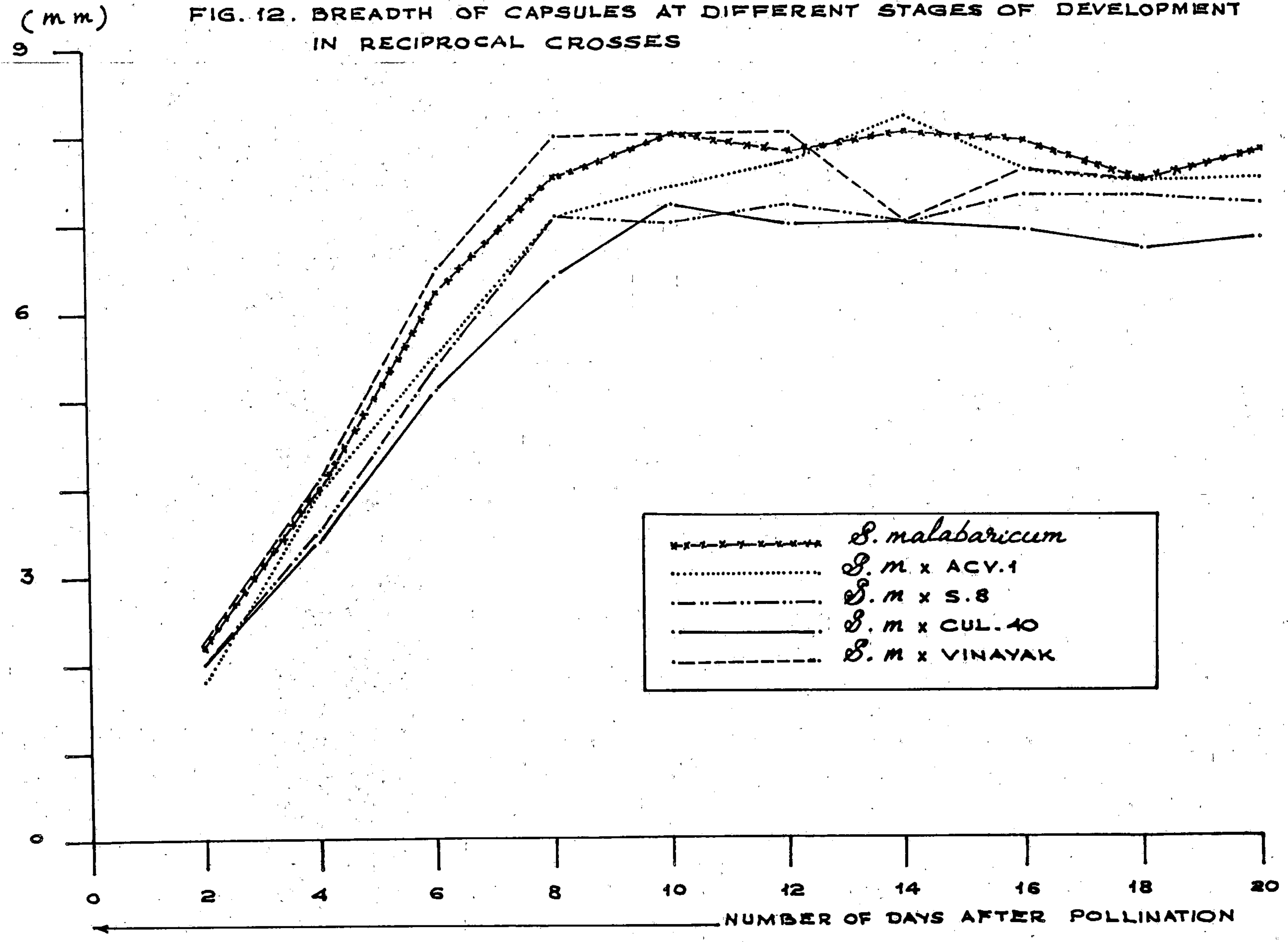


Figure 13. Capsule development in S.8 x *S. malabaricum* crosses.

a and d - 10 days after pollination

b and e - 20 days after pollination

c and f - 30 days after pollination

4 - S.8

12 - *S. malabaricum*

4 x 12 - Direct cross

12 x 4 - Reciprocal cross

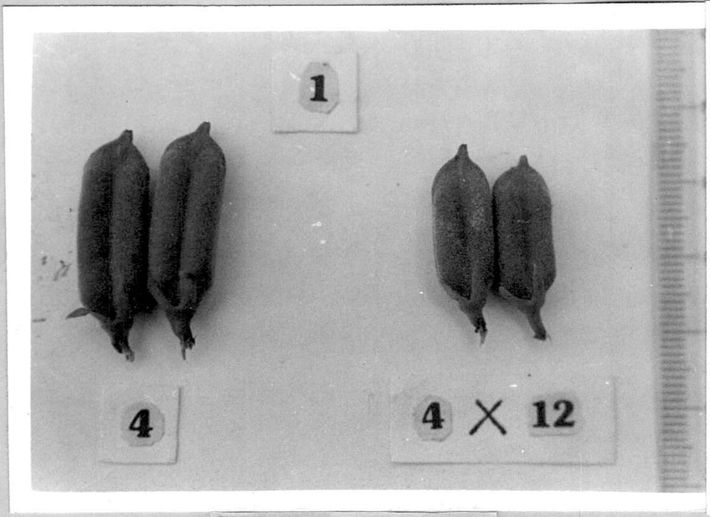


Figure 13a

Figure 13d

Figure 13b

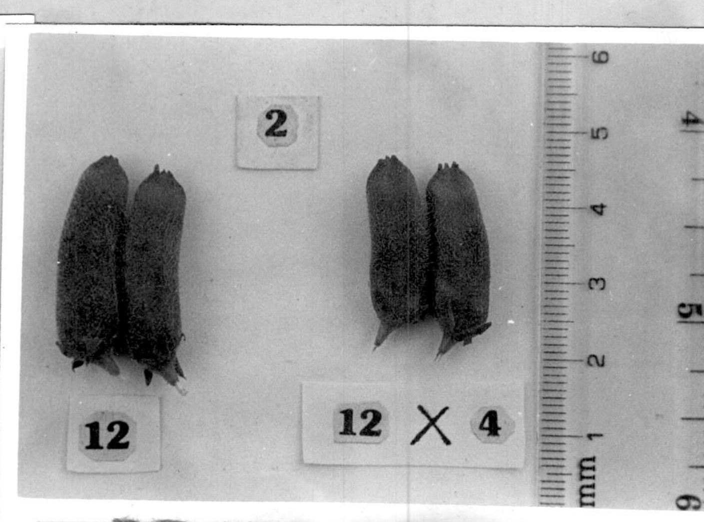


Figure 13e

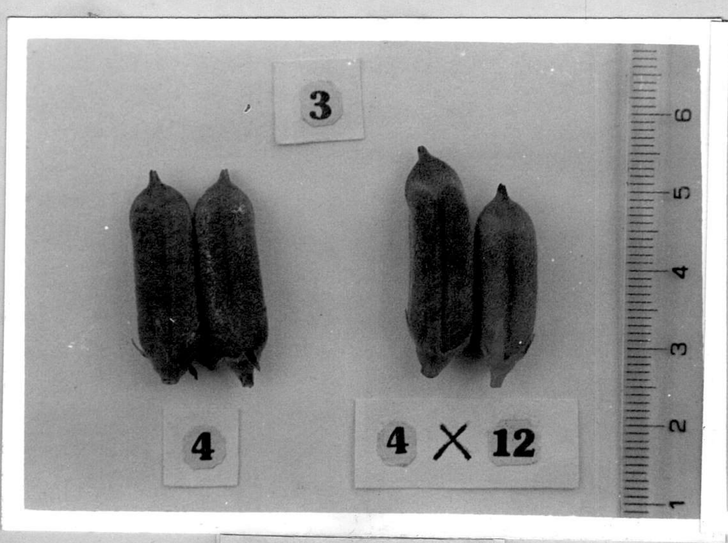


Figure 13c

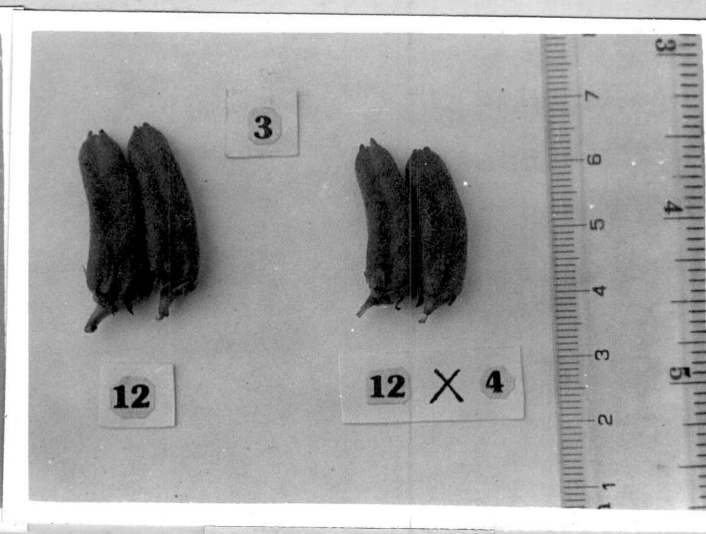


Figure 13f

in Table 10 and the data are graphically presented in Figure 12. Capsule development in direct and reciprocal crosses between S.8 and S. malabaricum at 10 days, 20 days and 30 days after pollination are shown in Figure 13a to f.

