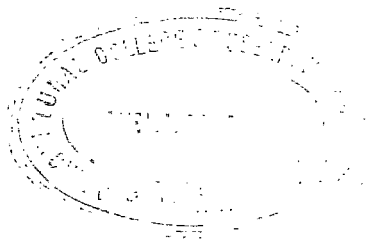


STUDIES ON THE C₂ GENERATION OF THE COLCHICINE INDUCED POLYPLOIDS OF CHILLIES

(*Capsicum frutescens* L.)



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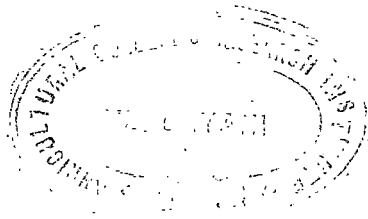
M. LATHIKA

THESIS

SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR
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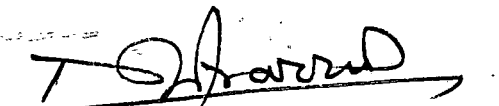
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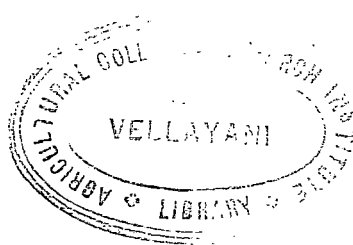
CERTIFICATE

This is to certify that the thesis herewith submitted contains the results of bonafide research work carried out by Smt. M. Lathika, under my supervision. No part of the work embodied in this thesis has been submitted earlier for the award of any degree.


(P. KUMARA PILLAI)
Principal.


(M. ARAVINDAKSHAN)
Lecturer in Horticulture.

Agricultural College &
Research Institute,
Vellayani, Trivandrum,
August, 1968.

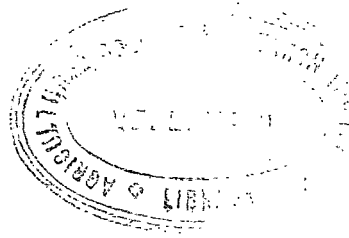


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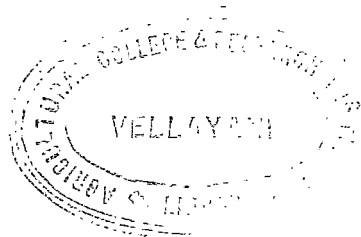
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INTRODUCTION



INTRODUCTION

Chillies (Capsicum frutescens L.) is widely used in India for culinary purposes. Induction of polyploidy has been suggested as a means of improvement of chillies by earlier workers (Pal et al, 1939; 1941; Aleksic, 1960). The scope of polyploidy breeding in producing new and superior plant types is well recognized. Substantial evidences are there in literature to support the fact that colchicine induced polyploids of several crop plants have established themselves as promising varieties.

Investigations on the induction of polyploidy in chillies by the application of colchicine was started in the year 1966 in the Agricultural College and Research Institute, Vellayani. Indiramma (1967) treated the seeds and seedlings of chillies with varying concentrations of colchicine for different durations in her attempts to produce tetraploid types. One tetraploid plant was successfully obtained by treating the seedlings with 45 per cent colchicine for 9 hours. In her studies Indiramma recorded the extent of morphological variations that occurred among the colchicine treated plants in addition to the characters associated with the tetraploid plant.

The study reported in this thesis is a continuation of the previous work, and embodies the details of investigations carried out on the C_2 generation. Seeds obtained from the colchicine induced tetraploid chilli were made use of in the present investigation. The objective of the work was to find out the extent of morphological variations among the C_2 progeny of the polyploid in comparison to that of the diploids. The studies were also directed to ascertain the plant characteristics of the polyploid types and abnormalities exhibited by them in the C_2 generation. Cytological studies were also made in the plants suspected to be polyploids as a confirmatory measure.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

The discovery, made independantly by Eigsti, Blakeslee and Avery (1937) and Nebel and Ruttle (1937), that polyploid strains of plants can be produced in large numbers through the action of the drug colchicine paved way for intensive studies on the artificial production of polyploids. Several methods of colchicine application have been adopted for the induction of polyploidy in plants. The principle underlying the colchicine treatment is that the alkaloid must be brought in contact with the actively dividing cells. This is effected either by treating pre-soaked seeds with colchicine solution or by applying colchicine to the growing tips of the seedlings. An attempt is made here to review the work done on the methods of application of colchicine in a wide variety of economic plants

Seed treatment

Treatment of seeds with colchicine at different concentrations for different durations has been found to be effective in inducing chromosome doubling.

Pal, Ramanujam and Joshi (1941) used concentrations of 0.05, 0.1, 0.2 and 0.4 per cent aqueous colchicine solution for duration ranging from 1 to 8 days and Aleksic (1960) 0.8 per cent colchicine solution for inducing polyploidy in chillies. These workers treated the seeds in their experiments.

Richaria^h and Persai (1940) and Srivastava (1956) produced autotetraploids of sesamum by treating the seeds with colchicine of various concentrations for varying durations.

Ramanujam and Joshi (1941) subjected germinating seeds of gram to varying doses of colchicine solutions. Treatment of seeds with 0.25 per cent aqueous solution for half an hour gave the best result from the point of view of survival of seedlings and induction of polyploidy.

Srivastava (1955) found in Cicer arietinum that seed treatment with 0.25 per cent colchicine for 30 minutes was superior to seedling treatment in inducing tetraploidy.

Reo et al (1944) produced tetraploid strains of Corchorus olitorius by treating dry seeds with colchicine solutions of 0.05 to 1.0 per cent concentrations for 12 to 24 hours.

Ramanujam and Deshmukh (1945) induced autotetraploidy in several self sterile species of Brassica by treating dry seeds in aqueous colchicine of 0.1 to 0.4 per cent strength for 24 to 48 hours and germinated seeds in 0.025 to 0.1 per cent for 8 to 24 hours. Warnke (1945) could induce polyploidy in Taraxacum kok - saghyz by immersing seeds in 0.05 to 0.8 per cent colchicine for 1, 2 and 4 days in covered dishes at room temperature. Hertzsch (1954) obtained tetraploid forms of Oryza sativa by immersing germinated seeds in 0.05 per cent colchicine. Sen and Hari (1955) induced polyploidy in Vigna sinensis by seed treatment.

Seedling treatment

Treatment of the apical buds of young seedlings with colchicine solution is yet another method for the induction of polyploidy. Evans (1955) working on Trifolium pratense compared the effectiveness of different techniques commonly used and found that seedling treatment gave the best results.


Toole and Bamford (1945) successfully doubled 8 haploid peppers to their diploid forms, by smearing 0.1 to 1.0 per cent colchicine emulsion on the growing tips, once or twice a week.

Shimamura (1939) induced tetraploidy in tomato by seedling treatment. Beasley (1940) produced polyploids in Gossypium by colchicine treatment of seedlings. Choudhary et al (1956) produced tetraploids in two varieties of Corchorus capsularis and three varieties of C. olitorius by treating seedlings with 0.1 to 0.2 per cent colchicine for 24 hours. Sen and Chedha (1958) treated the seedlings of Phaseolus mungo with 0.25 to 0.5 per cent colchicine for 6 to 9 hours for one or two days and produced 50 per cent polyploids.

Sen and Bhowel (1960) found that seedling treatment was much more effective than that of seeds in inducing tetraploidy in vigna sinensis. Bouharmont (1961) found that in rice treatment with 0.1 per cent colchicine solution for 2 hours could induce polyploidy. It was observed that 20 days old plants were better than newly germinated seedlings to induce polyploidy. Ramen and Kesavan (1963) applied 0.5 per cent colchicine in water for 90 minutes each day for 3 consecutive days on the apical buds of the seedlings of Arachis duranensis and doubled its chromosome number.

Other methods

Apart from seed and seedling treatment, colchicine had been applied to various other parts of plants and has



been reported to be equally effective in inducing chromosome doubling. Methods adopted by various workers are given below.

Pal et al (1938) induced polyploidy in chillies by treating roots of seedlings with 0.05 to 0.4 per cent colchicine for 4 to 6 hours at the time of transplanting.

Pierse Gavaudan and associates (1937) gave the first account of polyploidy induced by colchicine in Allium roots.

Nebel and Ruttie (1938) treated cuttings of Tradescantia reflexa with 0.2 per cent colchicine for 4 hours and induced tetraploidy. Langham (1940) treated the axillary buds of sesame with 0.5 per cent and 0.4 percent colchicine and new buds developed were tetraploid branches. Shifriess (1942) tried 0.3 to 0.5 per cent colchicine emulsion on the short apex of Cucumis sativus at the cotyledonary stage and this was found to be an efficient method for the mass production of polyploids.

Kumar et al (1957) in order to overcome the sterility of the triploid hybrid obtained from a cross between a tetraploid and diploid Arachis hypogae applied 0.2 per cent aqueous colchicine to the buds of the

sterile hybrids and produced a fertile allohexaploid plant.

Sobti (1963) produced tetraploid strains of Mentha piperata by treating the suckers of diploid plants with 0.1 per cent colchicine for 24 hours.

Morphological variations

Chromosomal abnormalities and morphological variations in the plant characters are associated with polyploidy. The usual symptom noticed is the gigas nature of plants. However, decreased growth rate, later flowering etc. have also been noted. Morphological variations in plants subjected to treatment with colchicine have been reported by several workers. The literature on the morphological variations observed in different crop plants are reviewed below.

According to Kumar and Abraham (1942) tetraploids of Phaseolus in general showed a decreased growth rate and consequently took longer time to reach maturity and flowering than the diploids. However, autotetraploids were larger, in many respects, than their related diploids as a result of increased cell size. In general, There was an increase in size of various plant parts, a

delay in growth and in flowering and often an increase in the darkness of the foliage.

According to Stebbins (1950) increase in chromosome numbers often resulted in abnormalities such as dwarfing, wrinkled foliage and weak plants. In most of the tetraploid plants growth is slower at first but later on it grows rapidly than the diploids (Janaki Ammal and Bezbaruch, 1962).

Pal, Ramanujan and Joshi (1941) induced polyploidy in chillies by treating the seeds with colchicine. They obtained 54 tetraploids, 4 periclinialploid chimeras (with $4n$ epidermis and $2n$ pollen) and 186 diploids from a population of 244. The tetraploids were recognised on the basis of their thicker stem, bigger and thicker leaves, larger stomata, bigger pollen grains with 30 to 90 per cent pollen sterility. In the G_2 generation, some progenies of the tetraploids were found to consist of only tetraploids but others of diploids and tetraploids showing that the parent plant of the latter was really mixoploid. The periclinialploid chimera gave rise to normal diploid progenies as was expected. In the G_3 generation the tetraploids again gave rise to tetraploids. The seed setting was higher in the G_3 generation.

Georgieva (1961) found that the autotetraploids of *Capsicum* are characterised by increase in size of the cell, leaf, stem, flower, stomata, pollen and seed. The viscosity of the cytoplasm, the size of the fruits and the number of seeds were reduced. Growth was rapid but the plant matured slowly; flowering and fruiting occurring much later than in normal plants. The autotetraploids were unstable and productivity decreased in subsequent generations. Aleksic (1961) found that in tetraploid *Capsicum annuum* the fruits were smaller and lighter but with higher dry matter content.

Ghodgaonkar, Deshpande and Beohar (1965; 1966; 1967) collected two samples of C_1 tetraploid seeds in chillies. They showed 16.9 per cent and 27.9 per cent increase in weight over normal diploid plant. The heavier seed produced tetraploid in C_2 generation. The C_2 plants were gigantic having increased height, broader leaves, flowers, stomata and pollen grain size. The seeds were larger and heavier. The $4n$ number of chromosomes was cytologically confirmed.

Srivastava (1956) observed in *Sesame orientale* L. that the colchicine treated plants showed an early retarded growth followed by a considerably enhanced growth rate. Richharia and Persal (1940) found that in

tetraploids of *Sesamum* the plants possessed smaller hypocotyl, thick cotyledon and short stunted root.

These plants were characterised by shorter, stiffer and thicker stem and dark green, broad and thick leaves.

The authors confirmed that such plants were polyploids by observing the chromosome number.

Langham (1942) found that in *Sesamum indicum* L. the tetraploid branches possessed large mucilage glands and larger but fewer number of stomata. The pollen mother cells of such branches showed the tetraploid chromosome number of $2n = 52$ while others had chromosomes between 26 and 52. Some of the tetraploid branches were fertile. The number of pods and number of seeds per pod did not show any difference from the diploids. But the seed weight was more in tetraploids. Kobayashi and Shimamura (1945) observed that the polyploids of *sesamum* possessed thicker stem, larger leaves and bigger stomata, flowers and seeds compared to the diploids and the same was stable even seven years after the treatment with colchicine. Shimamura and Kobayashi (1948) made the following observations in the colchicine induced tetraploids of sesame. The tetraploids bore shorter and fewer number of capsules, but the seeds were larger. The fertility in tetraploids were poor. However, cultivation

under stable conditions showed that the fertility of the tetraploids can be improved.

Parthasarathy and Kedarnath (1945) produced auto-tetraploids in sesamum and observed that tetraploids possessed gigas characteristics. The tetraploids surpassed their diploid progenitors in features like seed weight, number of branches and bigger size, flowers and pollen grains. But they were far behind the diploids in fertility. In tetraploids the capsules were lesser in number and they were smaller in size.

Srivastava (1956) studied the breeding behaviour of induced polyploids of Sesamum orientale L. The tetraploids of C_1 generation were characterised by the general features associated with polyploids such as delayed growth and flowering, larger but darker foliage, larger pollen grains and stomata and reduced fertility. In the C_2 generation 176 tetraploids and five mixoploids were obtained from 181 seeds raised from the tetraploid plant. Seeds from some treatment failed to germinate. The C_2 plants also were characterised by the gigas nature.

Kovacs-schneider (1959) reported that colchicine induced tetraploid tomatoes showed an early vegetative development. But flowering and fruit setting showed no difference from that of the diploids. They possessed smaller fruits with fewer seeds. Kuzdowicz (1964) observed that in the genus Lycopersicon, the autotetraploids were less fertile than the diploids. Baldy (1967) found that selection in $4x \times 4x$ hybrids of tomatoes led to improvements in earliness, fruit set and fruit size. Three lines reached or approached the diploids in earliness and surpassed them in dry matter, sugar and ascorbic acid content and were better in colour.

Smith (1939) found that the autotetraploids of Nicotiana rustica, N. tabacum and N. glauca possessed smaller plant and leaf size. But the leaves were thicker than their corresponding diploids. On the other hand, Warnke and Blakeslee (1939) observed larger stigma, thicker and longer anthers, thicker corolla and flower stalks in the tetraploids of Nicotiana.

Kundu and Sharma (1956) induced tetraploidy in Corchorus olitorius L. and found that the tetraploids were only slightly less vigorous than the corresponding diploids. At times the tetraploid jute showed higher

pollen fertility than the diploids and the meiotic abnormalities were comparatively fewer.

Pissarev (1955) reported that when the wheat x rye hybrids were treated with colchicine they gave high yield in grain and straw and showed higher protein content.

Terkowski (1965) found that in rye the diploid varieties had a greater number of fertile flowers and that they set seeds more easily than the tetraploid varieties.

Fraudson (1959) observed that tetraploids of red clover surpassed the diploids in persistence, yield of dry matter and crude protein but rather low in fertility. Armstrong and Robertson (1960) found that tetraploid alsike clovers surpassed the diploids in characters like pollen diameter, stoma size, number of plastids in guard cells, height of the plant, leaf area, seed size, hay yield etc.

Mehta, Subramanyam and Swaminathan (1963) studied the effect of colchicine on four strains of berseem (Trifolium alexandrinum - $2n = 16$). All the features of autotetraploids such as larger size of stomata, cells, pollen and plant parts and reduced pollen and seed fertility were found in tetraploid berseem. The

tetraploids showed increased vegetative vigour and ability to produce larger number of tillers than the parent diploids.

Odincova (1965) studied the pollen structure in red alsike and white clovers and in Trifolium resupinatum and found that the tetraploid forms had 10 to 98 per cent of the pollen irregular in form.

Mehta et al (1966) induced polyploidy in Trifolium foenum graecum. The C_1 plants showed the general characters associated with polyploidy. In the C_2 generation the plants showed difference in their response to polyploidy. However, there was an increase in forage yield and essential oil content. The seed set was generally low.

Luongdinhoua (1950) produced polyploid rice seedlings by colchicine treatment. He distinguished the induced tetraploids from the original diploids by the awn development, increased length of the ligule of the flag leaf and the increased size of the spikelet and pollen grains. Most of the pollen grains of the tetraploids showed 2 to 3 germ pores in contrast to one noticed in the diploids. Pollen sterility was more evident in tetraploids, as a result of which the spikelet fertility

was reduced to 40 per cent in the tetraploids.

Gopalakrishnan and Shastri (1964) distinguished the tetraploids of Oryza australiensis on the basis of their dark green foliage, stouter foliar veins, larger spikelets and elongated tip of palea.

Libedeva (1959) produced less vigorous and self sterile polyploid potatoes by colchicine application. He observed a linkage relationship between increased yield and decreased starch content.

Sando (1939) observed in colchicine treated Fagopyron tataricum that the surface of the hull was rough with more prominent lateral ridges than those in diploids.

Ramuson and Levan (1939) treated sugarbeet with colchicine and found that after seed treatment, the first year plants exhibited polyploid characters, but later showing a tendency to revert back to diploidy. Treatment of shooting flower buds produced branches or flowers with both tetraploid and diploid sectors. Pollen fertility was not reduced although 0 to 4 quadri-valents were observed. Of the 84 seedlings grown from the tetraploid branch, three were tetraploids, 13 triploids and 68 diploids. The tetraploids were as fertile

as the diploids. Even after removing all the diploid branches the majority of seeds produced by the tetraploid branches were normal diploids.

Amin (1940) reported that autotetraploids of cotton showed retardation of initial growth and abnormalities like swollen hypocotyledonary stem, prominent leaf veins, broader leaf lobes and bracts, bigger flowers and seeds and larger pollen.

Ramanujam and Joshi (1941) subjected the germinating seeds of Cicer arietinum to varying concentrations of colchicine. From the point of view of the survival of the seedlings and induction of polyploidy, treatment of seeds with 0.25 per cent aqueous solution of colchicine for half an hour gave the best results. A delayed germination and retardation of growth rate were reported in treated plants. G_1 generation showed a mixture of $4n$ and $2n$ tissues. But later generations revealed pure tetraploids. Besides the abnormalities like curling and twisting of stem and leaves and roughening of their surface, the tetraploid possessed large number of leaves per plant, bigger leaflets flowers, pods and seeds than the diploids. The pollen grains and stoma guard cells of the tetraploids were also bigger than those of the diploids. The pollen grains

showed 40 to 80 per cent sterility in tetraploids in comparison to 0 to 10 per cent sterility in diploids. With respect to plant height, the difference noticed between tetraploids and diploids was not significant. However, tetraploid plants possessed more number of branches.

Graner (1941) observed larger leaf lobes with a sudden constriction at the tip in the tetraploid of Manihot utilissima while in normal plants the leaves were narrower.

Shifriss (1942) noted in the tetraploid Cucumis sativus an accentuated serration of leaf margin.

Randolph et al (1932) reported that the autotetraploids of maize possessed thicker leaves and showed delayed flowering due to prolonged vegetative growth.

Reo et al (1944, 1945) distinguished the tetraploid jute from the diploid by their gigas character and from their structural differences in the chromosomes.

Warmke (1945) obtained tetraploid races of Taraxacum kok-saghyz by colchicine treatment. Nineteen tetraploids grown in green house showed significantly larger roots but a lower rubber percentage than a similar

number of green house grown diploids. Forty three field grown tetraploids had both significantly larger roots and higher rubber percentage than a group of thirty six comparable diploids.

Napp-zinn (1949) grew descendants of some *Oenothera* hybrids and separate the early germinating individuals from those which germinated much later. In the descendants of *O. flavens* x *franciscana* hybrids, triploid plants appeared almost exclusively among those germinating late.

Nishiyama (1950) observed a retarded growth rate in the colchicine induced polyploids of radish. The yield of the tetraploids were higher than that of the diploids.

Tandon and Chinoy (1950) suggested the possibility of producing superior types in *Amaranthus bilitum* through colchicine technique. They observed in the treated plants thicker stem, more number of leaves and branches, larger and dark green leaves and also prolonged vegetative growth.

Tandon (1951) reported that there was apparently no branching in the diploids of *Brassica oleraceae* var. *Botrytis*. But he could produce profusely branching individuals by colchicine treatment.

Derman (1954) collected better sized berries from the tetraploid grapes obtained by colchicine application.

Singh (1955) while studying colchicine induced tetraploids in Carica papaya found the tetraploids to be of no economic importance since they were highly sterile and produced round fruits which were much reduced in size.

Rajan and Ahuja (1956) studied the high and low fertility of diploids and induced autotetraploids in toria. He found that the lack of fertilisation of normal embryo sacs, prefertilisation abnormalities and abnormal development of embryo and endosperm were the chief causes for sterility.

Funke (1956) described a rapid method for the selection of polyploid plants. This method consisted of immersing mature pollen grains in a mixture of one part of concentrated sulphuric acid to three parts of 2 to 3 per cent methyl blue acetic acid, when the pollen pores were distinguishable as small indentations on the surface of the pollen grains. By this means the number of pollen pores which was found to be three in the case of diploids and to vary from 3 to 6 in the corresponding tetraploids may be readily ascertained under the microscope. This method was found to be very effective in selecting out tetraploids of

Antirrhinum majus, Polygonum, Convolvulus, Lycopersicum esculentum etc.

Spasojevic (1956) observed late flowering and ripening but larger seeds in the autotetraploids of Phaseolus vulgaris L.