iii) Number of seeds per capsule

Mean number of seeds per capsule in direct and reciprocal crosses of Sesamum indicum varieties with S. malabaricum in comparison to their respective female parents are given in Tables 11 and 12. Data on seed set are graphically presented in Figure 14a and b. Among the four varieties of S. indicum, the mean number of seeds per capsule was maximum in ACV.1 (90) and minimum in S.8(56). There was substantial reduction in number of seeds per capsule in all the direct and reciprocal crosses from the respective female parents. Among the four direct crosses, ACV.1 x S. malabaricum showed 34 percent reduction in number of seeds per capsule and S.8 x S. malabaricum showed 29 percent reduction. In Cul.40 x S. malabaricum and Vinayak x S. malabaricum it was 31 percent. In reciprocal crosses, the percentages of reduction were 25, 27, 32 and 20 in S. malabaricum x ACV.1, S. malabaricum x

Table 11. Number of seeds per capsule under natural and cross pollinations in *Sesamum indicum* varieties.

Sl. No.	Number of days after pollination.	ACV.1	ACV.1 x S.M*	S.8	S.8 x S.M	Cul.40	Cul.40 x S.M	Vinayak	Vinayak x S.M
1	2	89	70	53	43	67	59	75	63
2	4	85	56	53	35	65	56	72	51
3	6	90	57	54	40	71	49	78	53
4	8	92	66	56	35	73	57	77	42
5	10	84	58	58	40	74	49	76	55
6	12	87	61	56	38	78	39	75	53
7	14	90	58	54	39	72	46	72	54
8	16	95	50	57	47	71	55	74	45
9	18	96	50	56	38	71	54	75	47
10	20	93	59	58	43	75	37	76	54
Mean	-	90	59	56	40	72	50	75	52

*S.M - *Sesamum malabaricum*

Table 12. Number of seeds per capsule under natural and cross pollinations in *Sesamum malabaricum*.

Sl. No.	Number of days after pollination.	S.M*	S.M x ACV.1	S.M x S.8	S.M x Cul.40	S.M x Vinayak
1	2	83	60	87	87	76
2	4	82	68	67	59	69
3	6	94	68	68	58	69
4	8	94	79	70	57	80
5	10	114	80	56	70	66
6	12	89	67	60	62	67
7	14	100	70	61	61	77
8	16	94	61	66	59	75
9	18	85	77	72	58	78
10	20	94	66	72	57	85
Mean	-	93	70	68	63	74

*S.M - *Sesamum malabaricum*

Figure 14. Seed set in direct and reciprocal crosses.

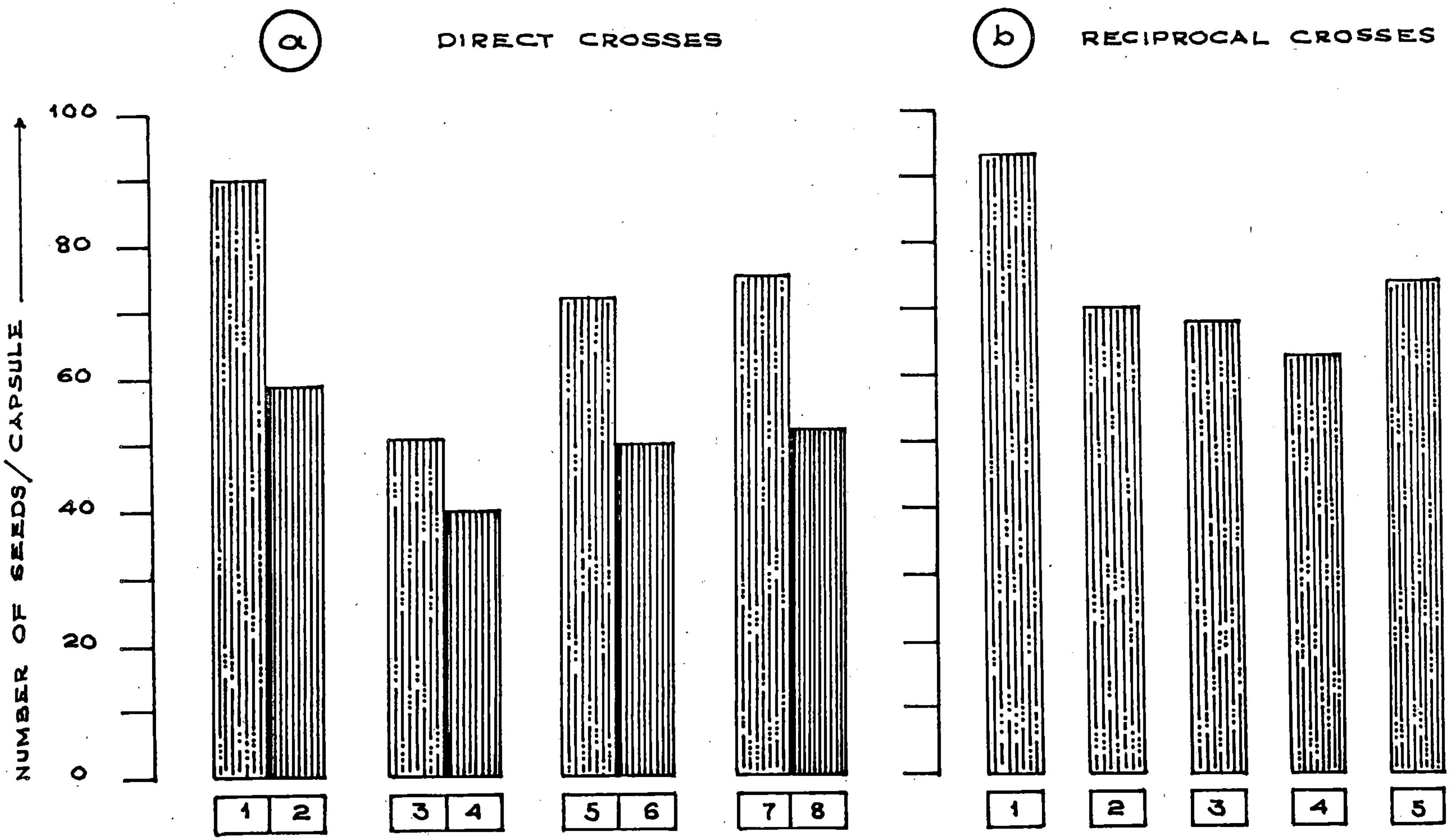
a) Direct crosses

1. ACV.1
2. ACV.1 x *S. malabaricum*
3. S.8
4. S.8 x *S. malabaricum*
5. Cul.40
6. Cul.40 x *S. malabaricum*
7. Vinayak
8. Vinayak x *S. malabaricum*

b) Reciprocal crosses

1. *S. malabaricum*
2. *S. malabaricum* x ACV.1
3. *S. malabaricum* x S.8
4. *S. malabaricum* x Cul.40
5. *S. malabaricum* x Vinayak

FIG. 14. SEED SET IN DIRECT AND RECIPROCAL CROSSES



S.8, S. malabaricum x Cul.40 and S. malabaricum x Vinayak respectively.

iv) Fresh seed size

Mean seed length at 2 days intervals upto 20 days from pollination in the four varieties of S. indicum and their direct crosses with S. malabaricum are presented in Table 13. Development of crossed seeds was slower than the open pollinated seeds of the respective female parents. At the initial stage of development (Two days after pollination) the difference in length between open pollinated and crossed seeds was 38% in ACV.1. Percentage of reduction became 16 and 15 after 10 and 20 days of pollination respectively. In S.8 x S. malabaricum the percentages of reduction were 30, 12 and 14 at 2, 10 and 20 days after pollination respectively. In Cul.40 x S. malabaricum, the percentages of reduction were 12, 7 and 11 respectively. In the case of Vinayak x S. malabaricum the crossed seeds exhibited 19%, 13% and 13% reduction after 2, 10 and 20 days of pollination. Maximum seed length was attained at 10 to 12 days after pollination in all varieties and crosses.

Table 13. Length of fresh seeds (mm) under natural and cross pollinations in *Sesamum indicum* varieties.

Sl. No.	Number of days after pollination.	ACV.1	ACV.1 x S.M*	S.8	S.8 x S.M	Cul.40	Cul.40 x S.M	Vinayak	Vinayak x S.M
1	2	0.85	0.53	0.80	0.56	0.76	0.67	0.70	0.57
2	4	1.68	1.44	1.34	1.10	1.66	1.39	1.55	1.23
3	6	2.42	2.38	2.42	2.13	2.78	2.41	2.45	2.13
4	8	3.65	3.16	3.12	2.73	3.49	3.57	3.07	2.75
5	10	4.08	3.43	3.31	2.92	3.67	3.43	3.68	3.21
6	12	4.15	3.49	3.36	3.07	3.72	3.31	3.72	3.35
7	14	4.00	3.44	3.45	2.78	3.82	3.24	3.68	3.39
8	16	4.11	3.49	3.36	2.84	3.56	3.36	3.77	3.24
9	18	4.18	3.54	3.42	3.08	3.71	3.38	3.76	3.31
10	20	4.12	3.51	3.47	3.00	3.74	3.36	3.74	3.26

*S.M - *Sesamum malabaricum*

Table 14 represents the mean seed length in Sesamum malabaricum and the four reciprocal crosses at 2 days intervals upto 20 days after pollination. Not much difference in the seed length between open pollinated and crossed seeds, at different stages of growth was noticed in this case. Two days after pollination, seed length in open pollinated capsules of S. malabaricum and S. malabaricum x Vinayak was 0.63 mm. Percentages of reduction in S. malabaricum x ACV.1 and S. malabaricum x Cul.40 were 14 and in S. malabaricum x S.8 it was 16. Ten days after pollination the percentages of reduction were 15, 23, 16 and 11 respectively in the four crosses. After 20 days of pollination these values became 15 (S. malabaricum x ACV.1 and S. malabaricum x S.8), 16 (S. malabaricum x Cul.40) and 19 (S. malabaricum x Vinayak). Seed development in direct and reciprocal crosses of S.8 x S. malabaricum at 10, 20 and 30 days after pollination in comparison to their female parents are given in Figure 15a to f.

v) Dry seed size

Size of dry seeds in ACV.1 x S. malabaricum and S.8 x S. malabaricum in comparison to their female parents are given in Table 15. Data on dry seed size are graphically

Table 14. Length of fresh seeds (mm) under natural and cross pollinations in *Sesamum malabaricum*.

Sl. No.	Number of days after pollination.	S.M*	S.M x ACV.1	S.M x S.8	S.M x Cul.40	S.M x Vinayak
1	2	0.63	0.54	0.53	0.54	0.63
2	4	1.59	1.22	1.11	1.10	1.26
3	6	2.82	2.01	1.93	1.84	1.97
4	8	2.95	2.44	2.48	2.42	2.68
5	10	3.12	2.64	2.42	2.62	2.79
6	12	3.14	2.73	2.61	2.60	2.70
7	14	3.18	2.70	2.63	2.60	2.72
8	16	3.17	2.78	2.66	2.60	2.62
9	18	3.20	2.72	2.64	2.68	2.65
10	20	3.20	2.71	2.72	2.70	2.60

*S.M - *Sesamum malabaricum*

Figure 15. Fresh seeds at different stages of development in S.8 x *S. malabaricum* crosses.