Bhattacharjee (1956) found that tetraploids of Cajanus cajan obtained by colchicine treatment had significantly lower number of branches and nodes than the diploids. The number of leaves, size of leaves and leaflets did not show much difference between tetraploid and diploid.

Saito (1957) reported that colchicine induced tetraploid strains of American watermelon were very late in maturing. Chopra and Swaminathan (1960) observed that seed fertility in the autotetraploids of watermelon was considerably reduced.

Sharma and Dutta (1957) found larger and darker plants in the polyploids of Coriandrum sativum. None of the seeds from this autotetraploid were germinated.

Zimmerman (1958) found that in forage grasses colchicine induced tetraploids were less vigorous and winter hardy than diploids.

Sen and Cheda (1958) obtained complete polyploids, branch polyploids or sectorial polyploids in black gram by colchicine technique. The polyploids had bigger flowers, seeds, pollen and stomata but shorter pods and heavier seeds. C_1 and C_2 plants showed less vigour and late flowering.

Zebrek (1959) made hybrids of Triticum monococcum and T. timopheevi which were sterile. But fertile hybrids were obtained from crossing T. durum x T. timopheevi and T. polonicum x T. timopheevi. Doubling the chromosome number of these hybrids with colchicine was found to decrease fertility by 15 times. The autopolyploids of T. durum ($2n = 56$) and T. timopheevi ($2n = 56$) had low fertility. There was only a set of 4 to 5 grains per ear. Crossing these autopolyploids resulted in increased fertility. As many as 30 grains were produced per ear in such hybrids.

Kluge (1959) induced tetraploid in Fragaria vesca var. Semper florens and compared with the diploids. He found that the tetraploids formed fewer inflorescence per plant and accordingly fewer but larger flowers with the same number of petals and fewer anthers when compared to the diploids. Pollen grains in tetraploids were fewer in number and their viability was too low to facilitate

uniform and adequate pollination. Seed weight of tetraploid was double that of diploids but showed only 20 to 30 per cent viability. The number of seeds per fruit amounted to about 100 in diploids against 30 to 40 in tetraploids. The fruits of tetraploids were often deformed and were smaller than those of the diploids.

Mackevic (1959) observed a retardation of growth in the colchicine induced tetraploids of Populus tremula L. and P. balsamifera.

Moffett and Nixon (1960) observed that tetraploids of Acacia were less vigorous than the diploids. The bark thickness and tannin content were greater, but yield per acre was lowered.

Thombre and Desai (1960) found broader and thicker leaves in the tetraploids of Agave centela.

Janaki Ammal (1962) observed that the tetraploid Rauwolfia serpentina B. showed an increase in alkaloid content and larger size of flower and vegetative part.

Vakili (1962) induced tetraploidy in Musa balbisiana which possessed thicker leaves with shorter petioles than the diploids.



Sankaran (1964) studied the morphological characters of the autotetraploid Sorghum vulgare and its hybrid with Sorghum halepense. The tetraploid resembled its fertile counter part and exhibited increased size of grain.

Vieweshwara and Chinnappa (1965) induced autotetraploidy in Coffea canephora. Gigas characters were noticed in the polyploids. Flowering was profuse but fruit set was low.

Mettin (1965) induced tetraploids by colchicine in Aegilops speltoides. The C_2 generation showed normal growth in the seedling stage. Later the culm thickened and tillering was reduced; ear emergence was delayed and height decreased. The ears became shorter and the number of spikelets and grains was reduced. The length of spikelets and awns was also reduced. The number of florets per spikelet was unchanged and the seeds were larger.

Tandon and Rao (1966) found that the hybrids of a $4x \times 2x$ Solanum nigrum complex were taller than either of the parents and had a well developed branching system. Leaves were large, thick, dark green, ovate with dentate margins. They resembled the tetraploid parent in foliar

characters. Although the hybrid plant flowered profusely no fruit was formed. Pollen fertility was as low as 0.25 per cent. Meiotic studies in the pollen mother cells of hybrids confirmed that they were triploids.

Laws (1967) induced polyploids in *Oenothera*. He observed that most of the tetraploid races showed a marked increase in pollen sterility and a decrease in the seed set per capsule. There was no evidence to show that tetraploid plants produced a greater amount of nonviable seeds than the diploids. Germination in diploids and tetraploids was comparable and seedling mortality was negligible.

Cytological observations

The action of colchicine on mitosis has been found to be greatly efficient, highly specific and totally selective. The reaction of colchicine depends upon its concentration and time of exposure. The observations by Nebel (1937), Nebel and Rattle (1938), Levan (1938) and many others subsequently showed that colchicine inhibited the formation of the spindle in a dividing nucleus without affecting the division of chromosomes. Due to this failure of spindle formation the daughter chromosomes do not move to opposite poles and the

chromosomes accumulate in pairs ("colchicine-pairs"). Cytokinesis does not take place and the original cell has a reconstituted nucleus with double number of chromosomes.

Meiotic abnormalities like multivalent formation, chromatin bridges, lagging chromosomes etc. associated with colchicine treatment have been reported by many workers. Pal, Ramanujam and Joshi (1941) found 0 to 7 quadrivalents and the rest bivalents during meiosis at diakinesis and metaphase I in the colchicine induced polyploids of Capsicum annuum. They also pointed out that apart from inducing polyploidy colchicine effected in inducing numerical and structural changes in the chromosome complement.

Richharia and Persai (1940) reported 0 to 5 quadrivalents in the autotetraploids of Sesame. Ramanujam and Joshi (1942, 1944) found that in the A_2 generation of Sesamum orientale ($2n = 26$) x S. prostratum ($2n = 32$) 29 bivalents were formed at meiosis. The anaphasic separation was normal with $29 + 29$, although in some cases $30 + 28$ separation was also noticed. Four plants of the A_2 generation with $2n = 45$ resembled the prostratum parent and showed a typical *Drosera* type of pairing with 16 bivalents and 13 univalents at meiosis.

Kobayashi and Shimamura (1949, 1952) studied the cytology of tetraploid Sesame after 7 years from the date of induction. They noticed frequent occurrences of 13 quadrivalents during meiosis which disjoined normally. Mazzani and Micheletti de Zerpa (1953) observed in the subsequent generation of colchicine induced tetraploids of Sesamum, that during meiosis 5 to 10 quadrivalents were formed and the remaining were bivalents while Kedarnath (1954) reported variable number of quadrivalents, trivalents, bivalents and univalents in the autotetraploids of sesamum. Irregular separation of chromosome was noticed during meiosis.

Laws (1967) studied the meiosis in the induced tetraploids of *Oenothera* and found that all tetraploids showed a reduction in chiasma frequency. Most commonly an irregular numerical disjunction of the chromosomes at the first anaphase was observed. Occasionally univalents were not included in the metaphase plate and their random inclusion in the dyad could make an alteration in the chromosome complement. Thus dyad nuclei with more or less number of chromosomes were formed due to the lagging univalents. Infrequently, bridge formation at anaphase I was noticed. Anaphase bridge and lagging chromosomes were found in the anaphase II also.

Ramson and Levan (1939) observed 0 to 4 quadrivalents per cell in the pollen mother cells of tetraploid sugarbeets.

Amin (1940) observed 0 to 5 quadrivalents in the autopolyploids of cotton.

Wada (1940, 49, 50) proved that in Tradescantia apindle fibres did not develop at prophase with concentrations of 0.05 or 0.1 per cent colchicine.

Muntzing and Prakken (1940) showed that there was a genotypic controlled tendency in an autopolyploid 'Phleum' to form bivalents.

Ramanujan and Joshi (1941) studied the meiosis in the induced tetraploids of Glycer arietinum. Meiosis was fairly regular with the formation of varying numbers of quadrivalents at diakinesis and metaphase I. Seven quadrivalents per cell occurring most frequently. They observed a chromatin bridge and a chromosome fragment in a few cells at anaphase I.

Sparrow (1942) reported an increase in univalents about 37 per cent among the treated plants of Antirrhinum.

Vaarma (1947) recorded the threshold value of C-mitosis in the root tips of Ribes nigrum to be 0.01 per cent.

Gilles and Randolph (1951) found after analysing an autotetraploid maize strain at the beginning and for a period of ten years, in which selection for various features was taking place, that there were more bivalent associations and fewer quadrivalents at the end of the selection period than at the beginning.

Carpentier (1954) treated roots and rootlets of groundnut with 0.005 per cent colchicine solution and produced C-mitosis cells which showed a decrease in the frequency of total number of dividing cells.

Raman and Kesavan (1963) studied the chromosome association of induced autotetraploidy in Arachis duranensis. In 38 pollen mother cells examined at metaphase I, the frequency of quadrivalents ranged from 0 to 8. The most frequent association being 6 quadrivalents + 8 bivalents. In a few cells trivalents were noticed, but a corresponding number of univalents were not present. A single case of 20 bivalents was observed. At anaphase I 20-20 separation was most frequent.

Uchikuwa (1956) reported irregular meiosis in the colchicine induced tetraploid strains of two Japanese varieties of Cucumis sativus.

Douglas Davidson (1957) noted the ability of colchicine to induce chromosome breakage in Vicia faba. Evans et al (1957) used colchicine as an indicator in mitotic rate in broad bean root meristems and found that three concentrations, 0.1, 0.05 and 0.025 per cent were similar in capacity for inducing metaphasic accumulations. The treatments for more than 6 hours, showed an inhibitory effect which slowed down the rate of entry of cells into mitosis, the time spent in interphase being increased.

Sen and Cheda (1958) found that the colchicine induced tetraploids of black gram did not show much meiotic abnormalities. Quadrivalents were few.

Damon (1958) observed that colchicine treatment of sorghum seedlings caused the arrest of cell division at metaphase with production of star, exploded and ball metaphase types in the shoot apices.

Smith and Hiner (1960) observed typical C. mitosis in Allium cepa.

Islam (1960) found comparatively lesser number of quadrivalents in the tetraploids of *Annona squamosa*, trivalents and quadrivalents were also noted. Laggards were observed at anaphase I and II.

Koul (1964) related meiotic abnormalities to pollen sterility. He found that the separation of the chromosome complement at metaphase II and anaphase II into three unequal but independent functioning group, together with nondisjunction of chromosomes lead to the production of more variable gametes. It was therefore concluded that autopolyploidy had contributed much to meiotic instability.

Tarkowski (1965) scored the frequency of quadrivalents, trivalents, bivalents and univalents in the tetraploids of rye and showed that chromosomal aberrations did not decrease fertility, but result in aneuploidy.

Jenaki Ammal and Kaul (1967) made cytological studies on the induced autotetraploids of *Asparagus officinalis* L. Meiotic features observed included quadrivalents, trivalents and univalents at metaphase, laggards and bridges at anaphase and in some cases of multipolar spindle and nuclear break down.

MATERIALS AND METHODS

MATERIALS AND METHODS

The investigation was carried out in the Division of Agricultural Botany, Agricultural College and Research Institute, Vellayani during the year 1967-68.

A. Materials

Indiramma (1967) studied the effect of colchicine in inducing polyploidy in chillies. The seeds and seedlings of Capsicum frutescens L. of the strain Dutta series No. 2 were treated with colchicine at different concentrations and at varying duration of time. From the C_1 generation she got one tetraploid plant. This was obtained by treating the seedlings with 0.45 per cent colchicine for 9 hours. Selfed seeds from 41 pods from this plant were collected. These seeds together with the diploids as control, provided the material for the study of C_2 generation reported in this thesis.

B. Methods

Seeds from one pod was considered as one line. Out of the 41 pods, seeds from 29 pods only germinated. From these 29 lines, only 10 were selected (those having a population of 9 plants and above) and laid out in a

Randomised Block Design with three replications, in order to study statistically the relative variations in the polyploids in comparison with the diploids. The remaining 19 lines were randomised in a separate block and used for cyto-morphological investigation. Diploids were planted as guard rows; two rows on the four borders and one row each in between the four blocks. 25 diploid plants from these were selected at random for observations.

Characters studied

The following characters were studied.

1. Germination percentage

The seeds of the polyploid and the diploid plants were soaked in water for 20 hours on 1st October 1967. These soaked seeds were kept for germination on moist blotting paper in petri dishes. Seeds from individual pods were kept separately. The number of seeds germinated were counted every day and they were transferred to the pots in the field. Germination percentage was calculated when the germination of seeds ceased completely in all the petri dishes. The dates of commencement and completion of germination in the case of different treatments were recorded.

2. Early deformity

Abnormalities such as crinkling of leaves, retardation of growth, dark pigmentation of the leaves etc. shown by the plants were noticed from the very beginning and the same were recorded with respect to the concerned plants.

3. Height and spread of plants

Height of individual plants was measured in cm from the soil level to the tip of the tallest branch. This observation was made at weekly intervals from the date of transplanting to the date of harvest.

Spread of the individual plants was recorded once i.e., three days before the first harvesting, when the spread was maximum.

4. Leaf characters

(a) Area of leaves

Measurement of area of leaves was made in Sq. cm using graph paper technique (Darrow's method). For this 25 leaves were selected at random from each line, 60 days after transplanting the seedlings.

(b) Thickness of leaves

The thickness of leaves of the suspected polyploids was recorded from the randomly collected samples. For each plant twenty five hand sections were taken from the central portion of the leaves and were measured in microns (μ), under the microscope.

5. Flower counts

Opened flowers on each plant were counted daily until the flower production in all plants ceased completely. The total number of flowers produced per plant per line was estimated. Date of flowering and time taken for flowering from the date of germination of individual plants were also recorded.

6. Fruit set

The number of fruits produced was recorded at each harvest. The total production of fruits by individual plant was estimated and the percentage of fruit set per line was calculated.

7. Size of fruits

From every line 100 fruits were selected at random and their length and girth were measured in cm.

8. Weight of fruits

The fruits from each plant ^{are} was weighed at the time of each harvest and the data were recorded and analysed statistically.

9. Weight of seeds

Weight of 1000 seeds from each line was recorded in gram and the data were statistically analysed.

10. Yield

Yield of fruits for all the plants was recorded.

11. Pollen studies

(a) Pollen sterility

25 Anthers were collected from plants selected at random under each line at about 9 a.m. Pollen grains of each plant was dusted on a clean slide and stained with 1:1 glycerine acetocarmine. The slides were examined under the low power of a compound microscope after 30 minutes. Pollen sterility was estimated by counting the fertile and sterile pollen grains separately from 30 microscopic fields. Well filled and deeply stained pollen grains were classified as fertile and shrunken

and unstained ones as sterile pollen. Sterility for each line was calculated.

(b) Pollen size

The size of the pollen was determined by measuring the diameter. Diameter of 100 fertile pollen grains was measured in microns (μ) for each line, using a standard ocular micrometer.

12. Stomatal studies

(a) Distribution of stomata

A sample of 10 leaves was collected at random from each line. The tissue from the lower surface was peeled off and stained with 0.5 per cent safranin. The frequency of stomata per unit area was determined by counting their number from 30 microscopic fields at random for each line.

(b) Size of stomata

The length and width of 25 randomly selected stomata were measured for each line, using a standardised ocular micrometer.

All the above mentioned characters were studied

separately for the confirmed polyploids and compared with those of diploids.

Cytological observations

Suspected polyploids were marked on the basis of their gigas characters peculiar to polyploids as well as the morphological abnormalities like crinkling of leaves, dark pigmentation of leaves etc. Besides, ten plants were selected as suspected polyploids based on the statistical analysis for the morphological characters.

Indiremma (1967) had fixed the morphological characters like increased height of plant, number of flowers, number of fruits and yield per plant for polyploid types. In the present study the plants which fell over and above the confidence limits for the above characters for the diploids were marked out as suspected polyploids.

The flower buds of appropriate size from these suspected polyploid plants as well as from control plants were fixed in 1:3 acetic alcohol. The fixation was done from 9.30 to 10.30 a.m. for 6 hours. After fixation, the material was stored in 70 per cent alcohol. Gentle tapping and judicious warming favoured excellent spreading and differential staining of the chromosomes in the

microsporocytes. The chromosome number and their behaviour such as abnormalities or irregularities in meiotic behaviour were studied in the diploids and suspected polyploids and the stages were photographed.

Statistical procedure

The recorded data pertaining to the different characters under investigation were subjected to statistical analysis.

EXPERIMENTAL RESULTS

EXPERIMENTAL RESULTS

Morphological characters of plants in C₂ generation

Seeds obtained from the 29 fruits were sown separately and the percentage of germination in each line was recorded. The morphological characters of the plants raised from these lines were also studied. The characters studied were height of plant, spread of plant, number of branches, time taken for flowering, number of flowers, percentage of fruit set, number of fruits produced, yield of fruits, size and sterility of pollen grains, distribution of stomata and weight of seeds.

Percentage of seed germination

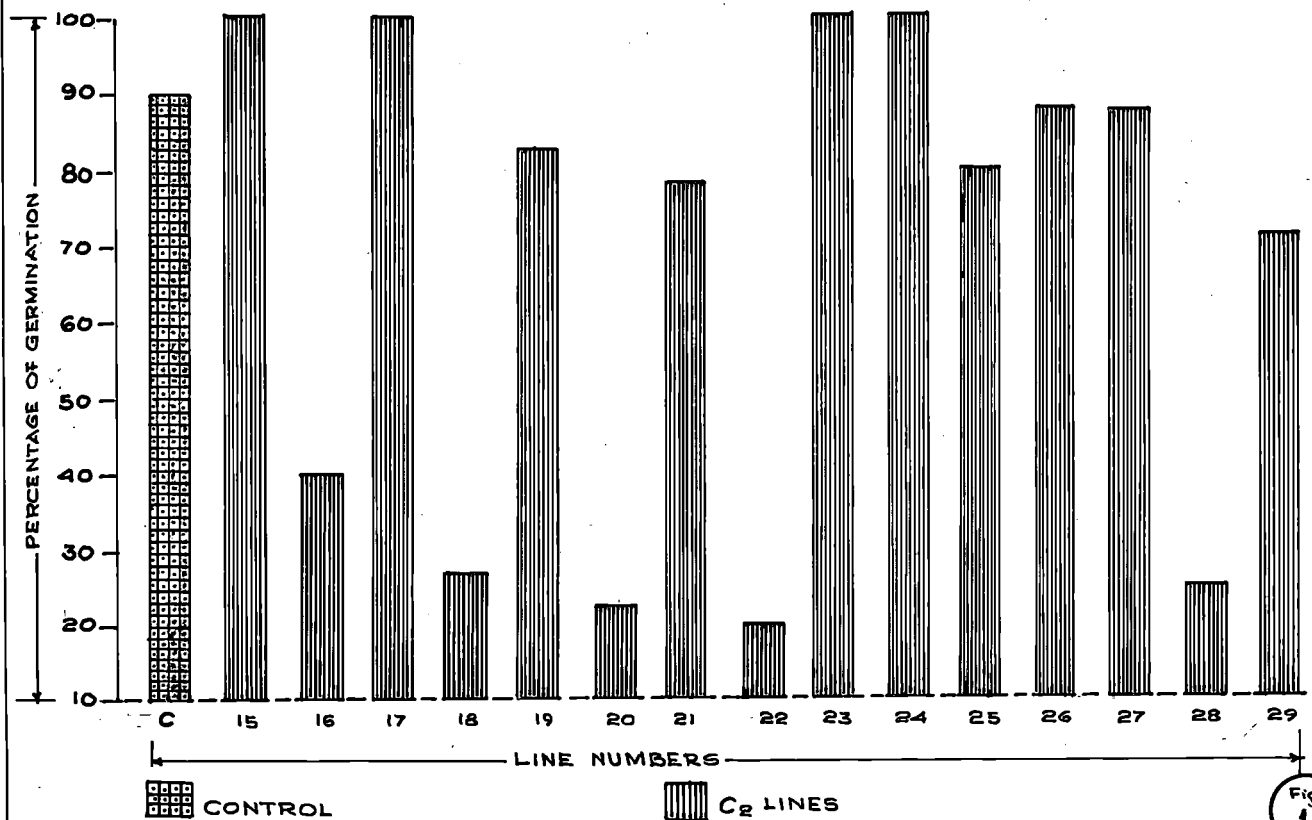
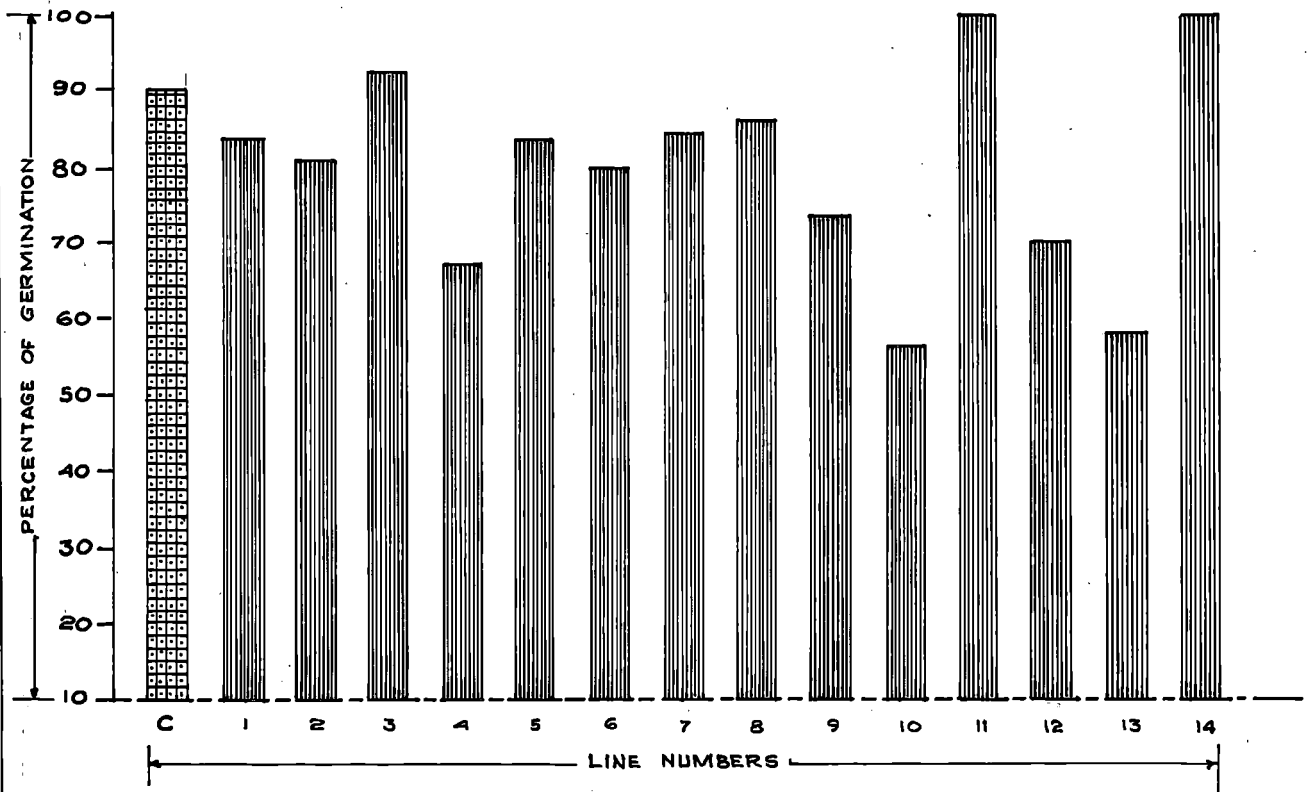
The data on the percentage of germination of seeds in 29 fruits obtained from the C₁ generation are presented in Table 1 a. There existed a wide range of variation in the percentage of germination. Seeds from fruits 11, 14, 15, 17, 23 and 24 showed 100 per cent germination. The minimum germination was noticed in the case of seeds obtained from line 22 (20 per cent). Germination in control was 90.06 per cent. The graphical representation for the percentage of seed germination is given in Fig. 1.