a and d - 10 days after pollination

b and e - 20 days after pollination

c and f - 30 days after pollination

4 - S.8

12 - *S. malabaricum*

4 x 12 - Direct cross

12 x 4 - Reciprocal cross

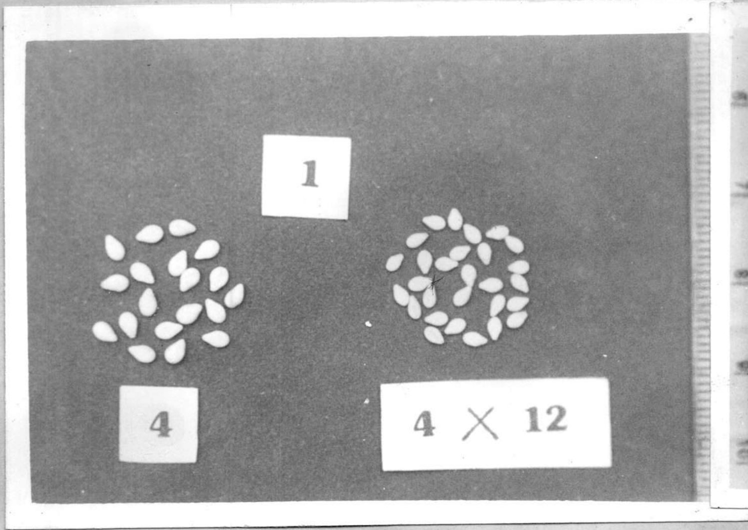


Figure 15a

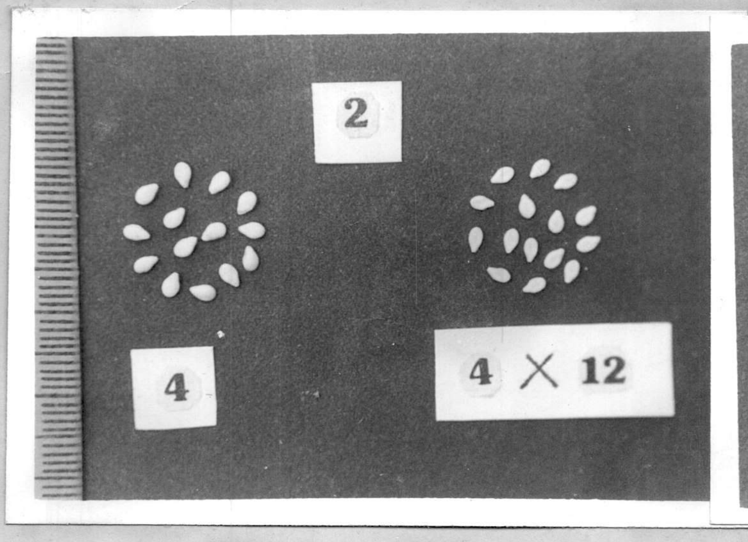


Figure 15 b

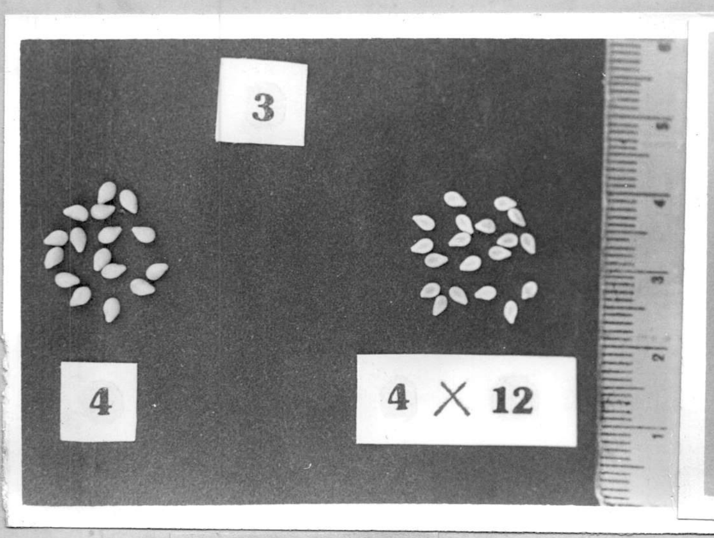


Figure 15c

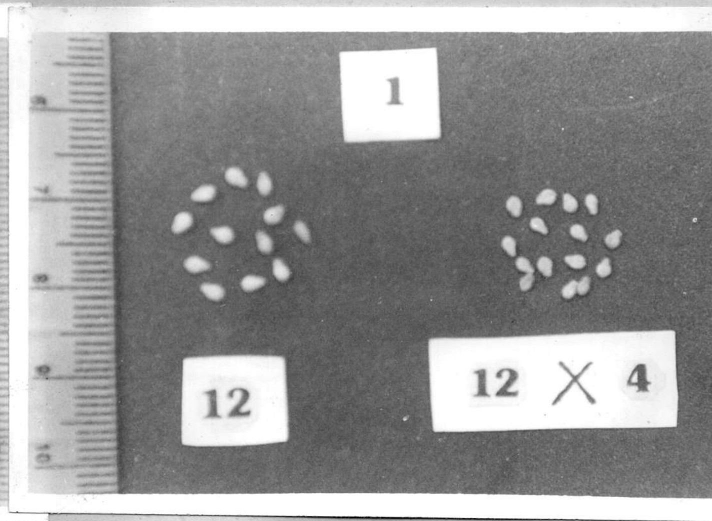


Figure 15d

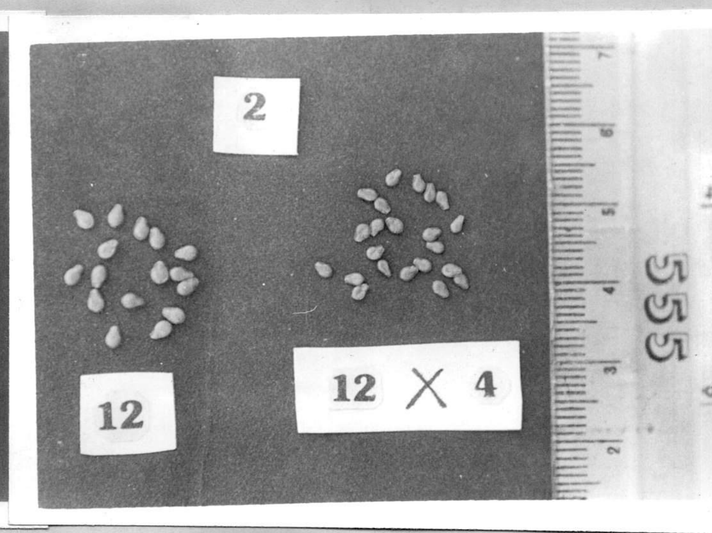


Figure 15e

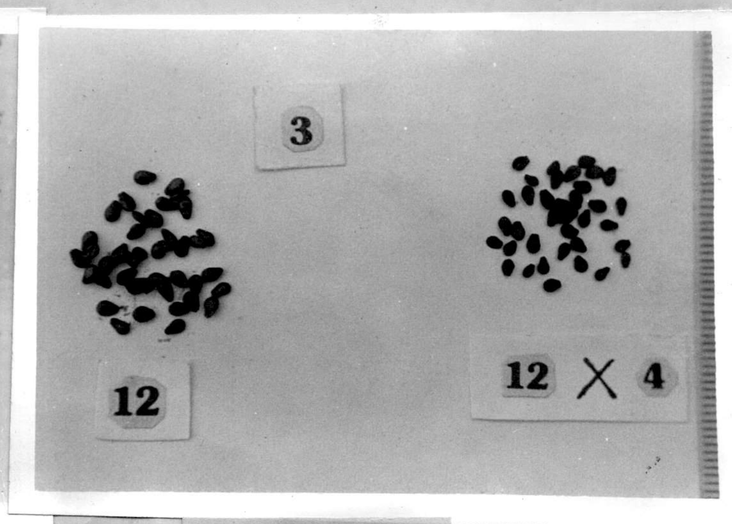


Figure 15f

presented in Figure 16a to d. There was substantial difference in length as well as breadth of dried seeds obtained from open pollination and cross pollination. Difference was minimum during the initial stages of deelopment and it increased gradually during the later stages of development. Percentage of reduction in breadth of the seed was more conspicuous than that in length.

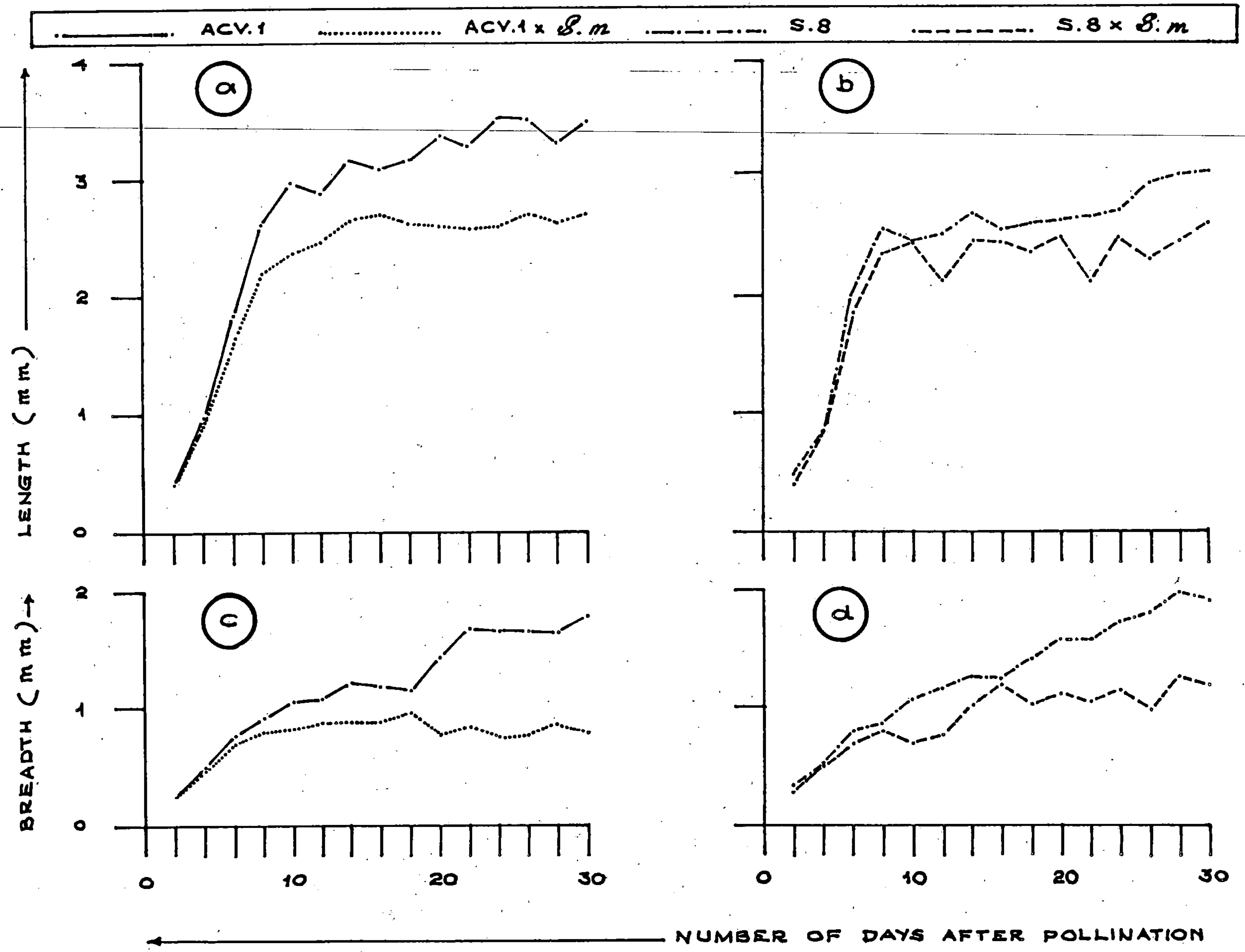
At two days after pollination, mean length of dry seeds was same in ACV.1 and ACV.1 x S. malabaricum (0.41 mm). In the case of S.8 x S. malabaricum there was 18 percent reduction. Breadth of dry seeds showed 4 and 14 percent reduction from their female parents in ACV.1 x S. malabaricum and S.8 x S. malabaricum respectively. Ten days after pollination, the percentage reduction in mean length of dry seeds was 20 in ACV.1 x S. malabaricum whereas it was negligible in S.8 x S. malabaricum. Mean breadth of dry seeds exhibited 23 percent reduction in ACV.1 x S. malabaricum and 33 percent in S.8 x S. malabaricum. Twenty days after pollination the percentages of reduction were 23 and 5 in the case of length and 45 and 28 in the case of breadth of dry seeds of the two crosses. Thirty days after pollination, length of dry seeds showed 23 and 15 percent reduction while breadth

Table 15. Size of dried seeds (mm) under natural and cross pollinations in *Sesamum indicum* varieties.