Table 1 a


Percentage of seed germination

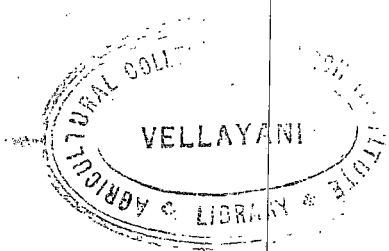
Line No.	Percentage of germination	Line No.	Percentage of germination
1	84.00	16	40.00
2	81.10	17	100.00
3	92.30	18	27.30
4	67.70	19	83.30
5	84.20	20	22.20
6	80.00	21	78.60
7	84.60	22	20.00
8	86.70	23	100.00
9	73.70	24	100.00
10	56.30	25	80.00
11	100.00	26	88.90
12	70.60	27	87.50
13	58.90	28	25.00
14	100.00	29	71.40
15	100.00	0	90.06

PERCENTAGE OF GERMINATION IN THE C_2 LINES



 CONTROL

 C_2 LINES



Rate of germination

Germination of seeds started from the fifth day of soaking in water in control as well as in most of the seeds obtained from the C_1 generation. Seeds from the fruits 17, 26 and 29 showed considerable delay in germination. The data on the rate of germination of seeds are presented in Table 1 b.

Height of plants

The data on the final height of plants recorded at harvest are given in Table 2 and the analysis of variance in Appendix I. The F ratio was found to be significant. Plants from the lines 2, 3, 5, 6, 7 and 8 were significantly taller from the control plants, while plants in the lines 1 and 4 showed a significant reduction in height. The mean plant height for different lines varied from 75.75 cm to 94.67 cm.

Spread of plants

The data on the spread of plants are presented in Table 2. From the results it will be seen that in lines 1, 2, 3 and 7, the spread of plants was significantly more than the control. The mean spread for the various lines varied from 56.75 cm (line 10) to 68.14 cm (line 3).

Table 1 b

Rate of seed germination

Line No.	No. of seeds kept for germination	Number of seeds germinated every 24 hours												Total No. of seeds germinated		
		5th day	6th day	7th day	8th day	9th day	10th day	11th day	12th day	13th day	14th day	15th day	16th day			
1	25	4	2	3	2	1	3	3	1	2						21
2	37	8	5	11	2	2	2	.	.	.						30
3	26	5	4	5	4	2	1	2	1	.						24
4	31	3	3	10	4	1						21
5	19	5	5	2	3	1						16
6	25	5	4	4	1	4	1	1	.	.						20
7	13	.	.	1	1	3	2	2	2	.						11
8	15	.	1	4	4	2	2	.	.	.						13
9	19	.	3	5	6						14
10	16	.	2	3	1	1	1	1	.	.						9
11	13	8	3	1	1						13

12	17	4	5	2	1	.	.	.							12
13	39	6	3	9	1	3	1	.							23
14	16	2	1	6	1	1	1	.							12
15	4	2	1	1							4
16	5	2	.	.							2
17	11	3	1	2	2	2	1		11
18	11	.	.	1	1	1		3
19	12	.	.	.	1	3	4	1	1		10
20	9	1	1		2
21	14	2	1	2	4	1	1		11
22	20	.	.	1	1	1	1		4
23	9	.	3	2	1	1	1	1		9
24	8	.	1	6	1	.	.	.							8
25	10	.	2	1	1	1	1	1	1		8
26	9	1	3	1	1	2	.		8
27	8	.	.	1	3	1	2		7
28	32	.	.	1	1	3	1	1	1		8
29	7	1	2	1	1		5
0	170	70	27	33	8	6	4	6		154

Table 2

Height, spread and number of branches

Line No.	Mean height (cm)	Mean spread (cm)	Mean number of branches
1	80.08	67.77	151.89
2	82.04	65.41	292.89
3	83.64	68.14	124.29
4	75.75	61.00	123.25
5	86.52	57.80	109.93
6	83.05	59.72	366.80
7	94.67	67.37	104.88
8	92.32	61.62	113.56
9	81.20	57.50	115.54
10	81.20	56.75	102.75
0	81.48	62.20	159.88

Number of branches

The data on the mean number of branches of plants in the different lines are given in Table 2. It will be seen from the table that the plants in lines 2 and 6 possessed significantly more number of branches than the control. Among the different lines the mean number of branches was maximum for line 6 (366.80) and minimum for line 10 (102.75). The number of branches recorded in control was 159.88. Analysis of variance for the number of branches is given in Appendix III.

Time taken for flowering

The data on the time taken for first flowering from the date of complete germination of seeds are presented in Table 3. The analysis of variance showed that F ratio was not significant. The days taken for flowering in different lines ranged from 55 in line 1 to 69 days in line 9. The time taken for first flowering by the control plants was 64 days.

Number of flowers

The data in Table 3 represents the mean number of flowers produced by the plants in different lines, which varied from 182.56 to 331.60. The maximum flower

Table 3

Time taken for flowering and number of flowers

Line No.	Time taken for flowering	Mean number of flowers
1	55.00	182.56
2	56.00	331.00
3	63.00	234.86
4	63.00	274.18
5	61.00	208.70
6	63.00	253.06
7	58.00	299.89
8	61.00	283.78
9	69.00	270.28
10	67.00	215.11
C	64.00	256.20

production was noticed in line 2 (331.60). Analysis of variance for number of flowers (Appendix V) showed that the F ratio was significant. Flower production in lines 2, 7, 9 and 10 was significantly higher than the control plants.

Percentage of fruits set

The data in the percentage of fruits set for the different lines are presented in Table 4. The fruits set showed variations between different lines ranging from 31.82 per cent in line 9 to 68.81 per cent in line 10. In control the fruit set was 63.82 per cent. The analysis of variance for the percentage of fruits set however, showed that F ratio was not significant (Appendix VI).

Number of fruits

Data on the mean number of fruits recorded in different lines are presented in Table 5. Line 10 recorded the maximum number of fruits (206.83) and the line 9 recorded the minimum (101.54). In control the mean number of fruits produced was 123.52. However, on statistical analysis there was no significant difference (Appendix VII).

Table 4

Percentage of fruits set and yield

Line No.	Percentage of fruit set	Mean weight of fruits in g
1	46.77	172.41
2	35.86	149.62
3	63.25	160.66
4	40.84	137.97
5	60.17	132.06
6	42.40	153.19
7	35.97	127.93
8	47.49	128.33
9	31.81	104.31
10	68.81	104.22
C	63.82	128.87

Yield of plants

The data pertaining to the yield of plants in grams in different lines is given in Table 4. Line 1 yielded the maximum (172.41 g). The minimum yield was obtained in line 10 (104.22 g). The yield of control plant is 128.87 g. The analysis of variance for yield of fruits is given in Appendix VIII. The F ratio was found to be not significant.

Size of fruits

The mean length and mean girth of fruits under different lines are presented in Table 5.

The length of pods was maximum in the line 3 (4.54 cm) whereas in the line 7 it was only 3.42 cm. In the control the mean length of pod was 3.89 cm. Except lines 6 and 7 all the other lines showed increased pod length than the control.

As far as the girth of the fruits was concerned all the lines except line 6 showed increased fruit girth, when compared to the control, in which case the pod girth was only 3.30 cm.

Area of leaves

The data on the mean area of leaves in different

Table 5

Number and size of fruits

Line No.	Mean number of fruits	Mean length of fruits (cm)	Mean girth of fruits (cm)
1	144.11	4.40	3.89
2	122.11	4.28	3.92
3	136.86	4.54	4.17
4	118.70	3.95	3.44
5	132.40	3.72	3.86
6	136.00	3.58	3.28
7	121.37	3.42	3.98
8	134.73	4.03	3.92
9	101.54	3.95	3.64
10	206.83	3.72	3.47
C	123.52	3.89	3.30

lines are given in Table 6. Appendix IX contains the analysis of variance for the area of leaves. The F ratio was not significant. However the plants in the lines 1, 5, 7, 8 and 9 showed a tendency for increased leaf area than the control.

Pollen sterility

Table 7 shows the percentage of pollen sterility among the different lines. Pollen sterility among the different lines varied from 21.75 to 37.42 per cent. In control 30.33 per cent of the pollen grains were found to be sterile. The X^2 analysis for the data on the extent of pollen sterility (Appendix XIII) did not show any significant difference for the G_2 lines when compared to the control.

Pollen size

The data on the mean diameter of pollen under different treatments is presented in Table 7. The mean diameter of pollen grains among different lines varied from $13.02 \pm 5.12 \mu$ to $17.09 \pm 1.63 \mu$. The analysis of variance for the data on pollen size under different lines is given in Appendix X. The F ratio was found to be not

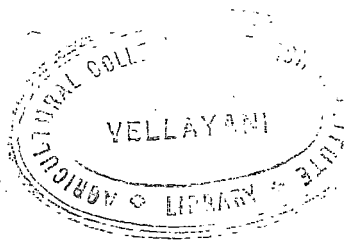


Table 6

Area of leaves

Line No.	Mean area (in Sq. cm)
1	208.56
2	126.78
3	140.75
4	137.77
5	152.29
6	122.40
7	168.99
8	161.22
9	158.39
10	127.67
0	144.44

Table 7

Pollen sterility and size

Line No.	Percentage of sterility	Mean diameter of pollen grains (in μ)
1	30.10	14.58 \pm 0.81
2	21.75	13.94 \pm 0.68
3	24.66	16.68 \pm 2.44
4	28.62	13.02 \pm 5.12
5	30.26	17.09 \pm 1.63
6	29.83	13.02 \pm 7.86
7	31.50	15.32 \pm 1.49
8	37.42	13.56 \pm 1.36
9	35.28	13.42 \pm 13.15
10	35.30	15.57 \pm 1.36
0	30.33	9.36 \pm 0.95

significant. However all the lines showed bigger pollen grains than the control plants.

Distribution of stomata

The mean number of stomata per unit area is given in Table 8. Among the different lines the maximum number of stomata per unit area was found in line 2 (9.07) and the minimum in line 9 (6.62). The analysis of variance for the distribution of stomata (Appendix XI) showed that the F ratio was significant. The number of stomata per unit area was significantly lower in all the lines from that of the control.

Size of stomata

Both length and width of stomata were found to be varying in the G_2 lines (Table 8).

The mean length of stomata was maximum in line 3 ($28.88 \pm 5.83 \mu$) and minimum in line 9 ($17.49 \pm 3.53 \mu$). In the control stomatal length was $25.22 \pm 5.01 \mu$.

The mean width of stomata ranged from $7.05 \pm 6.64 \mu$ in line 4 to $18.58 \pm 3.80 \mu$ in line 8. In the control plants the width of stomata was $13.42 \pm 2.71 \mu$.

Table 8

Distribution and size of stomata

Line No.	Mean number of stomata per unit area	Size of stomata (in μ)	
		Mean length	Mean width
1	7.69	17.89 \pm 2.71	13.42 \pm 2.71
2	9.07	26.03 \pm 5.42	13.15 \pm 2.71
3	8.85	28.88 \pm 5.83	14.78 \pm 2.71
4	8.74	24.41 \pm 5.42	7.05 \pm 6.64
5	8.32	26.98 \pm 5.42	11.39 \pm 2.44
6	7.59	22.37 \pm 4.07	13.42 \pm 2.71
7	7.36	18.03 \pm 4.07	14.10 \pm 2.71
8	6.69	26.71 \pm 5.42	18.58 \pm 3.80
9	6.62	17.49 \pm 3.53	14.50 \pm 2.71
10	7.14	23.19 \pm 4.07	14.50 \pm 2.71
0	9.96	25.22 \pm 5.02	13.42 \pm 2.71

Weight of seeds

The data presented in Table 9 represents the mean weight of 1000 seeds obtained from different lines. Analysis of variance for the data is given in Appendix XII. The F ratio is not significant, indicating that C_2 lines did not show significant difference from the control. However all the polyploid lines showed a tendency for higher seed weight than the control. The weight of 1000 seeds in control was 3.56 g.

Morphological abnormalities

Of the 236 plants raised in the C_2 generation certain plants exhibited morphological abnormalities like stunted growth and darker pigmentation of the foliage and crinkling of leaves. 32 plants showed crinkling of the leaves. In 2 plants the leaves had irregular margins. It was also observed that one plant possessed bilobed leaves.

Heterogeneity of the C_2 population

The data on the height and spread of plants, number of flowers and fruits and weight of fruits were subjected to χ^2 analysis in order to test the heterogeneity of the material used for raising the C_2 population. It

Table 9

Weight of seeds

Line No.	Weight of 1000 seeds (in g)
1	4.35
2	3.81
3	4.37
4	4.33
5	5.53
6	4.96
7	6.58
8	9.02
9	6.60
10	6.26
0	3.56

was found that the population was highly heterogenous. The X^2 values for all the five characters analysed were found to be highly significant. The X^2 analysis is given in Appendix XIV.

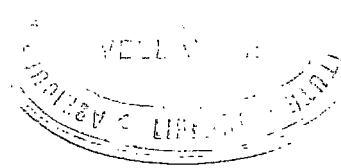
Isolation of suspected polyploids based on gigas characters

The studies on the morphological characters (vide Tables 1 to 9) showed that there existed a wide range of variation in the plants between the different lines. The plants which exhibited characters like increased height, (Plate 1, 2) maximum spread, (Plate 3, 4) more number of branches, maximum number of flowers, fruits and increased yield were marked out statistically, based on the confidence limits fixed for diploids. Plants which fell over and above the confidence limits were marked as polyploids. Thirteen plants were thus marked out. Three of them were found to be diploids and the remaining ten to be polyploids on cytological analysis. The data pertaining to the above characters of these ten plants are given in Table 10. Other characters such as time taken for flowering, percentage of fruits set, size of fruits, area of leaves and thickness of leaves of the respective plants are also given in Table 10. Table 11 contains sterility and size of pollen grains and Table 12 contains distribution and size of

Table 10. Morphological

Plants No.	Height of plants (cm)	Spread of plants (cm)	No. of branches	Time taken for flowering	No. of flowers	No. of fruits
32	102.20	88.00	148	61	380	188
34	103.60	71.00	124	65	368	161
61	99.40	78.00	202	47	304	163
84	105.00	68.00	140	57	331	176
134	88.20	67.00	150	51	312	189
135	100.80	75.00	165	64	333	196
187	107.80	70.00	128	69	326	147
197	98.56	67.00	126	70	296	158
201	89.88	74.00	152	49	302	152
236	106.12	69.00	110	72	290	141
Control	29.10	61.80	160	64	256	124

Note: Polyploid plants were marked out



variation among the polyploids

Size of fruits (in inch)		% of fruit set	Yield of plants (in g)	Area of leaves (in Sq. cm)	Thickness of leaves (in μ)
Length	Girth				
1.15	1.07	49.47	229.82	100.23	1.33 \pm 0.27
1.23	1.15	43.75	186.60	103.43	1.43 \pm 0.29
1.10	1.23	53.62	153.60	84.23	3.04 \pm 0.61
1.02	1.00	53.17	204.75	93.34	1.36 \pm 0.27
1.59	1.41	60.57	180.83	95.46	2.33 \pm 0.47
2.00	1.05	58.85	197.10	103.43	4.07 \pm 0.82
1.80	1.84	45.09	168.40	100.22	3.66 \pm 0.73
1.01	0.97	53.37	157.35	92.69	1.00 \pm 0.20
1.44	1.56	50.33	150.46	100.99	4.34 \pm 0.87
1.34	0.88	48.62	140.90	87.38	2.33 \pm 0.47
1.39	1.18	48.21	128.88	84.93	0.98 \pm 0.20

statistically and confirmed cytologically

Table 11

Variation in pollen sterility and pollen size
among the polyploids

Plant No.	Pollen sterility (in %)	Pollen size (in μ)
32	44.18	13.69 \pm 1.36
34	42.00	21.15 \pm 2.16
61	41.29	18.44 \pm 1.89
84	27.76	14.24 \pm 1.36
134	40.29	13.42 \pm 1.36
135	35.38	24.81 \pm 2.44
187	39.99	12.88 \pm 1.22
197	44.95	19.53 \pm 1.89
201	27.41	16.95 \pm 1.76
236	55.49	13.29 \pm 1.36
Control	30.07	9.36 \pm 0.95

Note: Polyploid plants were marked out statistically and confirmed cytologically

Table 12

Variation in the distribution and size of stomata
among the polyploids

Plant No.	Frequency of stomata/ unit area	Size of stomata (in μ)	
		Mean length	Mean width
32	5.34	26.58 \pm 0.27	11.12 \pm 2.17
34	4.33	21.15 \pm 2.17	1.08 \pm 2.17
61	8.46	13.69 \pm 1.36	5.02 \pm 1.89
84	4.83	26.58 \pm 0.27	10.71 \pm 2.17
134	7.16	17.89 \pm 1.76	13.15 \pm 2.58
135	8.34	19.79 \pm 3.93	9.89 \pm 2.03
187	4.23	18.03 \pm 3.66	10.17 \pm 2.03
197	5.93	16.14 \pm 3.25	12.07 \pm 2.44
201	7.16	28.48 \pm 5.69	11.53 \pm 2.31
236	6.86	31.05 \pm 6.24	15.32 \pm 3.12
Control	9.97	25.22 \pm 5.02	13.42 \pm 2.71

Note: Polyploid plants were marked out statistically
and confirmed cytologically

stomata of these plants. In the polyploid plants the leaves were thicker than the diploids (Plate 5). The size of the stomata was also larger in the polyploid types which resulted in the distribution of less number of stomata per unit area (Plate 6). The fruit size in the polyploids was smaller when compared to that of diploids (Plate 7). However the seeds were bigger in size (Plate 8). Details of cytological analysis of the above plants are given separately (vide Table 16).

Isolation of suspected polyploids based on morphological abnormalities

In the C_2 generation certain plants showed extreme abnormalities such as crinkling and lobing of leaves, darker pigmentation of the foliage and stunted growth. Such plants were separately studied and the data for their morphological characters, fruit set, yield, area of leaves and thickness of leaves were given in Table 13, pollen sterility and size in Table 14 and distribution and size of stomata in Table 15.

From the tables it will be seen that the plants which exhibited abnormalities showed increased pollen size, increased leaf area, less number of stomata per unit area, increased stomatal length and increased thickness of

Table 13. Morphological

Plant No.	Height of plants (cm)	Spread of plants (cm)	No. of branches	Time taken for flowering	No. of flowers	No. of fruits
15	112.00	34.00	35.00	57	78	15
16	95.20	32.00	40.00	57	180	100
25	82.60	41.00	128.00	59	413	336
35	85.40	71.00	139.00	65	485	240
65	105.00	50.00	95.00	51	305	217
86	69.72	60.00	118.00	76	219	137
116	111.00	61.00	80.00	56	48	20
140	85.40	68.00	120.00	64	228	140
142	92.40	36.00	52.00	88	103	83
146	109.20	45.00	162.00	71	379	217
153	84.00	40.00	98.00	65	197	145
169	120.40	68.00	86.00	75	265	216
180	106.40	32.00	46.00	77	85	75
213	75.60	50.00	118.00	73	627	368
Control	29.10	61.80	160.00	64	256	124

Note: Plants showing abnormalities were

variations among the polyploids

Size of fruits (in inch)		% of fruit set	Yield of plants (in g)	Area of leaves (in Sq. cm)	Thickness of leaves (in μ)
Length	Girth				
1.88	1.08	19.07	20.52	103.64	17.89 \pm 3.53
0.97	1.06	55.56	98.30	95.34	20.75 \pm 4.20
0.99	1.05	81.35	33.70	92.66	15.73 \pm 4.34
1.83	1.50	49.48	200.40	100.73	18.03 \pm 3.66
1.10	1.55	71.14	220.90	104.24	29.43 \pm 5.83
1.55	1.52	62.55	141.30	86.34	18.58 \pm 3.66
1.34	1.00	41.66	25.00	65.31	28.88 \pm 5.83
0.99	0.87	61.40	140.40	93.43	35.79 \pm 7.19
0.87	1.34	80.58	85.70	106.34	41.90 \pm 8.41
1.33	0.34	61.51	289.30	101.56	43.66 \pm 8.68
1.34	1.23	73.61	146.00	95.55	26.85 \pm 5.29
1.24	1.03	81.51	220.00	99.53	27.53 \pm 5.56
1.11	1.02	88.23	75.90	104.63	28.61 \pm 5.69
1.34	0.56	50.41	369.90	116.53	13.97 \pm 2.98
1.39	1.18	48.21	128.88	84.93	13.29 \pm 2.71

confirmed cytologically as polyploids

Table 14

Variation in pollen sterility and pollen size
among plants showing abnormal characters

Plant No.	Pollen sterility (in %)	Pollen size (in μ)
15	49.07	20.75 \pm 2.03
16	34.77	14.10 \pm 1.37
25	35.47	13.02 \pm 0.95
35	35.24	7.86 \pm 0.81
65	55.50	18.31 \pm 1.76
86	30.56	14.37 \pm 1.49
116	35.46	43.79 \pm 4.34
140	39.60	12.61 \pm 1.22
142	38.36	19.53 \pm 1.89
146	73.09	17.49 \pm 1.76
153	53.21	20.61 \pm 2.03
169	49.50	16.68 \pm 1.63
180	47.56	14.64 \pm 1.49
211	45.35	25.49 \pm 2.58
Control	30.07	9.36 \pm 0.95

Note: Plants showing abnormalities were confirmed cytologically as polyploids.

Table 15

Variation in the distribution and size of stomata
among the plants showing abnormal characters

Plant No.	Frequency of stomata/ unit area	Size of stomata (in μ)	
		Mean length	Mean width
15	6.03	17.76 \pm 3.53	9.35 \pm 1.89
16	7.46	19.39 \pm 3.93	11.39 \pm 2.31
25	8.53	24.41 \pm 4.88	12.61 \pm 2.58
35	8.44	11.12 \pm 2.17	11.12 \pm 2.17
65	5.76	26.04 \pm 5.15	13.42 \pm 2.58
86	7.46	29.56 \pm 5.97	11.25 \pm 2.31
116	5.34	26.98 \pm 5.29	10.17 \pm 2.03
140	9.76	27.12 \pm 5.42	11.79 \pm 1.63
142	6.43	26.85 \pm 5.29	9.89 \pm 2.31
146	6.26	33.09 \pm 8.00	10.17 \pm 2.03
153	6.44	28.75 \pm 5.83	11.25 \pm 2.31
169	7.76	13.42 \pm 2.58	13.02 \pm 2.58
180	7.34	12.61 \pm 2.58	10.58 \pm 2.17
213	6.63	24.00 \pm 4.75	12.61 \pm 2.58
Control	9.97	25.22 \pm 5.02	13.42 \pm 2.71

Note: Plants showing abnormalities were confirmed cytologically as polyploids

leaves. However with respect to height and spread these plants showed variations. These plants were cytologically examined and the conclusions derived are presented separately (vide Table 16).

Cytological observations on the suspected polyploids

Out of the 236 plants of the G_2 generation 24 plants which in general, exhibited gigas plant characters and morphological abnormalities, as already mentioned, were suspected to be polyploids. Studies on pollen and stomata were also used as criteria to confirm the polyploid nature of the above plants. The behaviour of chromosomes of these 24 suspected polyploids at meiosis was studied. Cytological studies of the diploids, which were used as control, also were conducted.

A detailed analysis of the cytological observations of some out of the 24 suspected polyploids are presented in Table 16.

The chromosome number of these polyploids were found to be $2n = 48$, as they form 24 bivalents at diakinesis and the chromosome number of the diploids $2n = 24$ as they form 12 bivalents at diakinesis. Even though regular bivalents at diakinesis and normal tetrads

Table 16

Cytological observations in polyploid plants

Plant No.	Meiosis I			Meiosis II		Abnormalities recorded
	Diakinesis	Metaphase I	Anaphase I	Anaphase II	Telophase II	
15	24 IIs (Plate 9)	..	24 + 24	4 groups of chromosomes	normal tetrads	2 laggards in one cell at anaphase I
25	22 IIs + 1 IV	..	24 + 24	4 groups of chromosomes	normal tetrads (Plate 10)	3 and 5 groups of chromosomes were observed at anaphase II in 4 out of 13 cells. Multivalents were noted in 2 cells
86	24 IIs	..	24 + 24 (Plate 11)	4 groups of chromosomes	normal tetrads	A chromatin bridge was found in one cell at anaphase I. (Plate 12) The number of lagging chromosome ranges from 1 - 5. 3 - 5 groups of chromosomes were also observed at telophase II. (Plate 13, 14).
135			24 + 24	4 groups of chromosomes	normal tetrads	Multivalents were noticed in 4 cells. 1 - 9 laggards were found in one cell

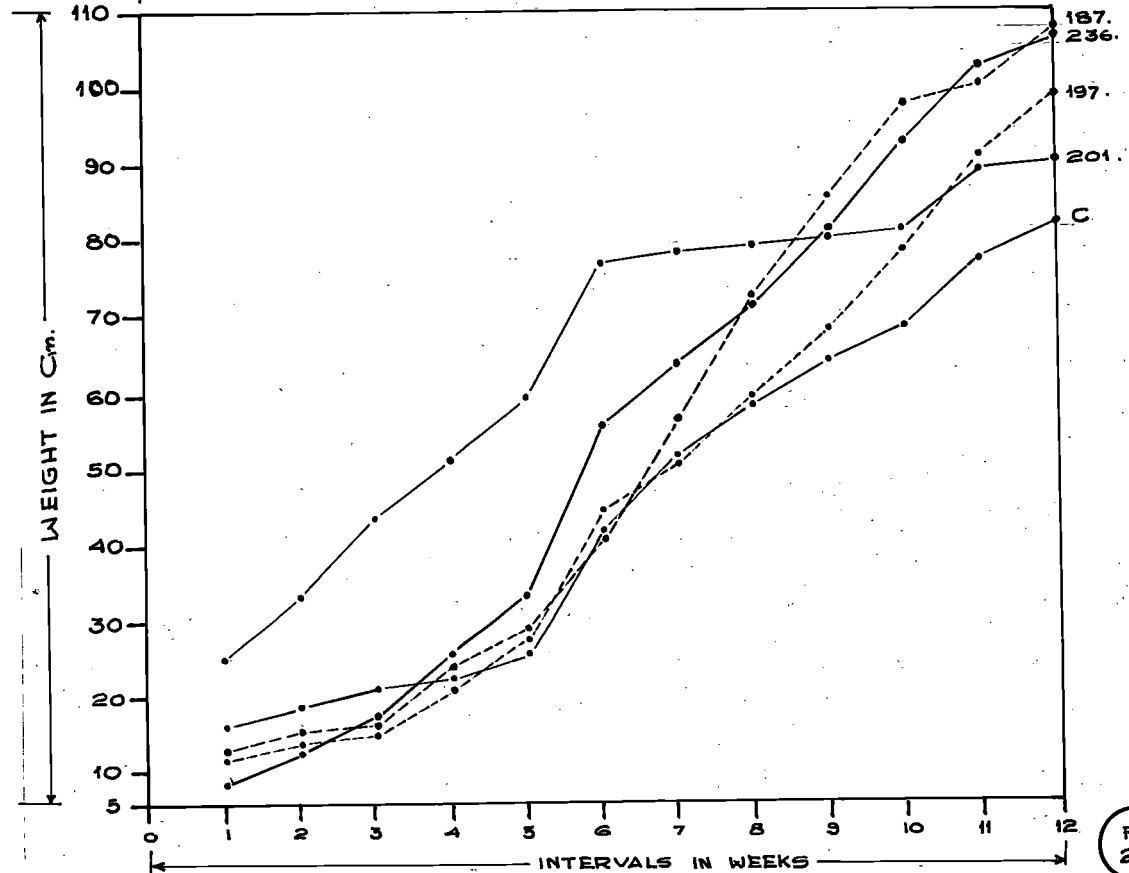
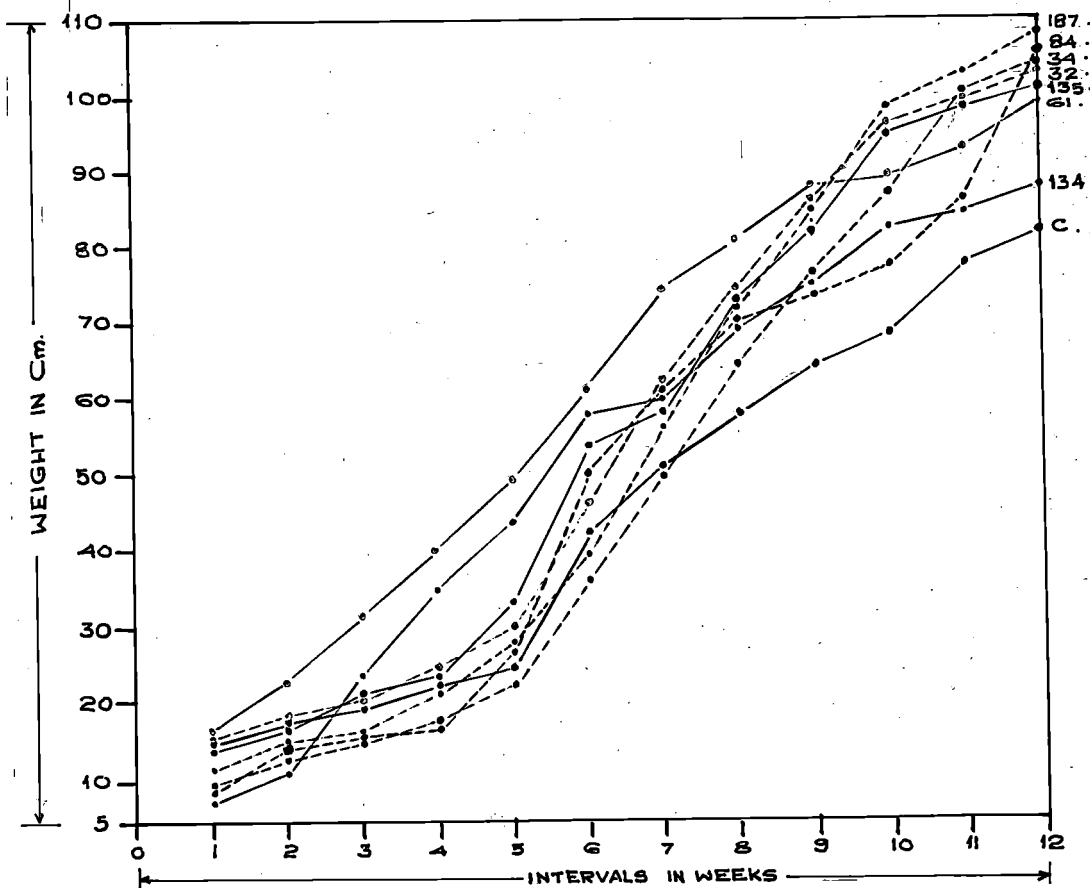
140	24 IIs	..	24 + 24	4 groups of chromosomes	normal tetrads	Nil
142	24 IIs	..	24 + 24	4 groups of chromosomes	normal tetrads	3, 5 & 6 groups of chromosomes were noticed in many cells at anaphase II.
146	24 IIs	..	24 + 24	4 groups of chromosomes	normal tetrads	4 laggards in one cell (Plate 15)
197	22 IIs + 1 IV	..	24 + 24	4 groups of chromosomes	normal tetrads	Multivalent formation was noticed in 2 cells (Plate 16)
201	22 IIs + 1 IV	..	24 + 24	4 groups of chromosomes	normal tetrads	Multivalent formation was noticed in 1 cell. 3, 5 & 6 groups of chromosomes laggards also were found at anaphase II (Plate 17)
236	24 IIs	(Plate 18)	24 + 24	4 groups of chromosomes	normal tetrads	3, 5 & 6 groups of chromosomes were found in some cells. Lagging chromosomes were observed in 6 cells. The number of laggards ranged from 2 - 8 (Plate 19)
Control	12 IIs (Plate 20)		12 + 12 (Plate 21)	4 groups of chromosomes	normal tetrads (Plate 22)	Nil

were formed at telophase, occasionally irregularities in the chromosome behaviour such as multivalent formation, laggards and many telophasic chromosome groups and rarely chromatin bridges were observed. Thus the polyploids showed considerably high degree of aberrations with respect to chromosome behaviour which is also evident from the Table 16.

The graphical representation for the various characters of the confirmed polyploids are given in figures 2 a to 12.

RATE OF GROWTH OF POLYPLOIDS

(PLANTS SHOWING GIGAS CHARACTERS)



RATE OF GROWTH OF POLYPLOIDS

(PLANTS SHOWING ABNORMALITIES)

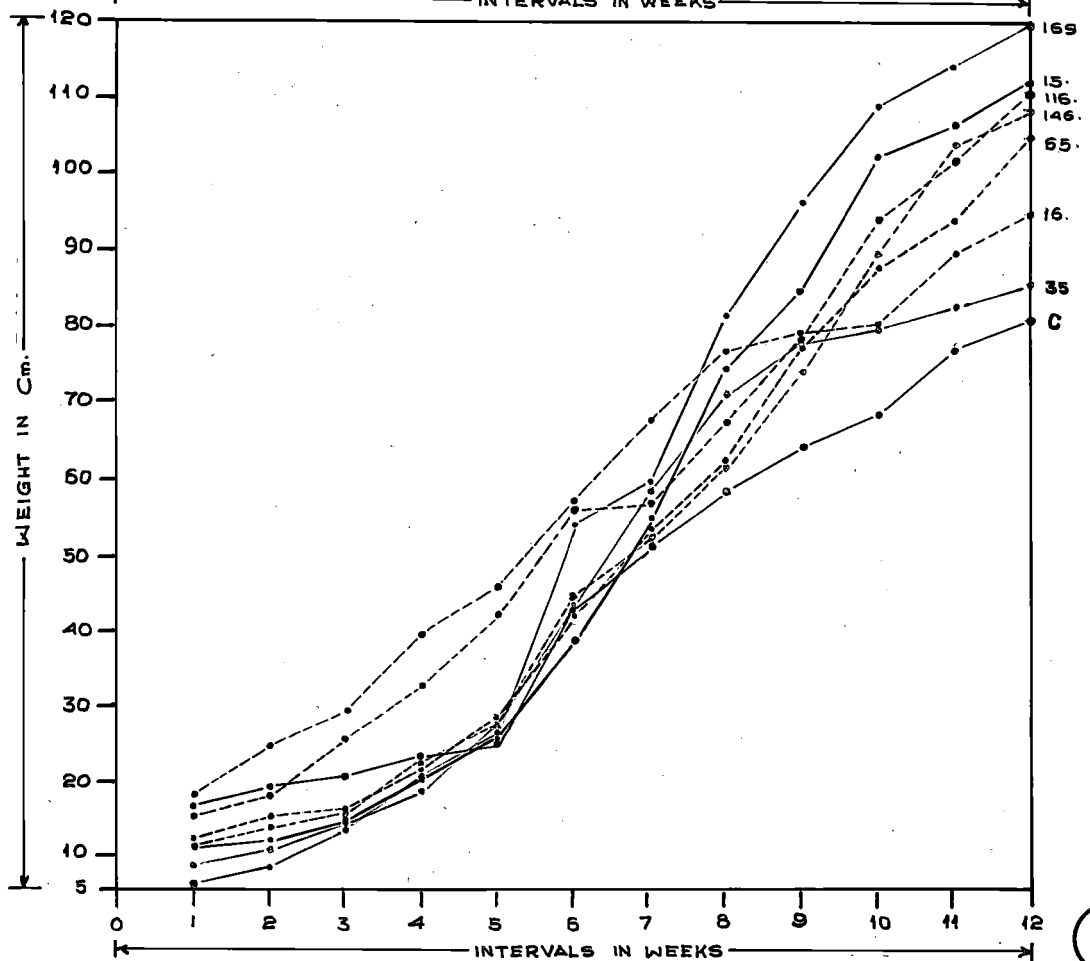
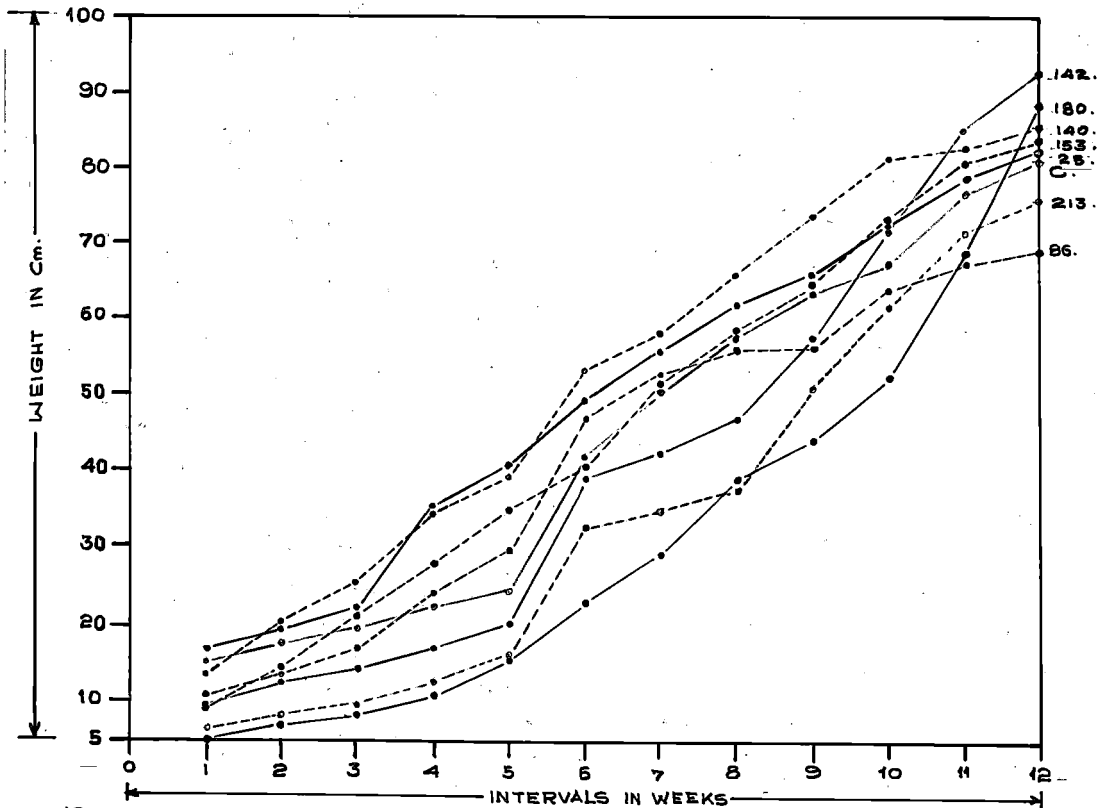
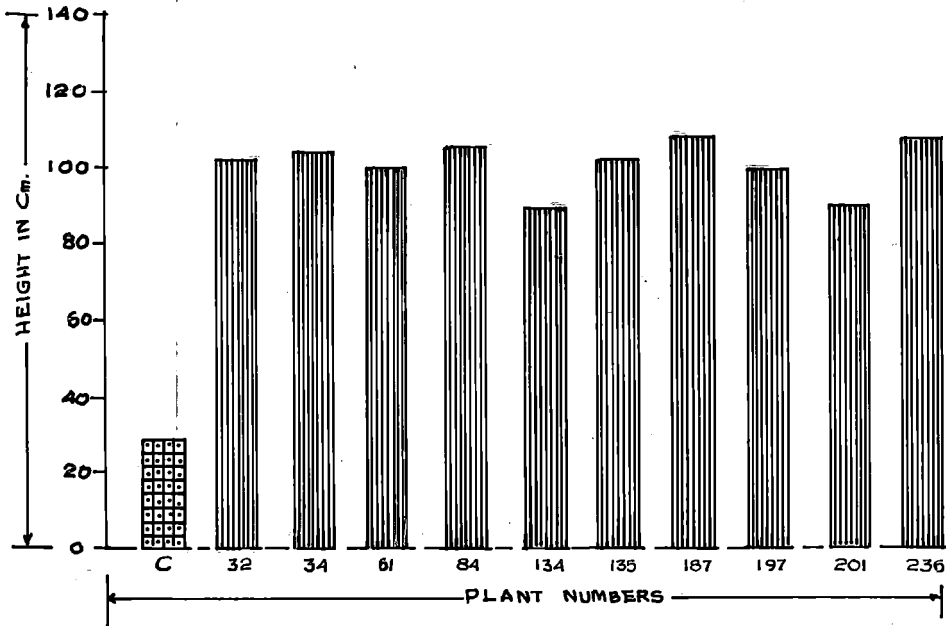


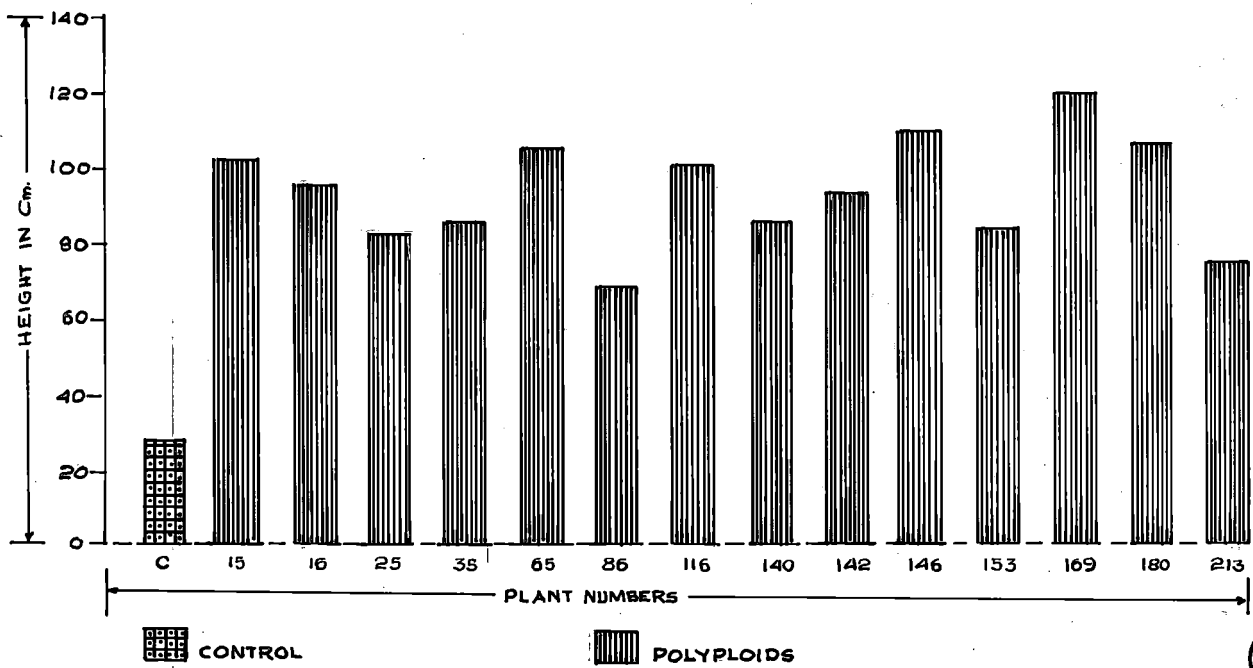
Fig 2b.