Sl. No.	Number of days after pollination.	ACV.1		ACV.1 x S.M*		S.8		S.8 x S.M	
		Length	Breadth	Length	Breadth	Length	Breadth	Length	Breadth
1	2	0.41	0.27	0.41	0.26	0.49	0.35	0.40	0.30
2	4	0.97	0.50	0.93	0.49	0.84	0.53	0.85	0.52
3	6	1.85	0.78	1.61	0.71	2.01	0.81	1.86	0.72
4	8	2.63	0.94	2.21	0.81	2.58	0.88	2.36	0.82
5	10	2.96	1.08	2.37	0.83	2.47	1.07	2.45	0.72
6	12	2.89	1.10	2.47	0.87	2.53	1.16	2.13	0.77
7	14	3.18	1.24	2.65	0.89	2.71	1.28	2.48	1.04
8	16	3.09	1.20	2.69	0.90	2.57	1.26	2.46	1.21
9	18	3.17	1.16	2.63	0.97	2.62	1.42	2.37	1.04
10	20	3.36	1.45	2.60	0.80	2.64	1.57	2.50	1.13
11	22	3.29	1.69	2.56	0.84	2.67	1.59	2.14	1.06
12	24	3.51	1.66	2.61	0.76	2.72	1.74	2.50	1.15
13	26	3.50	1.68	2.70	0.78	2.96	1.82	2.32	1.00
14	28	3.33	1.60	2.64	0.86	3.05	2.00	2.48	1.26
15	30	3.50	1.79	2.69	0.80	3.08	1.95	2.62	1.21

*S.M - *Sesamum malabaricum*

FIG. 16. SIZE OF DRIED SEEDS AT DIFFERENT STAGES OF DEVELOPMENT IN DIRECT CROSSES



of dry seeds showed 55 and 37 percent reduction in ACV.1 x S. malabaricum and S.8 x S. malabaricum respectively.

Size of dry seeds in reciprocal crosses are graphically presented in Figure 17 a and b. Dry seeds in direct and reciprocal crosses of S.8 x S. malabaricum at 10, 20 and 30 days after pollination are presented in Figure 18 a and b. In reciprocal crosses, Sesamum malabaricum x ACV.1 and S. malabaricum x S.8, the percentages of reduction in length and breadth of dry seeds showed a parallel change during the different developmental stages (Table 16). Two days after pollination the mean length and breadth were more for crossed seeds. After 10 days from pollination, the length of dry seeds in S. malabaricum open pollinated and S. malabaricum x ACV.1 cross showed no difference, while in S. malabaricum x S.8 cross, there was 11 percent reduction. Mean breadth of dry seeds was 5 percent more for S. malabaricum x ACV.1 and 9 percent less for S. malabaricum x S.8 than the wild open pollinated seeds. After 20 days from pollination, the length of dry seeds showed 17 percent reduction in S. malabaricum x ACV.1 and 16 percent reduction in S. malabaricum x S.8 from open pollinated seeds of the wild female parent.

Table 16. Size of dried seeds (mm) under natural and cross pollinations in *Sesamum malabaricum*.

Sl. No.	Number of days after pollination.	*S. malabaricum*		*S. malabaricum* x ACV.1		*S. malabaricum* x S.8	
		Length	Breadth	Length	Breadth	Length	Breadth
1	2	0.39	0.29	0.39	0.31	0.48	0.36
2	4	0.85	0.60	0.78	0.48	0.77	0.53
3	6	1.44	0.63	1.26	0.65	1.32	0.84
4	8	2.21	1.07	1.89	0.92	1.86	0.98
5	10	2.45	1.38	2.47	1.45	2.18	1.25
6	12	3.02	1.84	2.70	1.59	2.61	1.62
7	14	2.98	1.94	2.58	1.59	2.67	1.72
8	16	2.90	1.85	2.60	1.70	2.58	1.70
9	18	2.89	1.95	2.49	1.65	2.52	1.60
10	20	2.99	1.86	2.49	1.65	2.52	1.60
11	22	2.91	1.87	2.51	1.64	2.52	1.71
12	24	2.96	1.88	2.49	1.61	2.63	1.67
13	26	2.94	1.81	2.54	1.71	2.47	1.61
14	28	3.03	1.87	2.51	1.65	2.49	1.56
15	30	2.94	1.83	2.52	1.60	2.54	1.60

FIG. 17 SIZE OF DRIED SEEDS AT DIFFERENT STAGES OF DEVELOPMENT IN RECIPROCAL CROSSES

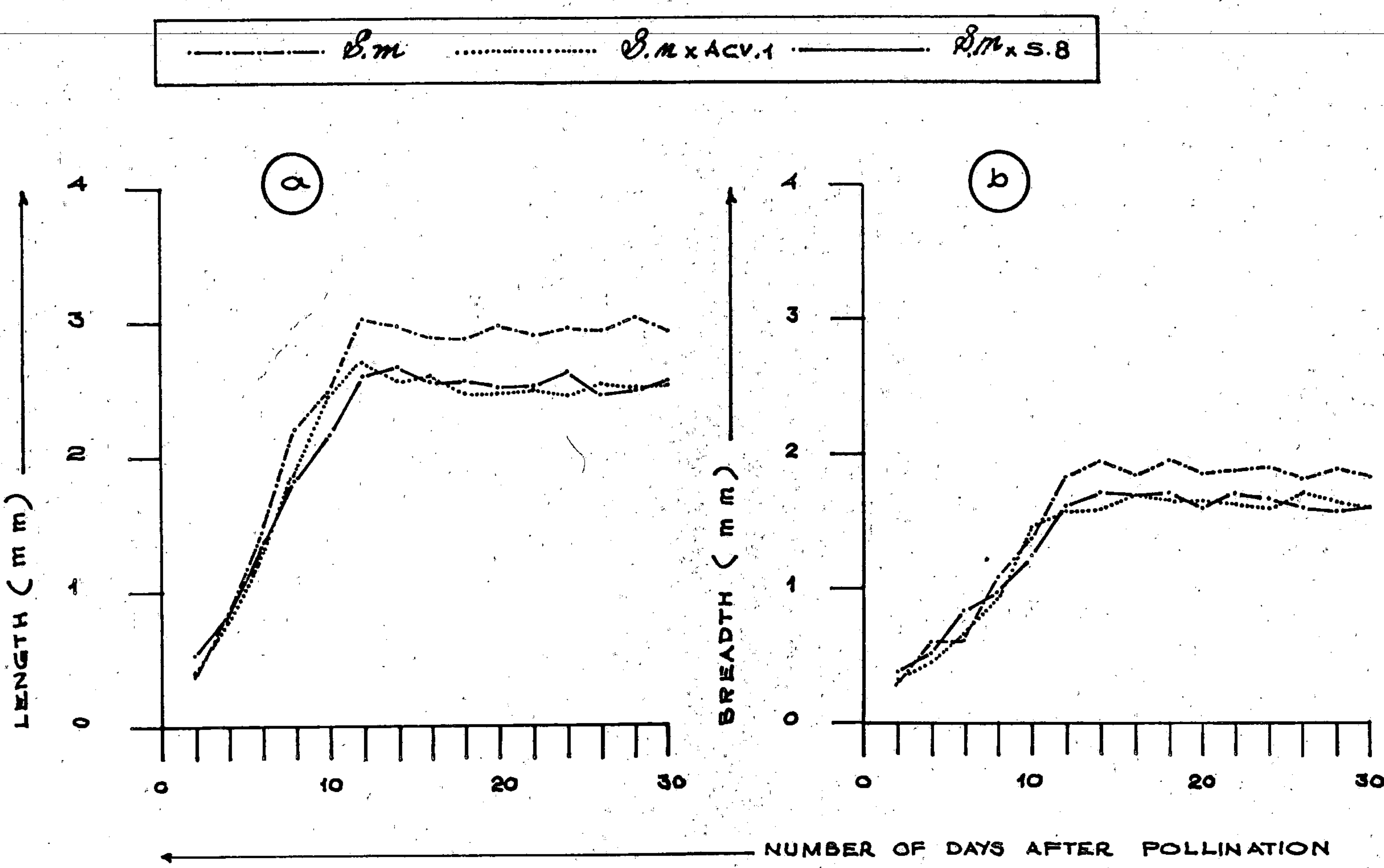


Figure 18. Dried seeds at different stages of development in S.8 x S. *malabaricum* crosses.