VARIATIONS IN HEIGHT AMONG POLYPOIDS

① (PLANTS SHOWING GIGAS CHARACTERS)



② (PLANTS SHOWING ABNORMALITIES)

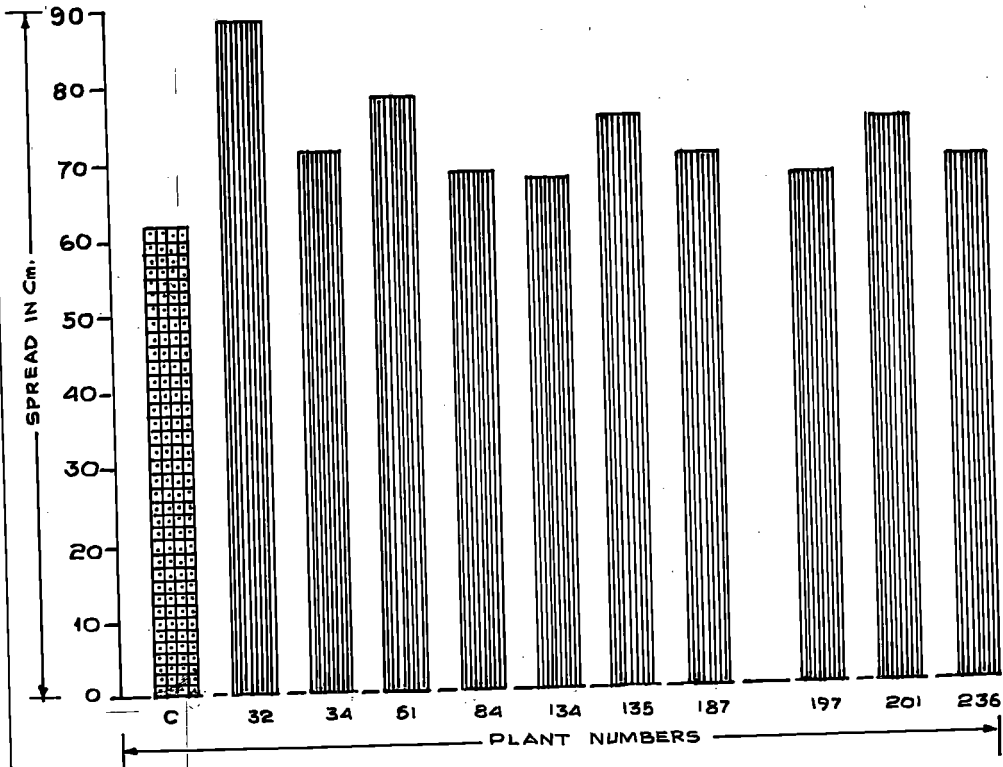


CONTROL

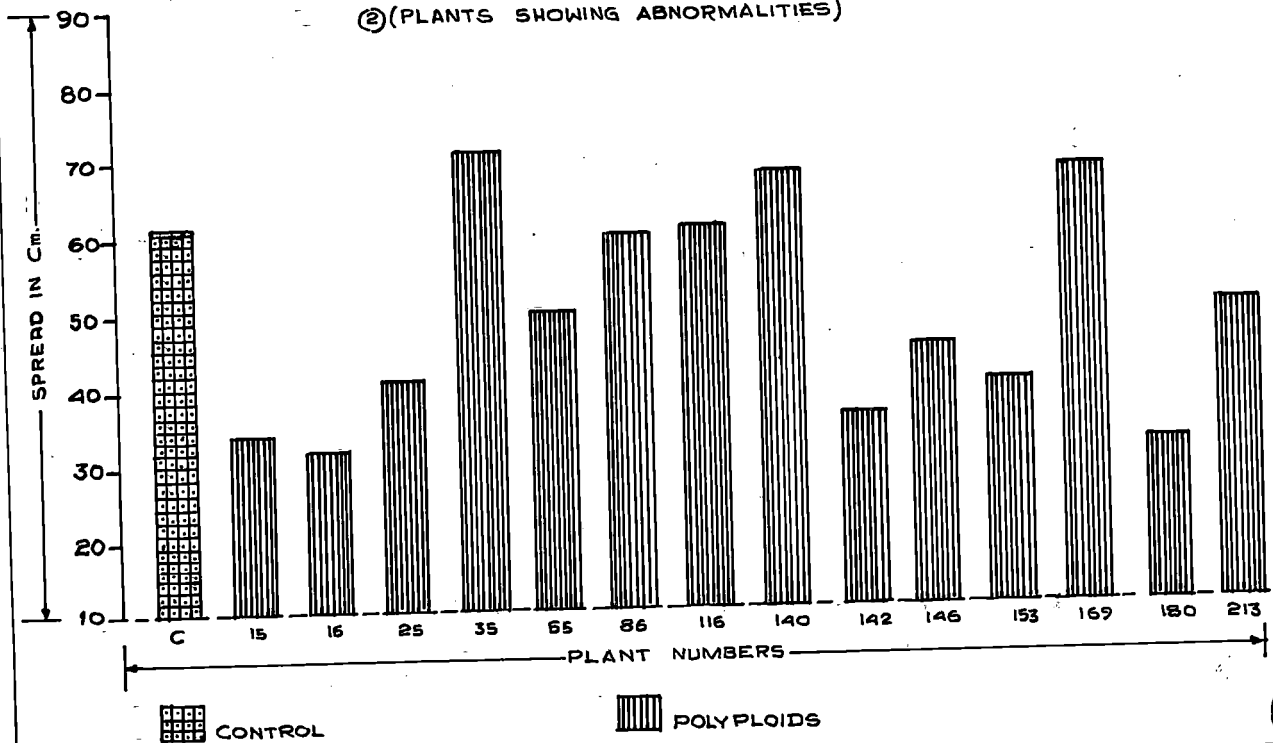
POLYPOIDS

VARIATIONS IN SPREAD AMONG POLYPLOIDS

① (PLANTS SHOWING GIGAS CHARACTERS)



② (PLANTS SHOWING ABNORMALITIES)



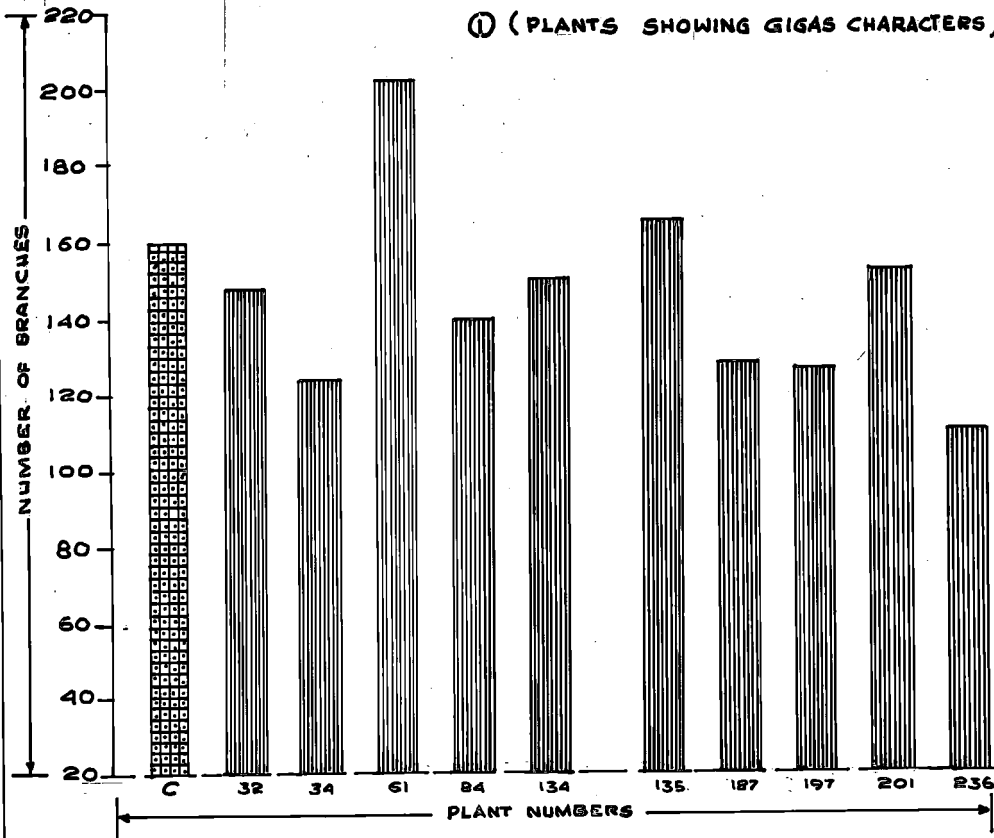
CONTROL



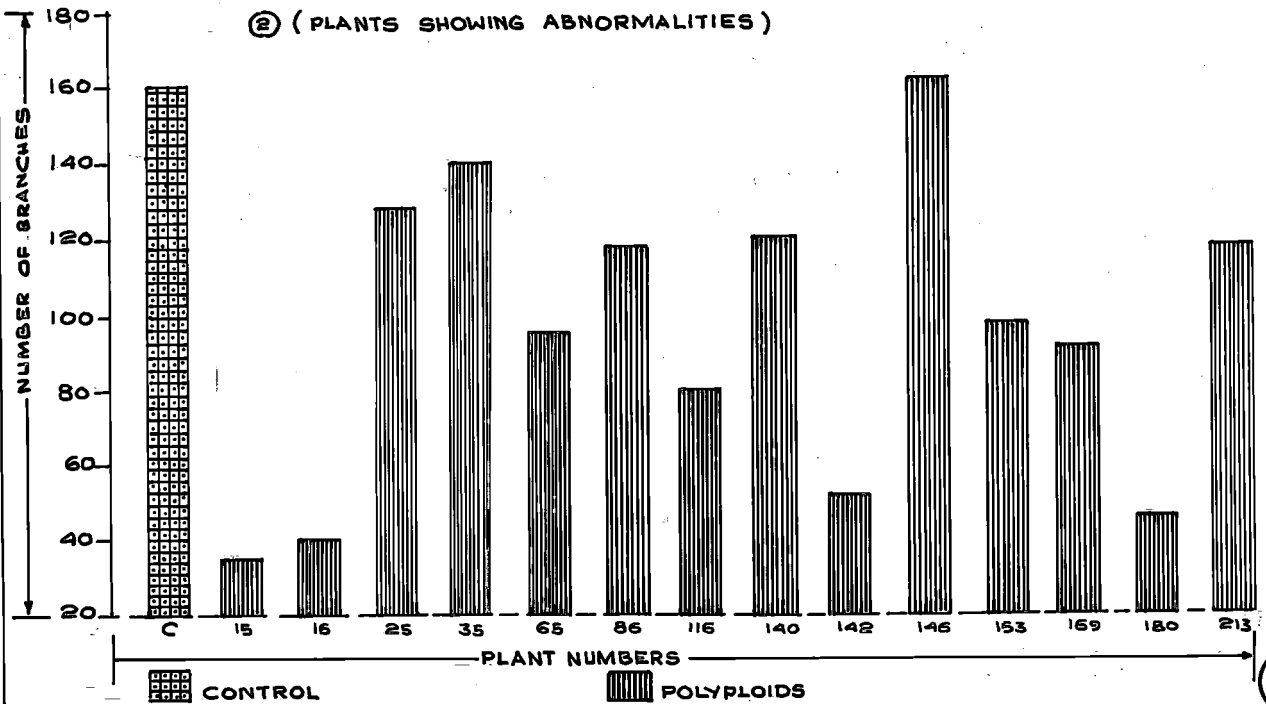
POLYPLOIDS

VARIATIONS IN NUMBER OF BRANCHES AMONG POLYPLOIDS

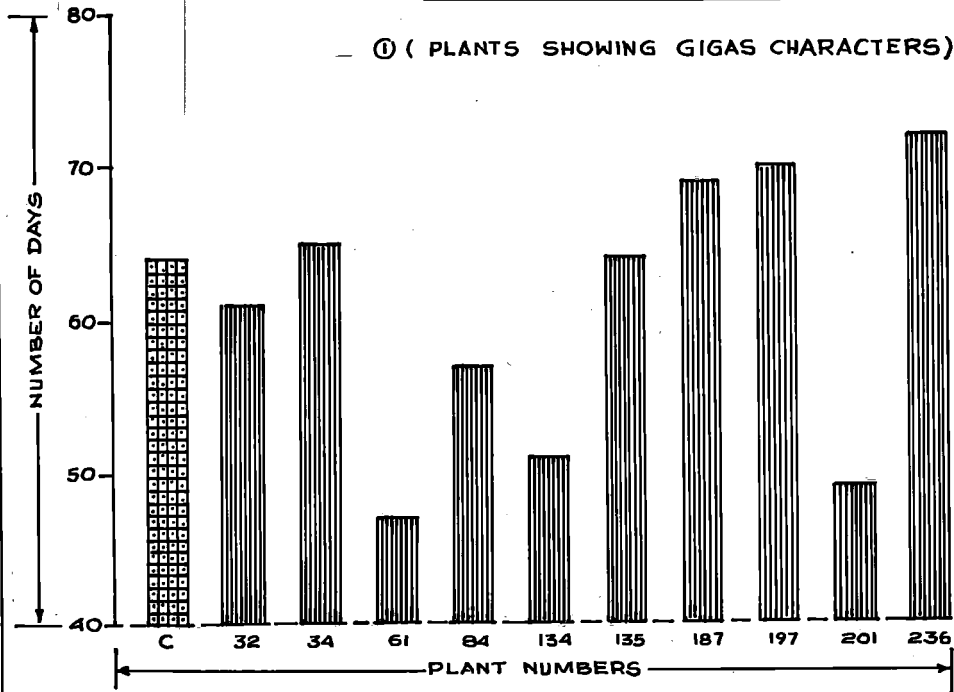
① (PLANTS SHOWING GIGAS CHARACTERS)



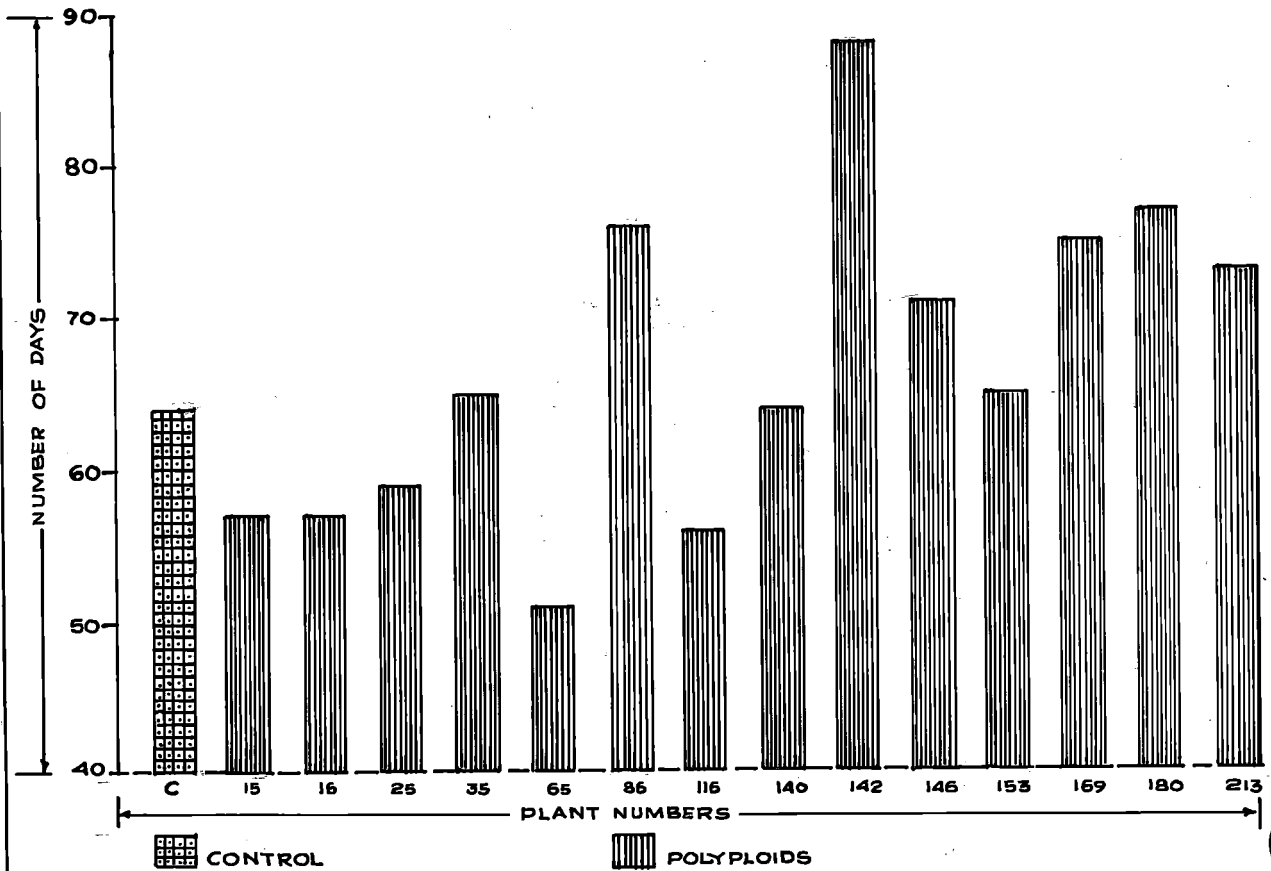
② (PLANTS SHOWING ABNORMALITIES)



VARIATIONS IN THE TIME TAKEN FOR
FLOWERING AMONG POLYPLOIDS

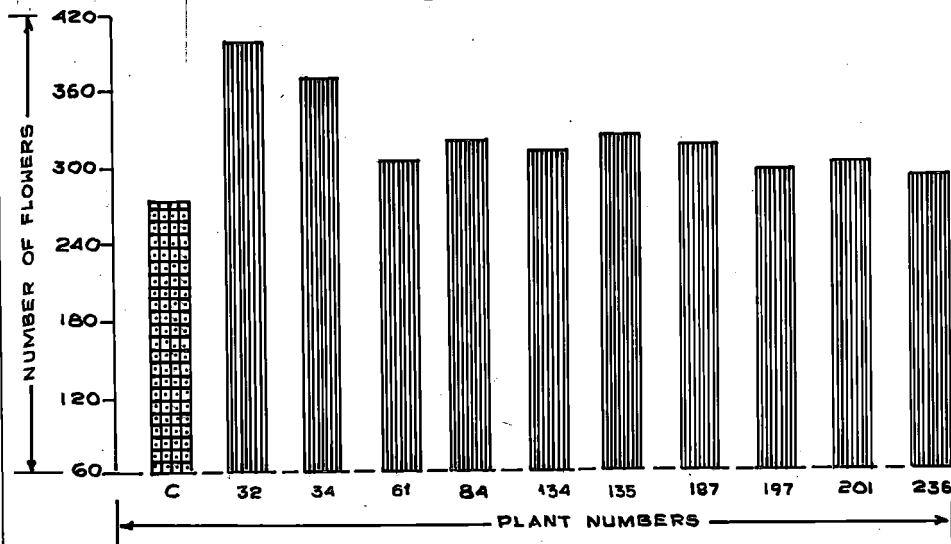


② (PLANTS SHOWING ABNORMALITIES)