a. Direct cross

b. Reciprocal cross

1 - 10 days after pollination

2 - 20 days after pollination

3 - 30 days after pollination

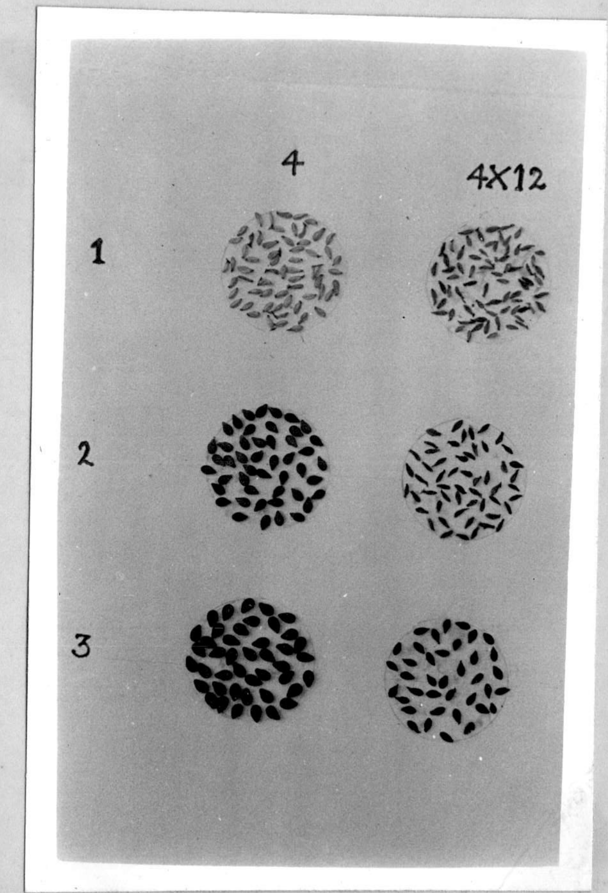


Figure 18a

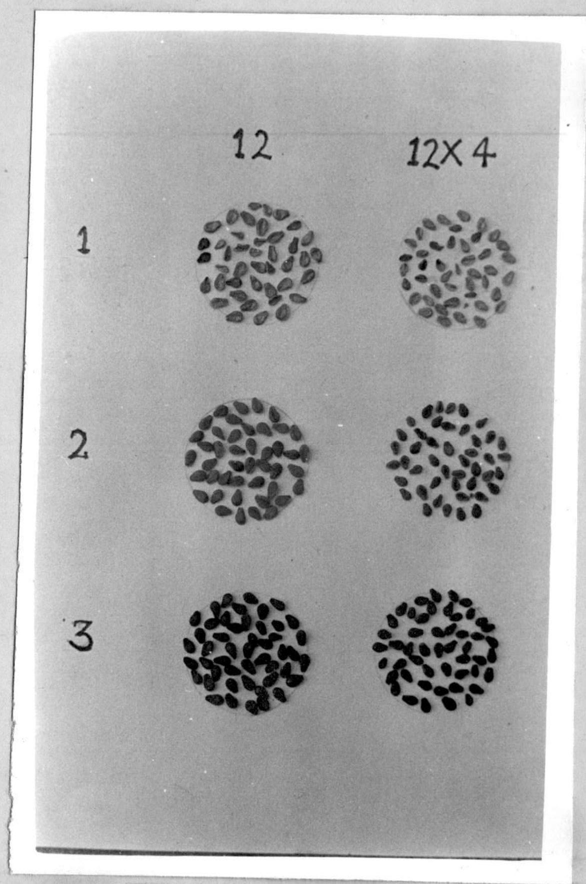


Figure 18b

Breadth of dry seeds at the same time exhibited 11 and 14 percent reduction in these two crosses respectively. At thirty days after pollination, the reduction in length was 14 percent and in breadth it was 13 percent in both the crosses.

DISCUSSION

DISCUSSION

Studies on interspecific relationships are useful in breeding programmes envisaging the transfer of desirable characters across the species barrier, usually from wild species to the cultivated ones. In *Sesamum*, eventhough about 34 species have been identified, only a few successful instances of interspecific hybridization have been reported. Most of the crosses attempted were unsuccessful due to interference of various reproductive isolation mechanisms, such as incompatibility, hybrid inviability and hybrid sterility. In a number of such crosses only shrivelled or empty nonviable seeds were obtained. Studies conducted on hybrid seed development indicate that hybrid inviability due to endosperm failure is the barrier to interspecific hybridization in some of these crosses (Dhawan, 1946; Anon., 1948; Dadlani, 1958). Results of studies on cross compatibility between *S. indicum* and *S. malabaricum* and the possible reasons for seed failure are discussed here.

Capsule setting was normal in direct as well as reciprocal crosses. In general, it was more in reciprocal crosses than in direct crosses. Out of the 22 crosses tried between eleven varieties of S. indicum on the one hand and S. malabaricum on the other (11 direct and 11 reciprocal crosses), two showed cent per cent capsule set (S. malabaricum x Cul.40 and S. malabaricum x N-62-32). Minimum capsule set (51%) was recorded in the cross, ACV.2 x S. malabaricum. Amirthadevarathinam (1965) reported 3.8% capsule set in S. indicum x S. capense cross and 3.9% capsule set in S. occidentale x S. indicum cross. Subramanian (1972) reported 30% capsule set in S. indicum x S. alatum cross and 8% in its reciprocal cross. He also reported 8% and 25% capsule set in direct and reciprocal crosses between S. indicum and S. radiatum and 6% capsule set in S. occidentale x S. indicum cross. These values are too low when compared to the results of the present study.

In spite of good capsule setting, normal seeds were not obtained in any of the 22 crosses. In direct crosses, medium shrivelled seeds were obtained in four and highly shrivelled seeds in the remaining seven. In reciprocal crosses, medium and highly shrivelled seeds were obtained in all the 11 crosses. Thus the reciprocal crosses showed higher percentage of medium shrivelled seeds than highly shrivelled seeds when compared to the direct crosses. The failure of normal seed set in crosses indicate that these two species have developed a strong isolation barrier which makes them completely noncompatible.

The direct crosses between Sesamum indicum and S. malabaricum and their reciprocal crosses appear to differ in the intensity of seed degeneration (Figure 5a and b). The comparatively better development of crossed seeds with S. malabaricum as female parent might be on account of the thicker seed coat of this wild species. Seed coat being a purely maternal tissue, the difference in seed coat thickness and development in the two species probably account for the minor differences in seed development in the two sets of crosses. Similarly, recovery of medium shrivelled seeds in four out of 11 direct crosses can also be attributed to the comparatively thicker seed coats of the female parents of these crosses viz. ACV.2, S.8, Vinayak and N-62-32. In all the crosses the colour of crossed seeds was the same as that of the selfed seeds of the female parent.

Recovery of shrivelled seeds in interspecific crosses in spite of normal capsule set has been reported in *Sesamum* by many earlier workers. Dhawan (1946) reported recovery of shrivelled seeds in *S. radiatum* x *S. indicum* and *S. radiatum* x *S. prostratum* crosses. *S. indicum* x *S. radiatum* cross also yielded similar results (Anon., 1948). Ramanathan (1950) obtained both shrivelled and normally developed nonviable seeds in the crosses, *S. radiatum* x *S. indicum*, *S. radiatum* x *S. prostratum*, *S. occidentale* x *S. indicum*, *S. occidentale* x *S. laciniatum*, *S. occidentale* x *S. prostratum* and *S. radiatum* x *S. laciniatum*. Mazzani (1952) reported that *S. indicum* x *S. radiatum* cross gave fruits without seeds. Kedharnath (1954) obtained some fruits containing shrivelled and nonviable seeds from *S. indicum* x *S. alatum*, *S. angolense* x *S. prostratum* and *S. angolense* x *S. laciniatum* crosses. Dadlani (1958) recorded nonviable seeds from the cross *S. occidentale* x *S. prostratum*. Amirthadevarathinam (1965) and Sundaram (1968) reported recovery of shrivelled seeds in *S. indicum* x *S. capense* cross. Subramanian (1972) obtained empty seeds in *S. indicum* x *S. alatum* cross and shrivelled seeds in its reciprocal cross. He also reported recovery of small and shrivelled seeds in the crosses, *S. indicum* x *S. radiatum*, *S. radiatum* x *S. indicum*, *S. occidentale* x *S. indicum* and *S. radiatum* x *S. laciniatum*.