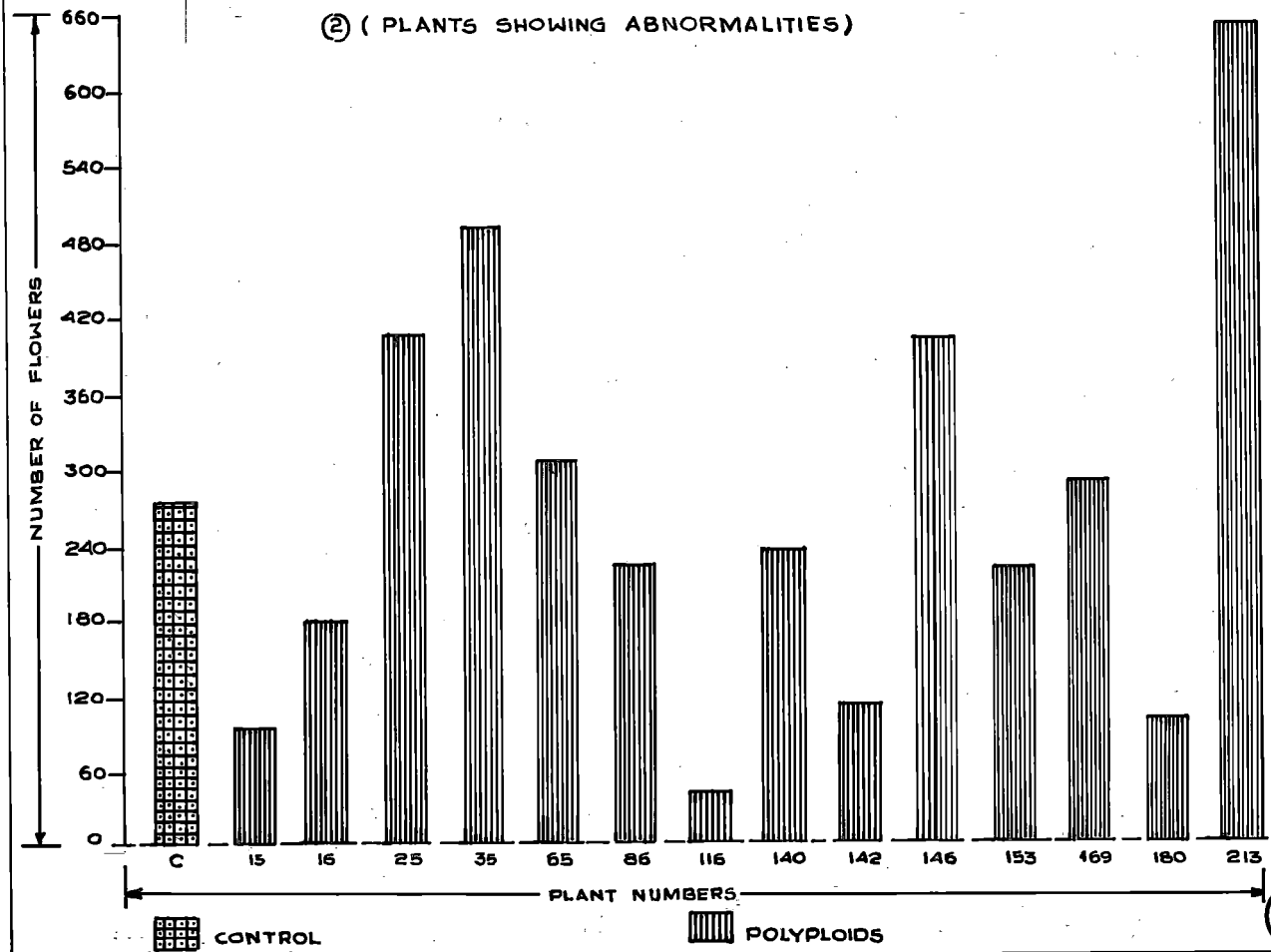


VARIATIONS IN THE NUMBER OF FLOWERS PRODUCED BY THE POLYPOIDS

① (PLANTS SHOWING GIGAS CHARACTERS)

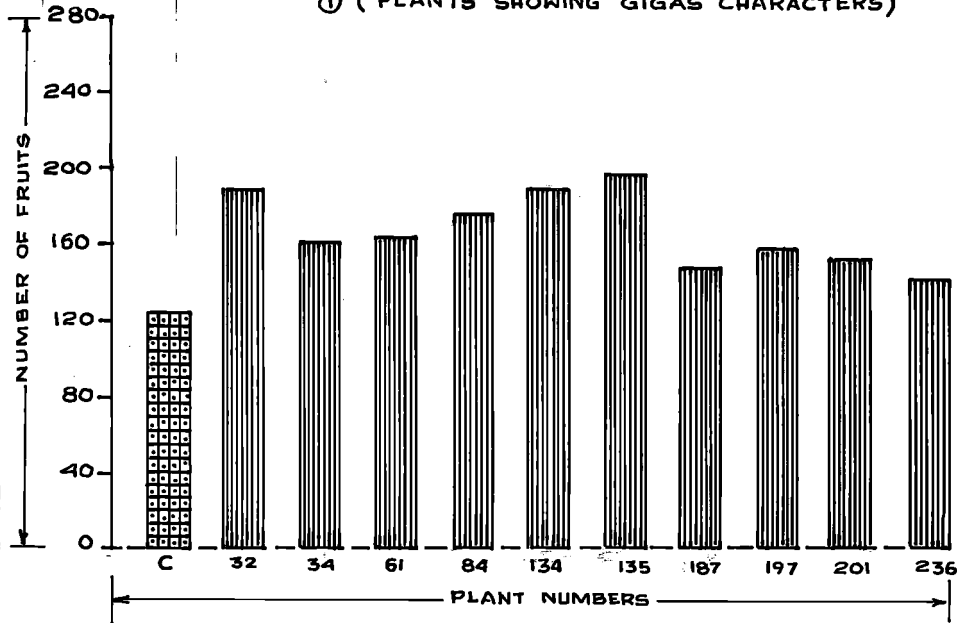


② (PLANTS SHOWING ABNORMALITIES)

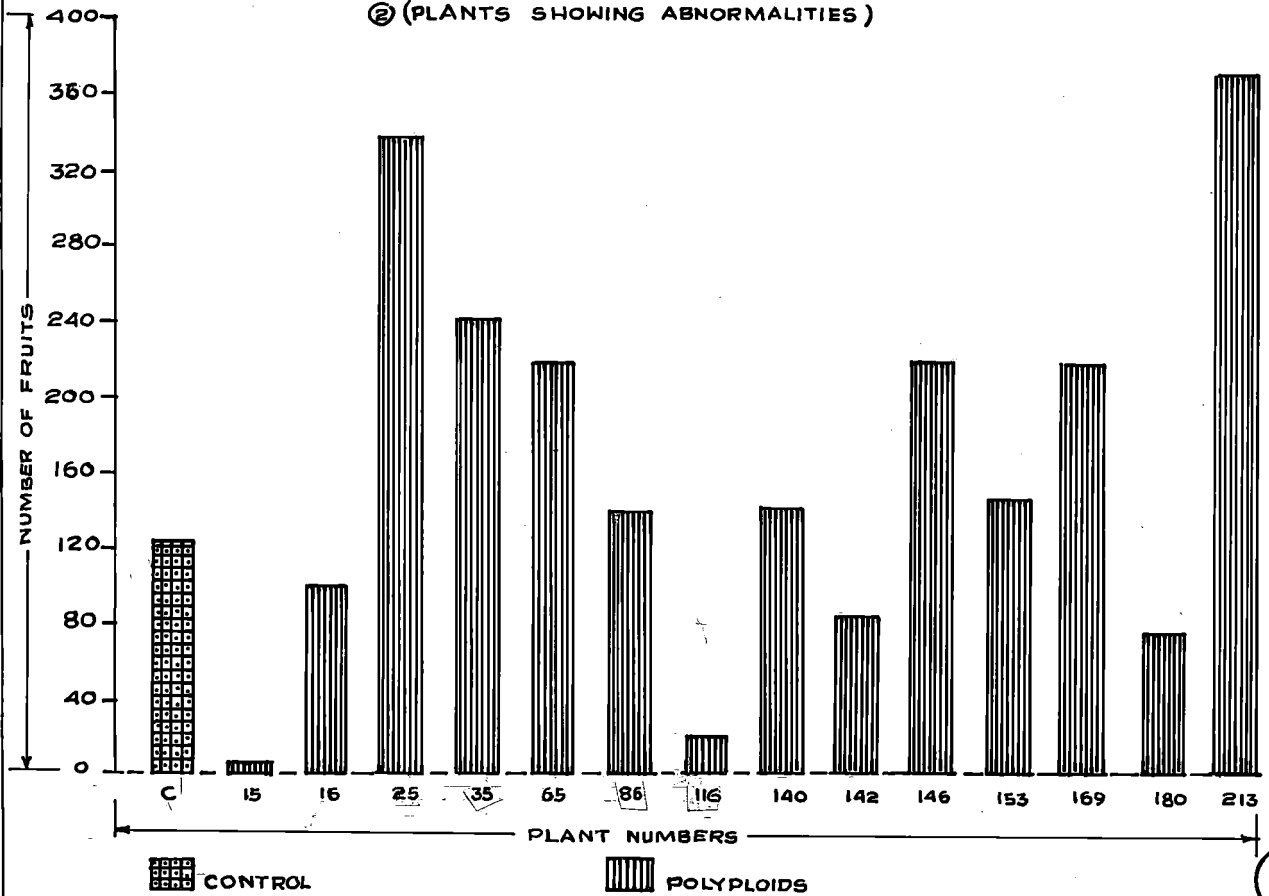


**VARIATIONS IN THE NUMBER OF FRUITS
PRODUCED BY THE POLYPLOIDS**

① (PLANTS SHOWING GIGAS CHARACTERS)



② (PLANTS SHOWING ABNORMALITIES)

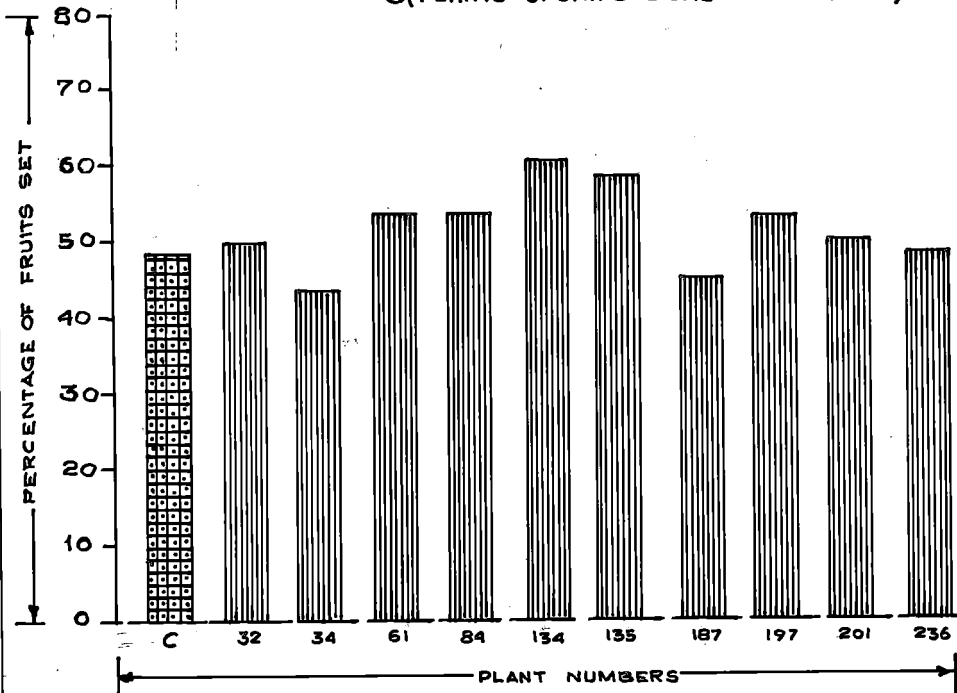


CONTROL

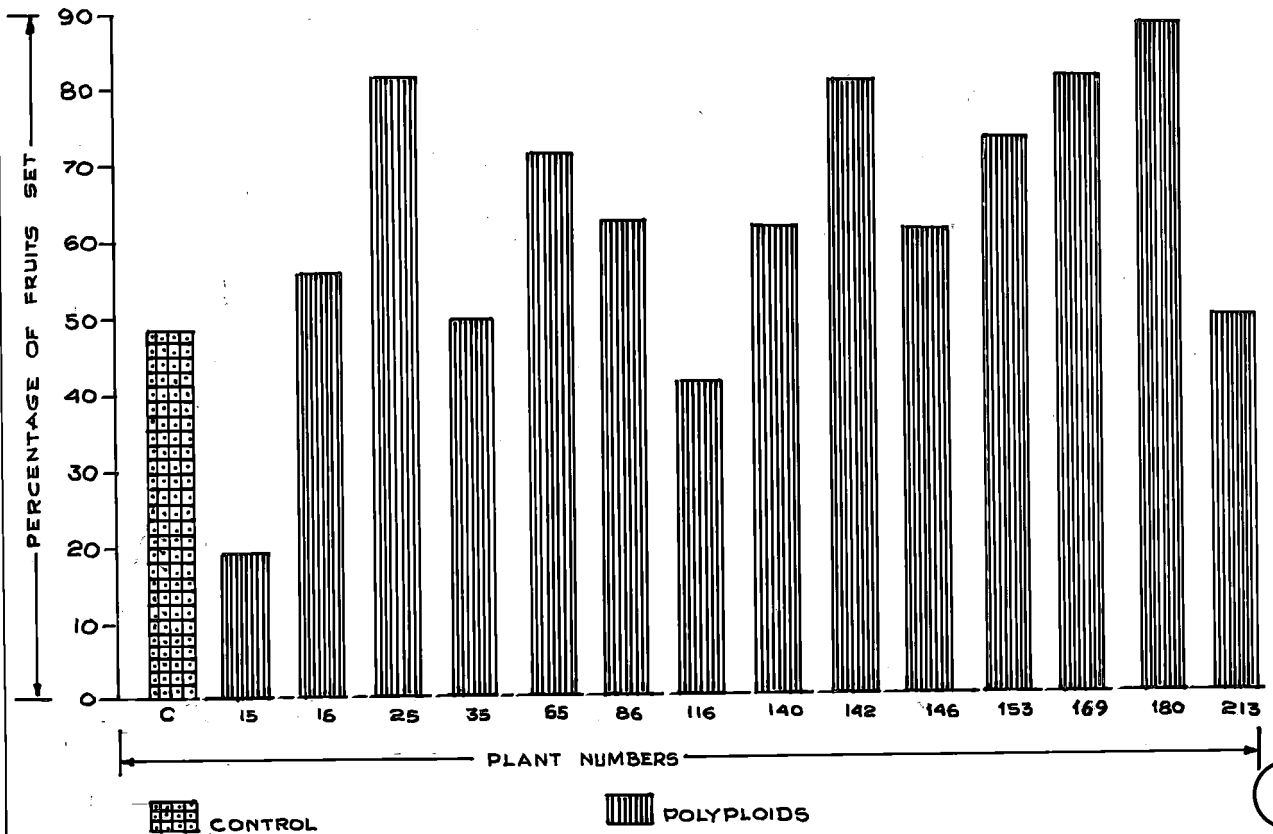
POLYPLOIDS

VARIATIONS IN THE PERCENTAGE OF FRUITS SET AMONG POLYPLOIDS

① (PLANTS SHOWING GIGAS CHARACTERS)



② (PLANTS SHOWING ABNORMALITIES)

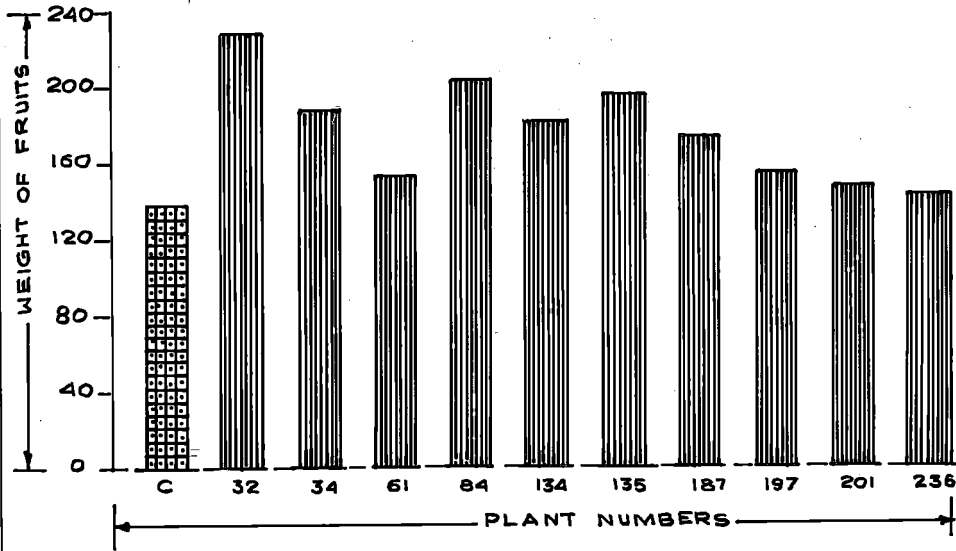


CONTROL

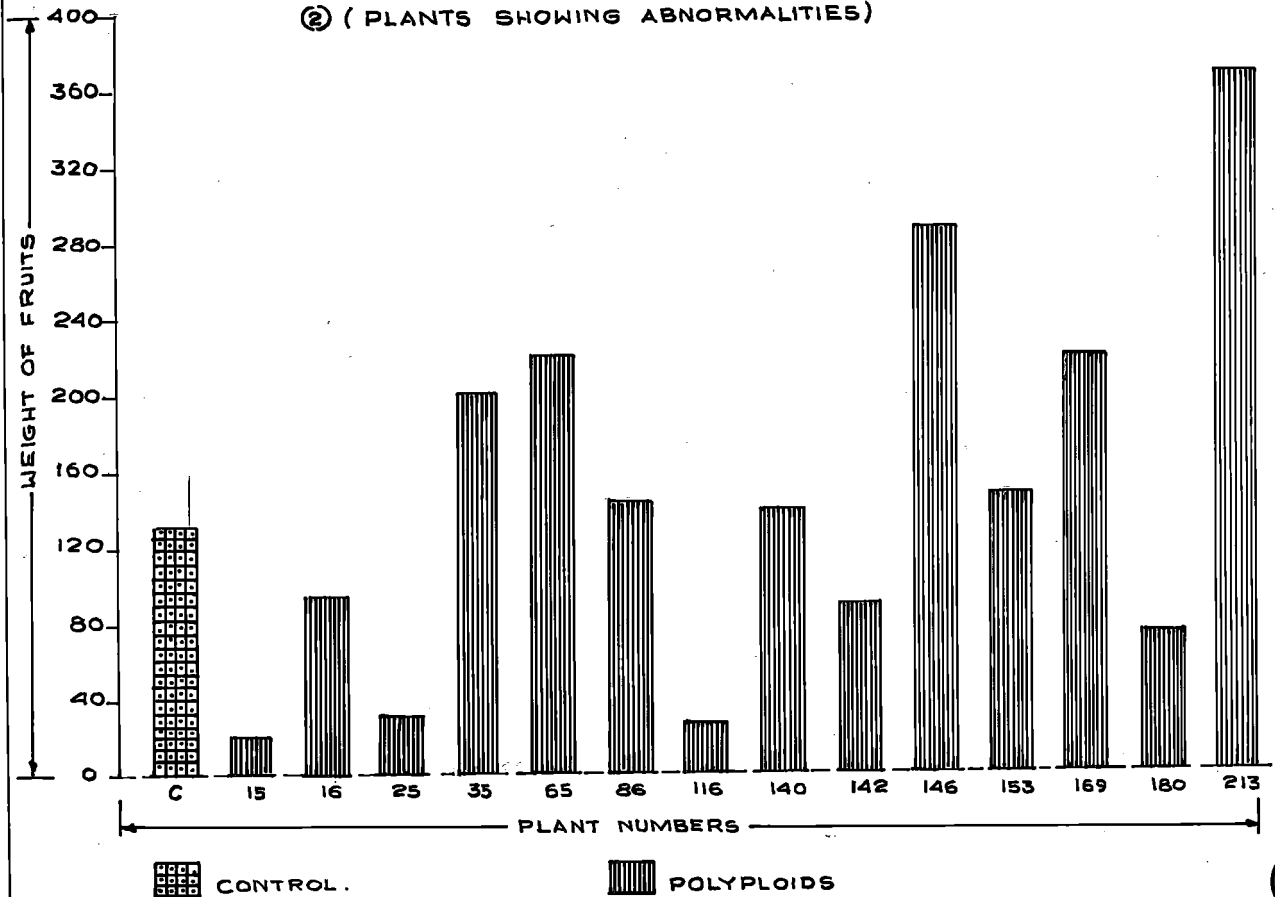
POLYPLOIDS

VARIATIONS IN YIELD AMONG POLYPLOIDS

① (PLANTS SHOWING GIGAS CHARACTERS)



② (PLANTS SHOWING ABNORMALITIES)

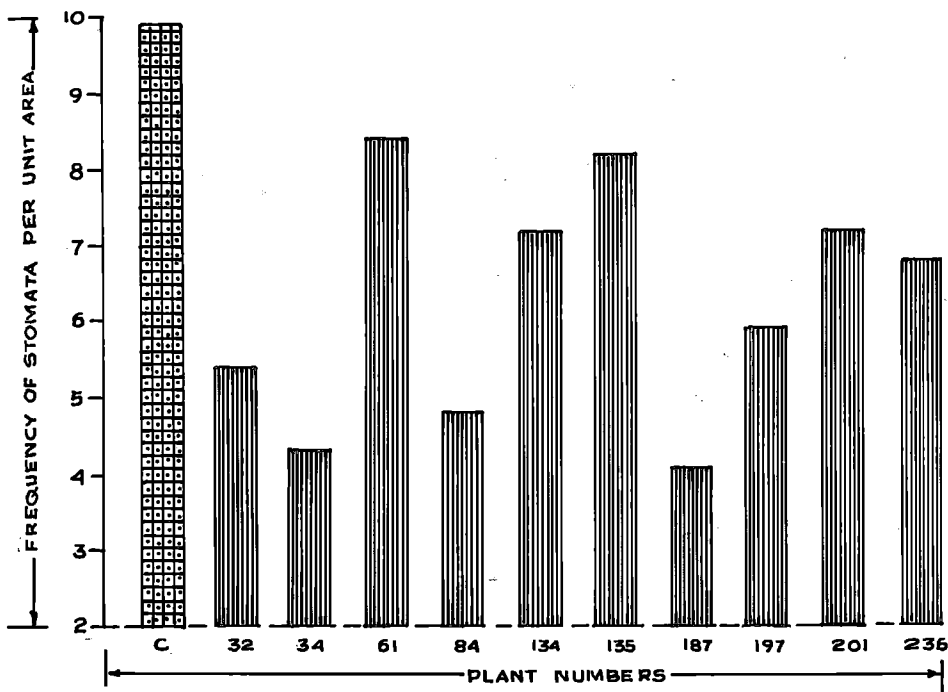


CONTROL.