In the present investigation, in all the crosses seed formation was inhibited at some stage after initial development which suggests that pollen grains although sufficiently successful to stimulate capsule formation, are inefficient to bring about normal seed development. This might be due to failure of either fertilization or growth and differentiation of the zygote into a normal embryo. Since no hybrid seed was obtained, the question of hybrid sterility or hybrid breakdown as an isolation mechanism does not arise here. Hence, the barrier between these two species could be either cross incompatibility leading to failure of fertilization or hybrid inviability resulting from failure of zygote development into the embryo.

Cross incompatibility.

The role of pollination in capsule development was critically studied in the style excision experiment. It was found that emasculated flowers did not develop into capsules if they were not pollinated. So the stimulus of pollination was essential for capsule development. Further, style excision done at different intervals after self pollination showed that when the styles were excised within four hours of pollination in *S. indicum* and within five hours of pollination in *S. malabaricum*, the flowers dropped off and capsules failed to develop. This indicated that for capsule development, styles should remain in tact for a minimum period of four hours in *S. indicum* and five hours in *S. malabaricum*. These results lead to the conclusion that some of the pollen tubes reach the ovary at four hours after pollination in *S. indicum* and at five hours after pollination in *S. malabaricum* and take part in fertilization. However, the percentage of capsule set remained low at this stage. For normal setting of capsules, the styles have to be retained for a minimum period of 5.5 hours in *S. indicum* and 6.5 hours in *S. malabaricum* (Table 6). If the pollen tubes traverse the stylar length during the interval between pollination and excision, style removal has no effect and normal capsule development is obtained.

Microscopic examination of pollen tube growth in the styles confirmed the above results. In selfed flowers of *S. indicum* and *S. malabaricum*, pollen tubes were found to enter the ovary 3.5 hours and 4.5 hours respectively after pollination. In the case of *S. indicum*, Joshi (1961) reported that in some ovaries pollen tubes were noticeable at four hours after pollination, though only a few apical ovules have received them. Six hours after pollination, pollen tubes were invariably observed in the ovary and they entered practically all the ovules. But Subramanian (1977) reported that pollen tubes were found just above the micropylar region four hours after pollination and pollen tubes have entered the ovule six to eight hours after pollination.

There is a difference of one hour in the time taken by the pollen tubes of *S. indicum* and *S. malabaricum* to reach the ovary. This difference could be attributed to differences between the two species, either in the rate of growth of pollen tubes or in the stylar length. Rate of growth of pollen tubes was found to be slightly different in selfed and crossed flowers. In selfed *S. indicum* flowers, the rate of growth was higher than that in flowers crossed with *S. malabaricum*. In the case of *S. malabaricum*, the rate of growth was slightly higher for the crossed flowers during the initial stages of growth. Dhawan (1946) reported that the rate of pollen tube growth was slightly less in *S. radiatum* x *S. orientale* cross than *S. radiatum* selfed, but fertilization was effected in all the ovules.

Dadlani (1958) reported that pollen germination, pollen tube growth and fertilization were normal and quicker in S. occidentale x S. prostratum than in S. occidentale selfed.

In S. indicum x S. mulayanum cross, pollen grains failed to germinate on the stigma of the recipient parent (Sastri and Sivanna, 1976). Non crossability due to restricted pollen tube growth in linseed crosses has been reported by Jindal and Kalia (1972). Abnormal pollen tubes were observed in the case of incompatible crosses in Brassica spp. by Sareen et al. (1970), Bochkarev (1973) and Khanna and Chowdhury (1981). Abnormalities were in the form of swelling at the tip of pollen tubes or its bursting in the stigmatic tissue. They showed anomalous growth and were unable to penetrate the tissue of the style. Incompatibility in those crosses was thus a pre-fertilization process. But in the present crosses between S. indicum and S. malabaricum apart from the slight variation in the rate of pollen tube growth in crossed and selfed flowers, the germination of pollen grains and growth of pollen tubes were uninterfered with. No abnormality was found in the growth and large number of pollen tubes were found to enter the ovary to fertilize all the ovules in direct as well as reciprocal crosses.

In all the 22 crosses, crossed capsules resembled the capsules resulting from selfing in the female parent. It shows that capsule development in this interspecific cross is not influenced by varietal difference. Studies conducted on selected crosses showed slight reduction in length and breadth of crossed capsules at all the stages of development. Reduction in the size of capsules could be due to a reduction in the number of seeds per capsule or due to a reduction in the size of seeds. Sharma and Khanna (1964) reported similar results in interspecific crosses in the genus Linum. Rate of growth of crossed ovaries was significantly lower than that of selfed ones and they concluded that slower development of the crossed ovaries might be due to any of the post fertilization causes, such as imperfect seed formation, slower growth of the embryo and poor development of the endosperm.

Mean number of seeds per capsule was substantially less in case of crossed capsules as compared to selfed capsules of the female parents (Tables 11 and 12). Percentages of reduction were more in direct crosses than the reciprocal crosses. Measurement of fresh seed length at different time intervals after pollination showed that eventhough there was some reduction at all the developmental stages, the length of seeds increased normally after pollination and maximum possible length was attained at about 10 to 12 days after pollination as in the case of selfed seeds.

A gross estimate of the frequency of fertilization may be made by determining the proportion of ovules which increase in size significantly after pollination (Cooper and Brink, 1945). Since pollen tubes have entered the ovary normally as in self pollinated flowers and the size of crossed ovules increased significantly during the subsequent stages it can be concluded that fertilization takes place in crossed flowers. However, the frequency of ovules getting fertilized is substantially less in crossed ovaries than in selfed ones. The difference in growth rate in the early stages of development of selfed and crossed seeds has been maintained as such throughout. These observations suggest that noncompatibility of these two species of Sesamum is not a pre-fertilization process.

Hybrid inviability.

The barrier which strikes at e different stages of development of seed after cross pollination has been catalogued as hybrid inviability. Estimation of dry seed size at different intervals of pollination revealed that size of open pollinated seeds increases gradually and reaches the maximum value at 24 to 28 days after pollination in S. indicum varieties (ACV.1 and S.8) and at about 12 days after pollination in S. malabaricum (Tables 15 and 16). But in the case of crossed seeds the rate of increase in size is very low. The hybrid seeds probably lag behind the normal ones in development from the initial stage of development itself. During the subsequent stages the lag increases so much that after 30 days of pollination the crossed seeds are left far behind the open pollinated seeds in size. In ACV.1 x S. malabaricum, mean breadth of the crossed seeds is only half of that of open pollinated seeds at 30 days after pollination. In S.8 x S. malabaricum there is 37 percent reduction. In the initial classification of crossed seeds, ACV.1 x S. malabaricum was included under the group of highly shrivelled seeds and S.8 x S. malabaricum under the group of medium shrivelled seeds.

Collapse of fertilized ovules during the early stages of post-fertilization development is frequent after interspecific crosses. This might be due to dis-hramonious interactions between the parental chromosomes or genes which are combined in the hybrid nuclei. In S. indicum x S. alatum cross, Kedharnath (1954) reported early abortion of young embryos as the reason for recovery of shrivelled seeds. Aneuploidy and markedly retarded embryo growth has been reported in the cross between Glycine max and G. tomentella (Sakai and Kaizuma, 1985). Disharmonious interactions between the parental chromosomes is most clearly evident in the first cleavage division of the zygote in the case of wide crosses and elimination of chromosomes occur during the cleavage mitosis and further development is arrested (Stebbins, 1958). But in hybrids between more closely related forms, no abnormality is evident in the early mitoses of the zygote, but they occur at later stages and often coincide with some critical or maximal period of differentiation of its tissues (Mc Cray, 1933).

In the present investigation, breakdown occurs at the initial stage of development of the crossed seeds. However, fresh crossed seeds in mature capsules do not show appreciable reduction in size from open pollinated seeds. So in this case, hybrid inviability due to disharmonious interactions between the parental chromosomes cannot possibly be considered as the reason for seed failure. Since there is no marked reciprocal difference in these crosses between S. indicum and S. malabaricum, the chance of noncompatibility involving cytoplasmic and plastid differences can also be ruled out.