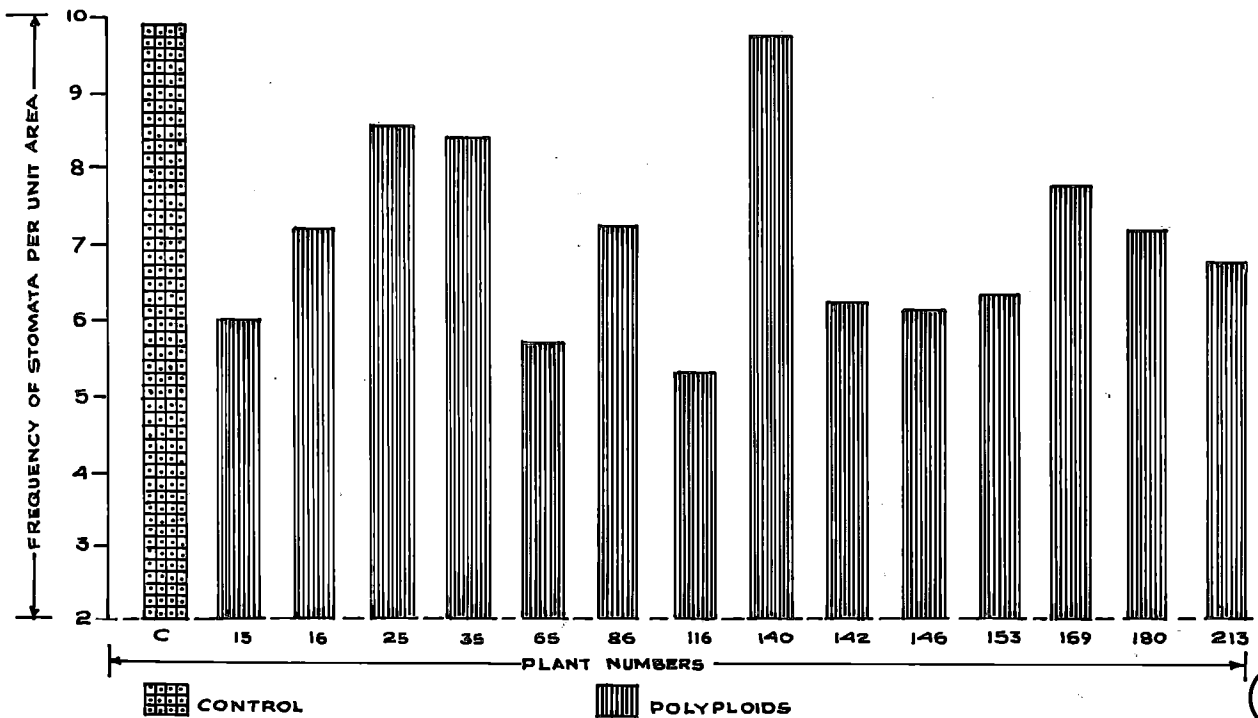
POLYPLOIDS

VARIATIONS IN THE DISTRIBUTION OF STOMATA AMONG POLYPLOIDS

① (PLANTS SHOWING GIGAS CHARACTERS)



② (PLANTS SHOWING ABNORMALITIES)



DISCUSSION

DISCUSSION

This study was mainly confined to the general morphological features of the C_2 population and the detailed cytological analysis of the suspected polyploids. The plants in the C_2 generation exhibited wide morphological variations which resulted in the heterogenous nature of the population.

The percentage of germination of seeds obtained from the C_1 plants ranged from 20 to 100 per cent. In the diploids 90.06 per cent seed germination was recorded. A higher percentage of seed germination was seen in many of the C_2 lines when compared with that of the diploids. Such a high germinability of tetraploid seeds have been reported in sesamum (Kobayashi and Shimamura, 1942, 1949; Srivastava, 1956) in corchorus (Kundu and Sharma, 1946) and in black gram (Sen and Cheda, 1958). However, there were lines with very low percentage of germination. Pal et al (1941) recorded reduced germination percentage in polyploid chillies. The seeds, in most of the polyploid types began germinating from the 5th day of soaking, as in the diploids. A delay in germination was noticed in certain lines which suggests that more number of

polyploids may be occurring in those lines. The rate of germination of polyploids was comparable with that of diploid seeds.

The data on the height, spread and number of branches (Table 2) showed that the plants in the C_2 lines in general possessed greater growth and vigour. This increased vigour of plants might have been contributed by the occurrence of more number of polyploids in the C_2 generation. Rao et al (1944, 1945) found in tetraploid jute and Srivastava (1956) in Sesamum orientale L. that the C_2 plants were characterised by the gigas nature. On the other hand Zimmerman (1958) reported that the tetraploids of forage grasses were less vigorous. Sen and Cheda (1958) also reported the less vigorous nature of the C_2 black gram. With respect to height and spread of plants, there were lines which showed increased and decreased growth, more and less number of branches, than the diploids. The number of branches is of importance in chillies where fruit is the economic product. But the increase or decrease in the number of branches may not be found in several cases proportionate to the yield. This can be explained as due to the low fruit set noticed in many cases.

Ramanujam and Joshi (1941) reported a reduction in height and lesser number of branches in the C_2 plants in Cicer arietinum L., while Mehta, Subramanyam and Swaminathan (1963) observed vigorous plant growth and more number of branches in the C_2 population of tetraploid berseem. Mettin (1965) observed that the height of plants in the C_2 population was decreased in Aegilops speltoides. Ghodgaonkar, Deshpande and Bechar (1965, 1966, 1967) showed that in chillies the C_2 plants were gigantic having increased height. Ahloowalia (1967) reported reduced tiller number in rye grass.

A delay in flowering by three to five days was noticed in two of the C_2 lines compared to the diploids. Randolph (1944) in maize, Tandon and Chinoy (1950) in Amaranthus bilitum, Srivastava (1956) in sesame, Spasojevic (1956), Kumar and Abraham (1942) in Phaseolus all observed delayed flowering in the tetraploids. In respect of the number of flowers produced by the different lines in the C_2 generation, there were plants with higher as well as lower flower production. High flower production in polyploids have been reported by Ramanujam and Joshi (1941) in Cicer arietinum.

Kluge (1957) found less number of flowers in Fragaria vesca. In the lines which showed significantly higher flower production (Table 3) it cannot be concluded from the evidence at hand whether it was due to the occurrence of more number of polyploids in these lines or due to other factors. Although a wide variation existed in the flower production, the fruit set in the G_2 population on the whole was lower than that in the diploids. One possible explanation is the poor fruit set noticed in most of the polyploids in the G_2 generation. The occurrence of different levels of polyploidy along with diploids might have also contributed towards this difference in the fruit set. Low fruit set in polyploid plants have been reported by several workers.

Parthasarathy and Kedaranath (1945) and Kobayashi and Shimamura (1945) reported lower fruit setting in the polyploid sesamum, while Kovacs-schneider (1959) found no difference in fruit setting from the diploids in tomatoes. As the percentage of fruit set was low a corresponding decrease in the yield was also noticed. Moffett and Nixon (1960) reported similar findings in Acacia. A lower fruit set is not always the case in polyploids. Nishiyama (1950) found that the tetraploid radish gave higher yield than the diploids. Necessarily

the contributory factor for yield in radish is the roots. The C_2 lines surpassed the corresponding diploids with respect to the weight of seeds. High seed weight in polyploids have been reported in Fragaria vesca (Kluge, 1959) and in Sesamum indicum (Langham, 1952).

The morphological characters of polyploids which were cytologically confirmed, showed considerable variation (Table 10, 11) from the diploids in characters like height, spread, number of branches, number of flowers, number of fruits, percentage of fruit set, yield etc. Such variations have been reported by Parthasarathy and Kedarnath (1945) in Sesamum (increased height and more number of branches and less number of capsules) Bhattacharjee (1956) in Cajanus cajan (lower number of branches), Ramanujam and Joshi (1941) in Cicer arietinum (large number of pods) and Mehta et al (1966) in Trifolium foenum-graecum (lower seed set). The polyploids produced smaller fruits when compared to the diploids. Sen and Gheda (1958) obtained shorter pods in the polyploids of black gram. Singh (1955) also reported reduced fruit size in Carica papaya. Production of smaller fruits in tetraploid chillies was also recorded by Aleksic (1961).

The polyploid plants exhibited a slower growth rate in the initial stages and a rapid growth rate later (Fig. 2). This observation is in agreement with the earlier findings of Janaki Ammal and Bezbarruch (1962). Retardation of initial growth on polyploids were also observed by Amin (1940) in cotton.

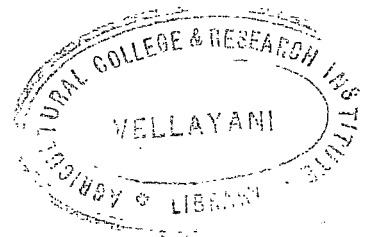
Apart from the gigas characters exhibited by the polyploid plants there were extreme abnormalities noticed in several cases in the C_2 generation. Such plants were also separately studied and cytologically analysed. It was interesting to note that plants exhibiting extreme abnormalities happened to be polyploids. This means that like increased vigour, polyploidy can also find expression as abnormalities in chillies. The abnormalities noted were initial stunted growth, crinkling of leaves and darker pigmentation of the foliage. Stebbins (1950) had reported abnormalities such as dwarfing and wrinkled foliage associated with polyploidy. Gopalakrishnan and Shastry (1964) also distinguished tetraploids of Oryza australiensis on the basis of their dark green foliage.

Polyploidy is usually accompanied by gigas characters expressed as increased vigour, larger leaf

area, larger pollen and stomatal size. Indiramma (1966) reported that the polyploid chillies exhibited increased plant height, increased leaf area, stomatal size and pollen size. She had fixed these characters for the polyploid plants.

In the present study, therefore, an attempt was made to isolate polyploids based on the above morphological characters as it was not possible to study all the plants individually by cytological methods. Based on the confidence limits fixed for the morphological characters for the diploids 17 plants were isolated and cytological studies showed that 14 plants thus isolated were polyploids. Three were found to be diploids. Ramanujam and Joshi (1941) also observed the occurrence of diploids in the C_2 population of tetraploid gram (Cicer arietinum). This confirms the earlier findings that in chillies polyploidy is exhibited by increased vigour in morphological characters like plant height and vigour. In polyploidy breeding in chillies, therefore, the above morphological characters thus helps to select the polyploids from a large population.

Such methods have been adopted by Ramanujam and Joshi in selecting polyploids of Cicer arietinum in the



C_2 generation. Pal, Ramanujam and Joshi (1941) also found that the polyploids of chillies in the C_2 generation were characterised by larger stomata, bigger pollen grains, increased thickness of leaves etc. In the present study also increased stomatal and pollen size were recorded in polyploids. The same observation was made by Georgieva (1961) in the tetraploids of Capsicum. Increased stomatal size in the polyploids were noticed by Kobayashi and Shimamura (1945) in sesamum. Armstrong and Robertson (1960) found that tetraploid alsike clovers surpassed the diploids in characters like pollen diameter, stoma size etc. Luongdinhcua (1950) noticed increased size of pollen grains in the polyploid rice seedlings. High percentage of pollen sterility was noticed in the case of all the confirmed polyploids. Pal, Ramanujam and Joshi also reported 30-90 per cent pollen sterility in tetraploid chillies. Similar observation was reported by Kundu and Sharma (1956) in the tetraploids of Corchorus olitorius.

The polyploids in general showed wide differences in features such as slower growth rate, later time of blooming and less fertility than the diploid progenitors and an overall increase in size of the various organs as

observed by various earlier workers. The cytology of polyploids also is a matter of wide variation. Many abnormalities were met with in the meiotic division of these polyploids which may be the cause for low seed set in certain cases. In the first meiotic division the diakinesis was normal in most cases, although quadrivalents were occasionally met with. Pal, Ramenujam and Joshi (1941) found 0-7 quadrivalents and the rest bivalents. Quadrivalent formation was also noticed to occur in diakinesis by Richharia and Persal (1940), Kobayashi and Shimamura (1949, 1952) in sesamum and Ramson and Levan (1939) in sugarbeet. But the frequency of quadrivalents observed in this case was much less than that was reported by the above authors. Sen and Cheda (1958) reported few quadrivalents in black gram. Chromatin bridge and laggard formation were also observed. Ramenujam and Joshi (1941) observed one chromatin bridge at anaphase I. Occurrence of laggards have been reported by Laws (1961) in the tetraploids of *Oenothera* and Islam (1960) in *Annona squamosa*. As many as nine laggards were observed in some cells. Three, five and six telophasic groups of chromosomes were also observed instead of normal tetrads. Koul (1964) reported three unequal but independent functional groups at anaphase II. Such a deviation from

the tetrad formation may probably due to the formation of laggards at anaphase and the unequal disjunction of the quadrivalents formed at diakinesis. This may also be due to the abnormalities in the formation of spindle which may be multipolar in nature.

The many abnormalities found in the meiotic division of the polyploids can be attributed to high pollen sterility and low percentage of fruit set observed in many of the polyploids. The cytological studies of 27 plants isolated from the C_2 population based on morphological characters and abnormalities showed that 24 were tetraploids and three were diploids. This gives us enough reason to think that the C_2 generation may contain larger number of diploids. It seems therefore possible that the C_1 tetraploid was really a mixoploid. Such an occurrence of C_1 tetraploids which were later confirmed to be mixoploids, based on the progeny performance, was described by Ramanujan and Joshi (1941) in Cicer arietinum.

SUMMARY

S U M M A R Y

The present study was undertaken in the Division of Agricultural Botany, Agricultural College and Research Institute, Vellayani with a view to study the morphological variations that existed in the C_2 generation of the colchicine induced tetraploid chillies. Detailed morphological studies of the plants in C_2 generation and cytological analysis of the suspected polyploids were conducted.

The seeds from the colchicine induced polyploid chillies were carried over to the C_2 generation. From the cytological observations made on selected individuals in the C_2 population, it was revealed that there occurred 24 tetraploids and three diploids, leading to the conclusion that the C_1 tetraploid was possibly a mixoploid.

Some of the individuals showed stunted growth in early stages. But most were found to be gigas in size. A general increase in the vigour of morphological characters such as height of plants, spread of plants, number of branches and yield of fruits were found in all cases of polyploids. However, abnormalities like dwarfing,

crinkling and dark foliar pigmentation were also noticed with certain polyploids. This may be the persisting effect of colchicine in inducing deformities in the G_2 generation also.

Based on the morphological variations, plant abnormalities and stomatal and pollen characters, 27 plants were isolated as suspected polyploids. A detailed study on the cytology of these plants was conducted and out of the 27 suspected polyploids, 24 were found to have 48 chromosomes and these were confirmed to be tetraploids. All the morphological characters were studied for these 24 polyploids and it was found that in general they exceeded the diploids in the over all vigour of the plants. These plants were also characterised by an increased pollen sterility and larger but fewer number of stomata. In general seed setting and yield were found to be low in the tetraploids. The possible role of polyploidy breeding in chillies has been discussed.

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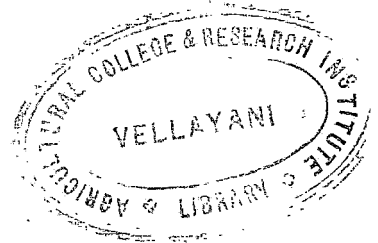
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* Original not seen

APPENDIX I

Analysis of variance for the height of plants

Source	S.S.	D.F.	Variance	F ratio
Total	137253.98	159	863.23	
Treatment	109281.55	10	10928.16	58.21**
Error	27972.43	149	187.73	

** Significant at 1% and 5% levels

APPENDIX II

Analysis of variance for spread of plants

Source	S.S.	D.F.	Variance	F ratio
Total	56503.56	155	364.53	
Treatment	12377.86	10	1237.79	4.06**
Error	44125.70	145	304.31	

** Significant at 1% and 5% levels

APPENDIX III

Analysis of variance for number of branches

Source	S.S.	D.F.	Variance	F ratio
Total	220449.34	151	1459.93	
Treatment	71376.50	10	7137.65	6.7**
Error	149072.84	141	1057.25	

** Significant at 1% and 5% levels

APPENDIX IV

Analysis of variance for duration of flowering

Source	S.S.	D.F.	Variance	F ratio
Total	543.87	30	18.13	
Treatment	261.87	10	26.19	1.8
Error	282.00	20	14.10	

F ratio is not significant

APPENDIX V

Analysis of variance for number of flowers

Source	S.S.	D.F.	Variance	F ratio
Total	2244005.97	159	14125.82	
Treatment	390340.26	10	39034.03	
Error	185665.71	149	12454.13	3.13*

F ratio is significant at 5% level

APPENDIX VI

Analysis of variance for percentage of fruit set

Source	S.S.	D.F.	Variance	F ratio
Total	5773.68	30	192.46	
Treatment	32.88	10	3.29	
Error	5740.80	20	287.04	0.67

F ratio is not significant

APPENDIX VII

Analysis of variance for number of fruits

Source	S.S.	D.F.	Variance	F ratio
Total	19481100.00	144	135285.42	
Treatment	53197.91	10	5319.79	0.93
Error	19427902.09	134	144984.34	

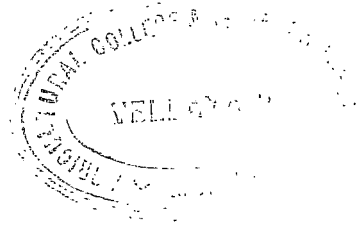
F ratio is not significant

APPENDIX VIII

Analysis of variance for yield of fruits

Source	S.S.	D.F.	Variance	F ratio
Total	19273350.80	144	133842.60	
Treatment	265763.00	10	26576.30	0.91
Error	19007587.80	134	141847.67	

F ratio is not significant



APPENDIX IX

Analysis of variance for area of leaves

Source	S.S.	D.F.	Variance	F ratio
Total	107.44	30	35.81	
Treatment	5.30	10	0.53	
Error	102.14	20	5.04	0.11

F ratio is not significant

APPENDIX X

Analysis of variance for size of pollen grains

Source	S.S.	D.F.	Variance	F ratio
Total	92.20	29	3.2	
Treatment	1.95	9	0.22	
Error	90.25	20	4.51	0.05

F ratio is not significant

APPENDIX XI

Analysis of variance for distribution of stomata

Source	S.S.	D.F.	Variance	F ratio
Total	174.80	30	5.83	
Treatment	127.69	10	12.77	5.41*
Error	47.11	20	2.36	

* F ratio is significant at 5% level

APPENDIX XII

Analysis of variance for 1000 seed weight

Source	S.S.	D.F.	Variance	F ratio
Total	100.75	29	3.47	
Treatment	40.98	9	4.55	1.52
Error	59.77	20	2.99	

F ratio is not significant

APPENDIX XIII

χ^2 table for pollen sterility

Treatments	χ^2 value
Line 1 Vs Control	0.03
Line 2 Vs Control	0.15
Line 3 Vs Control	0.008
Line 4 Vs Control	0.43
Line 5 Vs Control	0.097
Line 6 Vs Control	0.008
Line 7 Vs Control	0.0001
Line 8 Vs Control	0.02
Line 9 Vs Control	0.17
Line 10 Vs Control	0.017

Critical value for χ^2_1 (0.05) = 3.841

None of the values are significant

APPENDIX XIV

Characters	χ^2 value
Height of plants	$\chi^2 = 32.8$
Spread of plants	$\chi^2 = 52.6$
Number of flowers	$\chi^2 = 75.2$
Number of fruits	$\chi^2 = 175.01$
Weight of fruits	$\chi^2 = 147.8$

Critical value of χ^2 (0.05) = 16.92

None of the χ^2 values are significant.

LIST OF ILLUSTRATIONS

Figures

- 1 Percentage of germination in the C_2 lines
- 2 a Rate of growth of polyploids (Plants showing gigas characters).
- 2 b Rate of growth of polyploids (Plants showing abnormalities)
- 3 Variations in height among polyploids
- 4 Variations in spread among polyploids
- 5 Variations in number of branches among polyploids
- 6 Variations in the time taken for flowering in the polyploids
- 7 Variations in the number of flowers produced by the polyploids
- 8 Variations in the number of fruits produced by the polyploids
- 9 Variations in the percentage of fruits set among the polyploids.
- 10 Variations in yield among polyploids
- 11 Variations in pollen sterility among polyploids
- 12 Variations in the distribution of stomata among polyploids.

Plates

- 1 Polyploid plant showing height
- 2 Polyploid plant showing vigour
- 3 Comparison in spread between polyploid and diploid

Plates

- 4 Polyploid plant showing the spread
- 5 Leaf section of the polyploid plant
- 6 Size and distribution of stomata in diploids
Size and distribution of stomata in polyploids
- 7 Comparison of size of fruits between polyploids
(P) and control (C)
- 8 Comparison between size of ^{seeds} fruits in polyploids
(P) and Control (C)
- 9 Diakinesis in polyploid showing 24 bivalents
- 10 Late telophase II in polyploid showing normal
tetrad
- 11 Anaphase I in polyploid showing 24 chromosomes
at each pole.
- 12 Anaphase I in polyploid showing one chromatin
bridge in one cell and laggards in the
other cells.
- 13 Late telophase II in polyploid showing three
groups of chromosomes
- 14 Late telophase in polyploid showing 5 groups of
chromosomes
- 15 Late anaphase I in polyploid showing 4 laggards
- 16 Diakinesis in polyploid showing multivalent
formation
- 17 Late anaphase II in polyploid showing 6 groups
of chromosomes and laggards
- 18 Metaphase I in polyploid
- 19 Anaphase I in polyploid showing 8 laggards
- 20 Diakinesis in diploid showing 12 bivalents.