Another possible reason for seed abortion is the malfunctioning of the hybrid endosperm associated with anomalous development of the surrounding maternal tissue. In S. indicum x S. radiatum (direct and reciprocal crosses), disintegration of the endosperm resulted in death of the zygote due to starvation (Dhawan, 1946; Anon., 1948; Mazzani, 1952). The same reason was reported by Dadlani (1958) for the seed failure in S. occidentale x S. prostratum cross. In the present study also, hybrid inviability due to endosperm failure can be considered as the possible reason for seed failure. Further studies on embryo and endosperm development would throw more light on the specific cause of crossed seed breakdown.

S. indicum x S. alatum and S. indicum x S. capense crosses showed seed failure eventhough they possessed the same chromosome number ie. 2n = 26 (Kedharnath, 1954; Amirthadevarathinam, 1965; Sundaram, 1968 and Subramanian, 1972). But in S. indicum x S. radiatum (direct and reciprocal crosses) and in S. occidentale x S. prostratum cross, the parents differed in chromosome number (Dhawan, 1946; Anon., 1948; Ramanathan, 1950; Mazzani, 1952; Dadlani, 1958; Subramanian, 1972). Embryo and endosperm failure takes place in one way by altering the chromosomal relations and in a different way by genome substitution (Cooper and Brink, 1945). Since the chromosome number of S. malabaricum is not clearly known, the genetic cause of hybrid inviability in the present study cannot be discussed further here.

Overcoming hybrid inviability by employing ovary/ovule/embryo culture techniques have been suggested by many authors. Mazzani (1952) obtained three plants by culturing the embryos of S. radiatum x S. indicum cross. But they resembled S. radiatum and were considered to be the result of diploid parthenogenesis. The stage at which ovule or embryo is to be collected for embryo culture can be decided only after conducting proper embryological studies in the crossed seeds. For ovary culture also, the technique is to be standardised by excising the ovaries at different intervals after cross pollination. Interspecific hybrids between S. indicum and S. malabaricum with desirable qualities can be obtained by perfecting any of these techniques of artificial culture.

The present study has thus clearly indicated noncompatibility of S. indicum with S. malabaricum in direct as well as reciprocal crosses. Normal growth of pollen tubes in the style, capsule development and substantial increase in size of ovules consequent to cross pollination indicate cross fertilization in this interspecific cross, eliminating the role of cross incompatibility as a species barrier. However, the inhibition of seed development and the absence of normal seed indicate the involvement of hybrid inviability due to endosperm failure as the possible reason for noncompatibility. As such embryo culture is suggested as an effective method for attaining interspecific compatibility between these two species of Sesamum.

SUMMARY

SUMMARY

The study of cross compatibility between *Sesamum indicum* and the wild species *S. malabaricum* was undertaken during 1985-'86. Eleven adapted varieties of *S. indicum* were crossed directly and reciprocally with *S. malabaricum*.

There was very good capsule set in all the direct as well as reciprocal crosses. In general, capsule set was better in reciprocal crosses than in direct crosses. Crossed capsules developed normally and they resembled the capsules of the female parents in all respects.

In spite of good capsule set these crosses failed to produce any normal seeds. In direct as well as reciprocal crosses, medium shrivelled and highly shrivelled seeds were obtained. Failure of normal seed development indicated that these two species are noncompatible. Slightly better development of seeds in crosses with *S. malabaricum* as female parent might be due to the comparatively thicker seed coat of this wild species.

The role of pollination in capsule development was studied by emasculating flowers and avoiding pollination. No capsule developed in this case indicating that the

stimulus of pollination is essential for capsule development.

Capsule development takes place only when styles were retained for a minimum period of four hours in S. indicum and five hours in S. malabaricum. This indicate that pollen tubes take a minimum period of four to five hours to reach the ovary. Microscopic examination of pollen tube growth in the styles further confirmed these results.

Pollen tube growth was slightly slower in crossed flowers of S. indicum than in the selfed flowers. In S. malabaricum on the other hand, pollen tube growth was slightly faster in crossed flowers than in selfed flowers. In spite of such slight differences in the rate of growth of pollen tubes in selfed and crossed flowers, large number of pollen tubes were found to enter the ovary to fertilize the ovules.

There was slight reduction in length and breadth of capsules in direct and reciprocal crosses between S. indicum and S. malabaricum at the different stages of development when compared to the open pollinated capsules on female parents. The reduction in size of crossed capsules can be due to the reduction in the number of seeds per capsule as well as the smaller size of crossed seeds.

In crossed as well as open pollinated capsules, the length of fresh seeds increased gradually and attained a maximum at 10 to 12 days after pollination. Crossed seeds showed reduction in length from open pollinated seeds of the female parents at all the stages of growth.

Since pollen tubes in crossed flowers reach the ovary normally and the size of crossed seeds increase gradually after pollination it can be inferred that seed failure in this interspecific cross is not a pre-fertilization process ie. cross incompatibility is not the barrier in this cross.

Open pollinated seeds attained the maximum size at about 24 to 28 days after pollination in *S. indicum*. Crossed seeds lagged behind at the early stages of development itself. In reciprocal crosses there was a similar reduction in size of crossed seeds from open pollinated seeds of *S. malabaricum*. These observations show that development of crossed seed is inhibited at an early stage of growth. Hence hybrid inviability is the possible reason for breakdown of this interspecific cross. Apart from slight reduction in size, fresh seeds looked normal even in the mature crossed capsules. So endosperm failure can be considered as the cause of hybrid inviability.

Hybrid inviability can be overcome through embryo culture and hybrids with desirable qualities can be obtained. The stage at which ovule or embryo has to be collected can be decided by conducting embryological studies in the crossed ovules.

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*Original not seen

CROSS COMPATIBILITY BETWEEN *Sesamum indicum* L. AND *S. malabaricum*

BY

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ABSTRACT OF A THESIS
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ABSTRACT

Sesame is most valued annual oilseed crop of Kerala. Lack of varieties with resistance to drought and diseases is the main problem confronting sesame cultivation in the state. The wild species *Sesamum malabaricum* shows tolerance to drought and most of the diseases. But this wild species appears to be non-compatible with the cultivated *S. indicum*. An investigation was therefore undertaken to study cross compatibility between these two species.

Eleven adapted varieties of *S. indicum* were crossed reciprocally with *S. malabaricum*. Capsule set was normal in direct as well as reciprocal crosses. In general it was more in reciprocal crosses than in direct crosses. In spite of normal capsule set, these crosses failed to produce any normal seed. The seeds were either medium shrivelled or highly shrivelled. Failure of normal seed development in a wide range of crosses confirm that these two species are noncompatible.

Emasculated flowers did not develop into capsules in the absence of pollination. So the stimulus of pollination is essential for capsule development. Style

excision done at different intervals after self pollination indicated that pollen tubes take a minimum period of four hours to reach the ovary in S. *indicum* and five hours in S. *malabaricum*. Pollen tube growth was of course slightly slower in crossed flowers of S. *indicum* than in the selfed flowers. But in S. *malabaricum*, pollen tube growth was slightly faster in crossed flowers than in selfed flowers. Microscopic examination revealed normal growth of pollen tubes in the styles of these two species. Pollen germination and pollen tube growth were normal in crossed flowers also and large number of pollen tubes were found to enter the ovary for fertilizing the ovules.

Crossed capsules showed slight reduction in size than open pollinated ones on the female parent at all the stages of growth. Mean number of seeds per capsule showed substantial reduction in the crosses. Crossed seeds also showed reduction in length from open pollinated seeds of the female parent. Since the pollen tubes show normal growth and reaches the ovary, and that the crossed ovules show substantial increase in size, it can be inferred that seed failure in these crosses is not a pre-fertilization process.

Measurement of dry seed size at different intervals after pollination indicated that the crossed seeds lag behind the open pollinated seeds of the female parent from the initial stages onwards. These observations show that development of crossed seed was inhibited during the early stage of development itself. Hence hybrid inviability could be the reason for the recovery of shrivelled seeds in these crosses. Endosperm failure can be considered as the possible cause of this inviability.

Ovary/ovule/embryo culture, can be adopted to overcome this barrier and to get hybrids with desirable qualities of S. indicum and S. malabaricum. Attempts in this direction will enable a breaking down of noncompatibility in this interspecific cross and thereby widen the scope for stress resistance breeding in sesame.