PLATE I

Polyploid plant showing height

PLATE II

Polyploid plant showing the vigour



PLATE III

Comparison in spread between polyploid
and diploid

(R₂ T₃ 86 - Polyploid
C - Control)

PLATE IV

Polyploid plant showing the spread

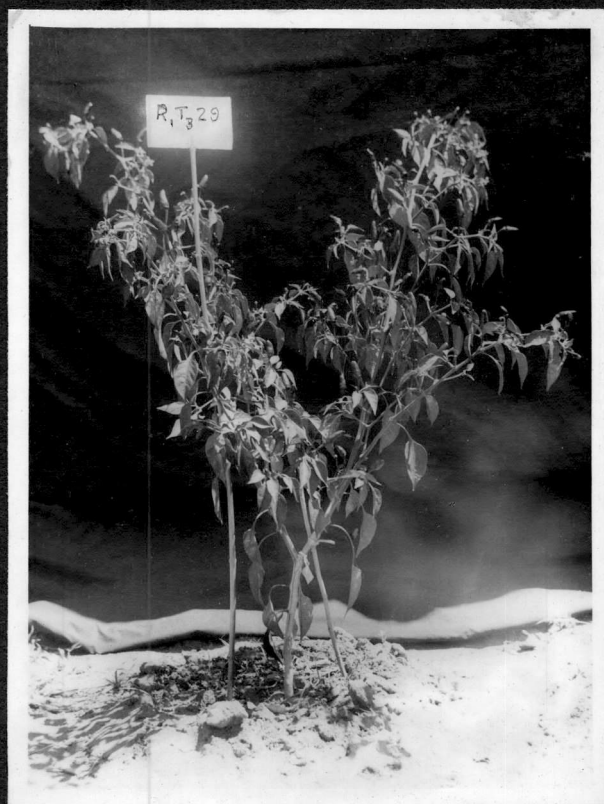


PLATE V

Leaf section of the polyploid plant.

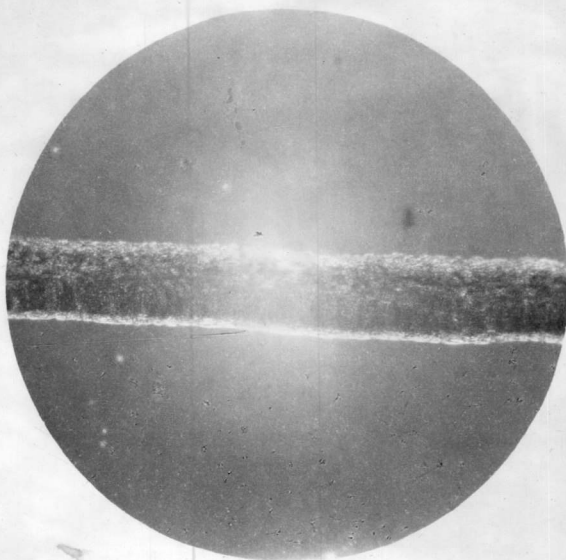


PLATE VI

Size and distribution of stomata in diploids

Size and distribution of stomata in polyploid.

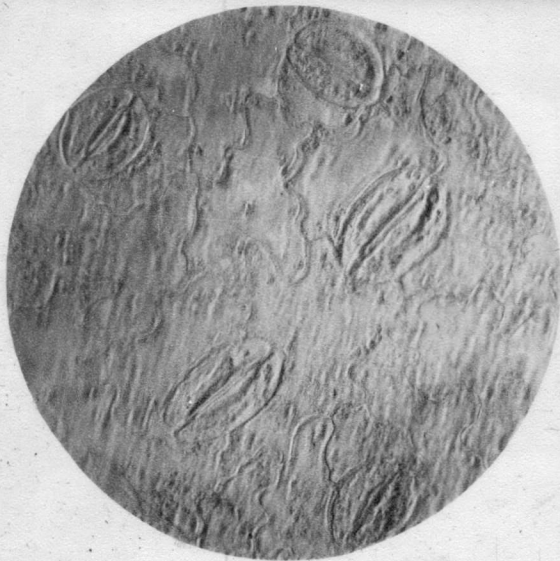
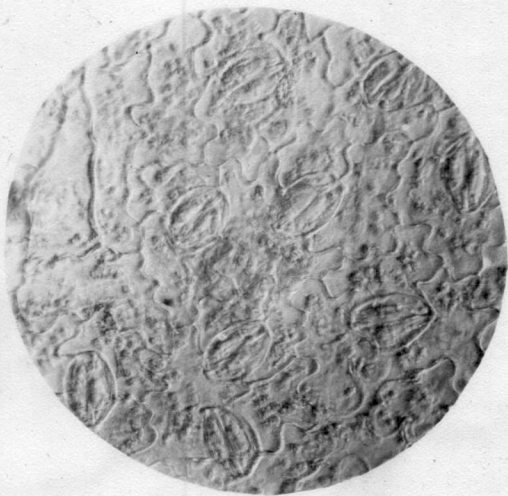


PLATE VII

**Comparison of size of fruits between
polyploids (P) and Control (C)**

PLATE VIII

**Comparison of size of seeds between
polyploids (P) and Control (C)**

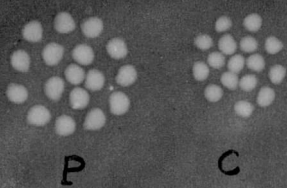
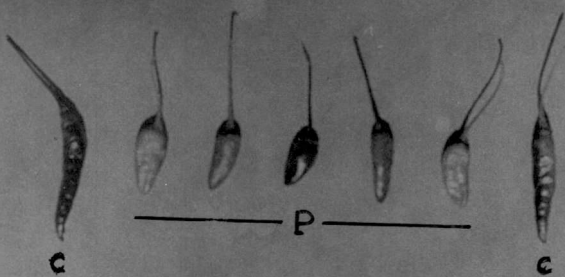


PLATE IX

Diakinesis in polyploid showing 24
bivalents

PLATE X

Late telophase II in polyploid showing
normal tetrad

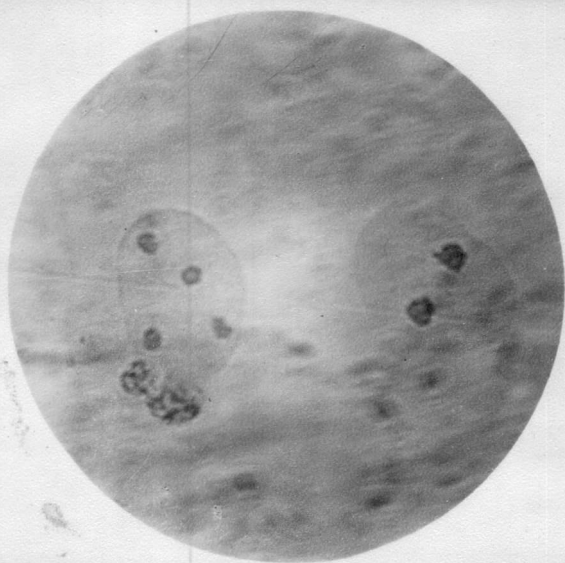
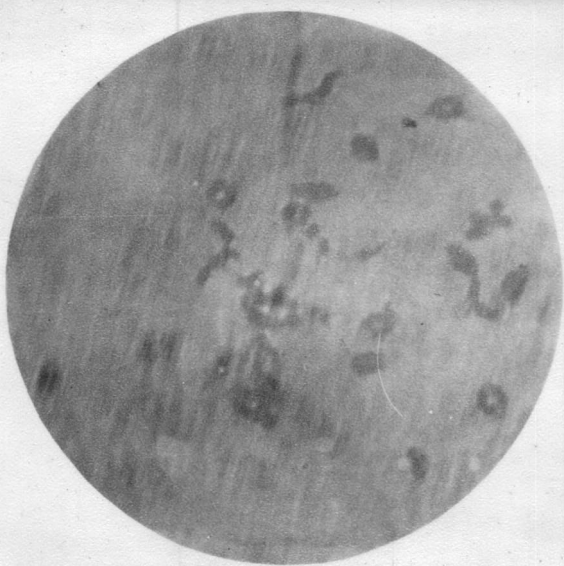


PLATE XI

**Anaphase I in polyploid showing 24
chromosomes at each pole**

PLATE XII

**Anaphase I in polyploid showing one chromatin
bridge in one cell and laggards in
the other cells**

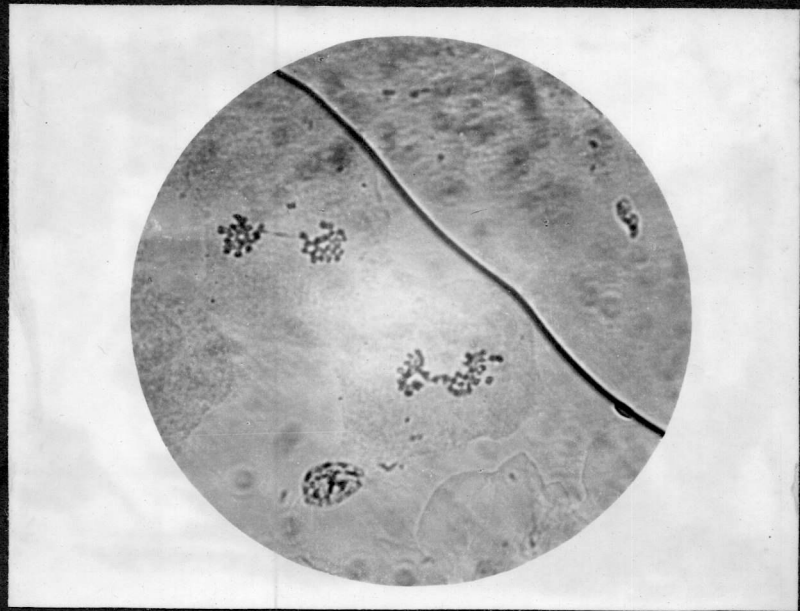
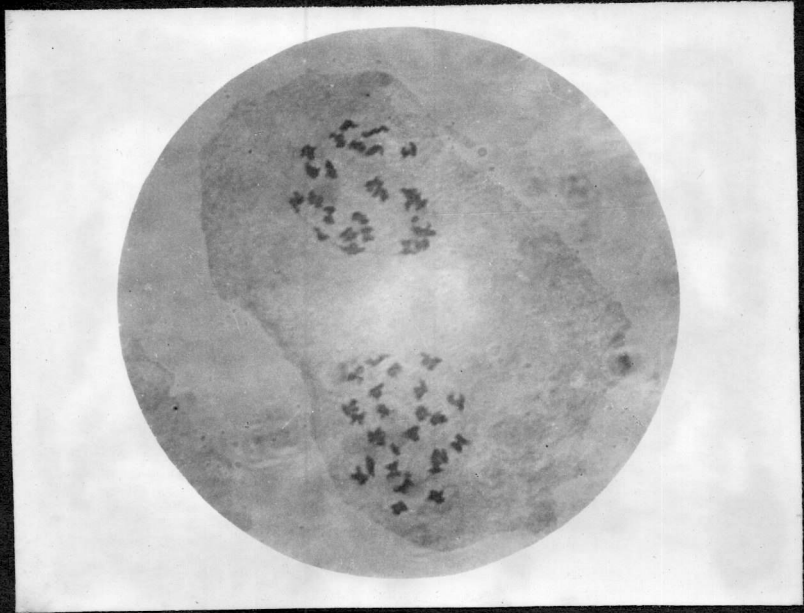


PLATE XIII

Late telophase II in polyploid showing
three groups of chromosomes

PLATE XIV

Late telophase in polyploid showing
five groups of chromosomes

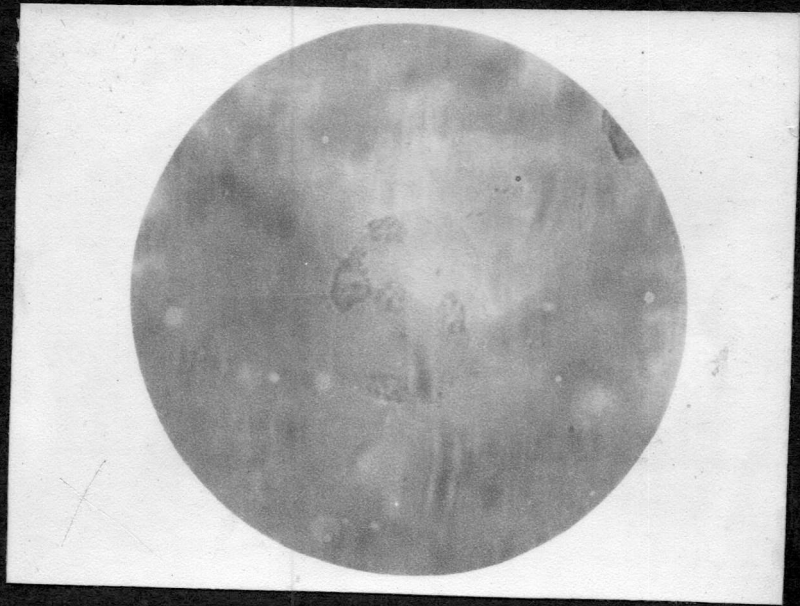
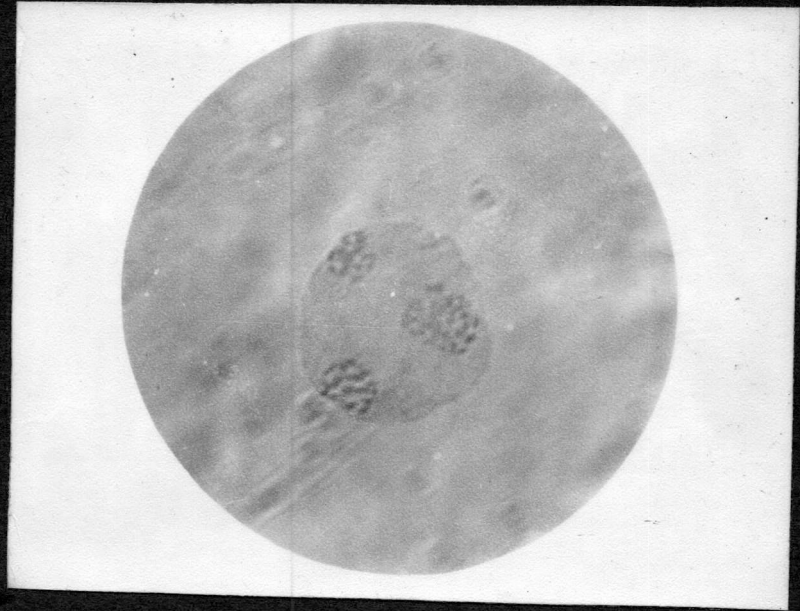


PLATE XV

Late anaphase I in polyploid showing
4 laggards

PLATE XVI

Diakinesis in polyploid showing multivalent
formation

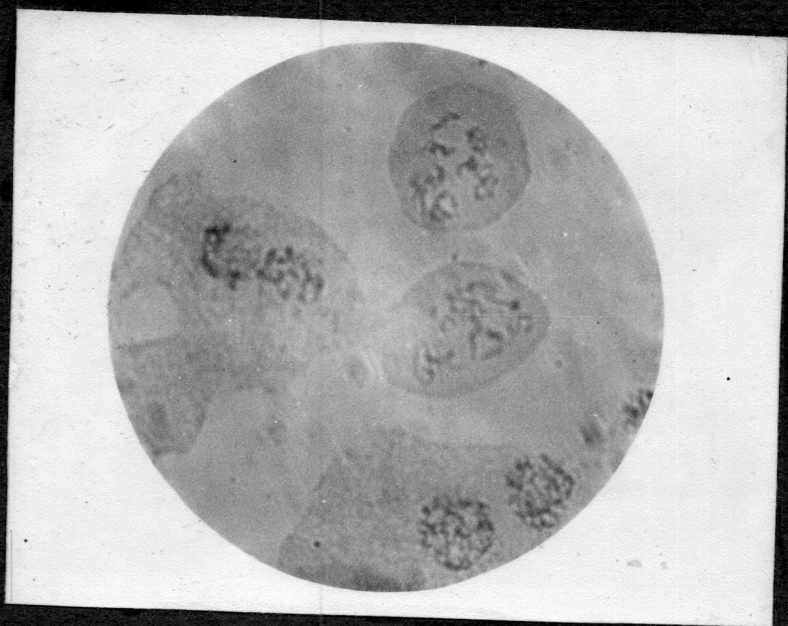
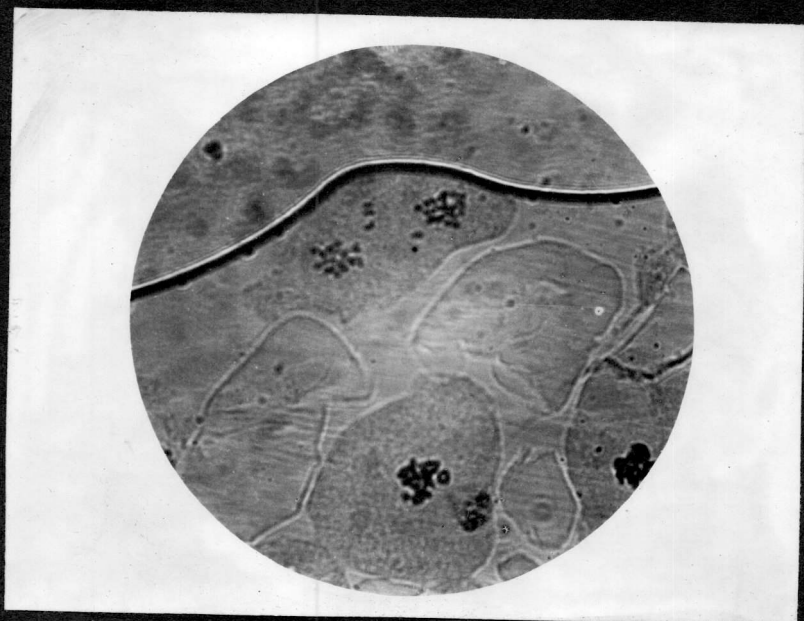


PLATE XVII

**Late anaphase II in polyploid showing 6 groups
of chromosomes and laggards**

PLATE XVIII

Metaphase I in polyploid

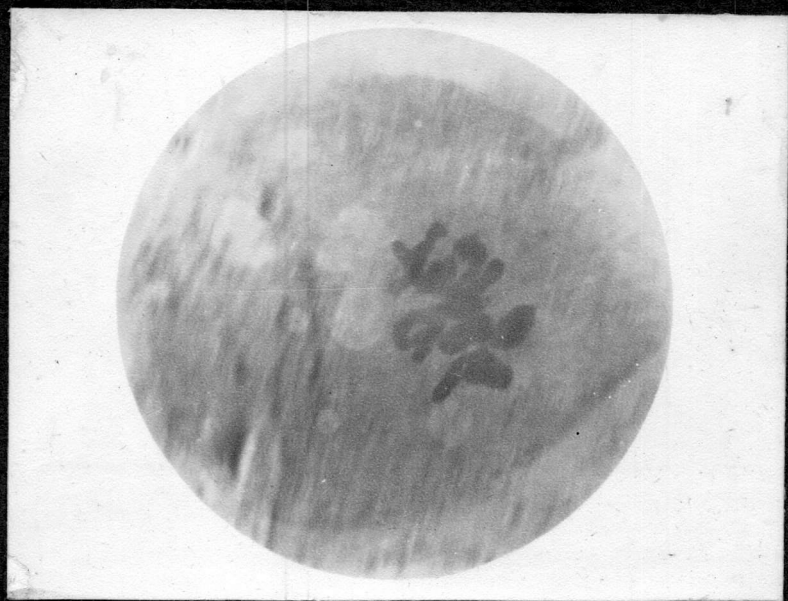
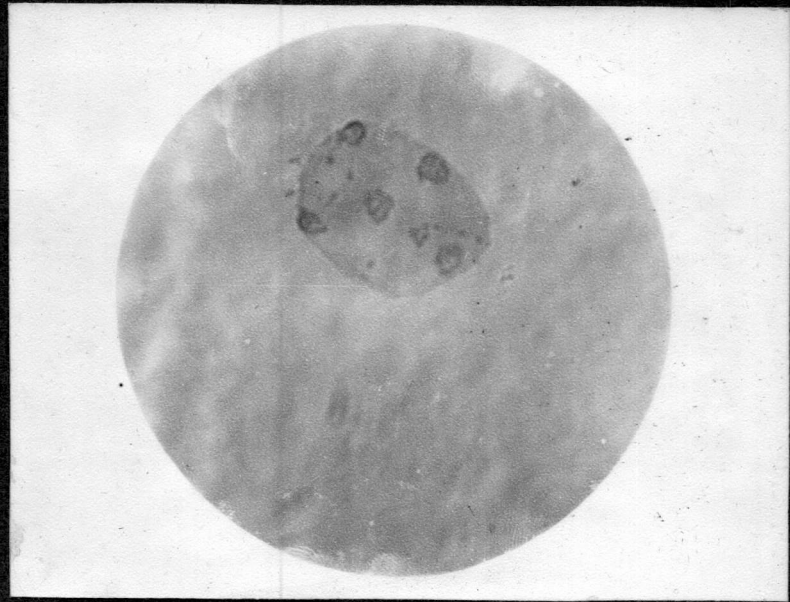


PLATE XIX

Anaphase I in polyploid showing 8 laggards

PLATE XX

Diakinesis in diploid showing 12 bivalents

