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A STUDY ON HAPLOID PLANTS IN EGYPTIAN COTTON

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1 - INTRODUCTION

Cotton haploids have been found as small twin seedlings by Harland in 1920, in Sea Island cotton or as solitary field plants known by the natives of Trinidad as "Man cotton". He also recorded in (1936) two cases of haploidy in Upland cotton (G. hirsutum L.) which have been found in the cultivar Acala Okra, first case was observed in field culture and the second as one member of a pair of twins, He stated also that neither twin embryos nor haploids have been found in Egyptian cotton (G. barbadense L.) variety Emans Brown and showed that other Egyptian varieties had not been thoroughly examined. It was also noticed by Beasley (1941) and Harland (1955) that occurrence of polyembryonic seeds was common in the New World cottons. However, the frequency of twins differed between species of Gossypium. Haploid/diploid twins have been reported to be the most common type from which the haploids could be recovered. The doubling of the chromosome number in these haploids using colchicine would produce theoretically homozygous pure lines of cotton that are not only useful in breeding but also utilized in physiological experiments, as reported by Harland (1959). Harland (1955) discussed that the use of the doubled haploids in genetic and breeding programs for several important characters could be recorded.

In the haploids, with the assurance that the corresponding diploid would also possess them to a similar degree. Resistance to disease, lint length, colour and fineness and seed fuzz are among the characters desirable to study.

Some Egyptian cotton seeds Giza 67 and Memufie each of which gave two embryos were separated, at the Seed Testing Laboratory, Ministry of Agriculture at Giza. These seeds were sown in pots and produced three types of twin seedlings having diploid/diploid or diploid/haploid or haploid/haploid members. The latter type usually dies out at an early stage of development, while the haploid partner of the diploid twin can survive to maturity. The haploid cotton is easily distinguished from normal diploid by its thin zigzag stem, short internodes, small leaves and squares and small sterile flowers. The haploid seedlings were treated with colchicine to produce double haploid seeds from each of the cultivars Giza 67 and Memufie in the seasons 1967 and 1968. Double haploid were compared with their nucleus control cultivars for their yield component characters and their fiber quality characteristics. Furthermore, double haploid pure lines originating from self pollinated seeds of the double haploid plants were grown in the next season and compared

with their control cultivars for seed weight, Ginning out turn, Seed index, Holo length, fiber strength and fiber fineness and other fiber quality characters.

Field trial experiments were carried out at Giza and Sakha Experimental Stations in the season 1970, including the double haploid pure lines and their nucleus seed cultivars as controls.

The aim of this work is :

- 1) To realise cytologically the haploid number of chromosomes in the weak member of the twin seedlings in Egyptian cotton.
- 2) To produce their double haploids which are theoretically homozygous.
- 3) To evaluate their agronomic characteristics as compared with their original commercial cultivars.

II- REVIEW OF LITERATURE

a) Polyembryony and the occurrence of haploids

The occurrence of more than one embryo in a seed "the phenomenon of polyembryony" has attracted much attention ever since its initial discovery in oranges by Leeuwenhoek (1719) (c.f. Maheshwari, 1950).

1- Polyembryony and haploids

Lehrman (1925) stated a case of polyembryony in cotton.

Harland (1936) described the haploid state in Sea Island cotton. He tested several thousand seeds of Sea Island cotton and noticed twenty seeds containing two embryos, one of the two was usually smaller than the second. By careful culture, Harland was able to raise to maturity sixteen pairs of plants. Fourteen of them each pair consisted of one haploid and one diploid and the two rest pairs each of them were of identical diploids.

Webber (1940) stated that the twins were mainly haploid-diploid in the tetraploid species ($n = 26$) and

smaller number was haploid and the larger was dioid.

Roux (1958) reported that it was probable that a tendency to polyembryony was an inherited character.

Roux and Chirikian (1959) noted that although haploidy in cotton was previously considered exceptional, research work at Bebedjia in 1958 - 1959 resulted in the isolation of more than 50 haploid plants from population of certain varieties of Upland cotton. The precise origin of haploid was as yet not clear but it was suggested as induced by abnormalities in fertilisation.

Turcotte and Roaster (1963) reported that progenies on a double haploid line, L 56-4 of G. barbadense Pima S-1, contained a high proportion of haploid plants. These plants, in contrast to those previously reported in cotton originated from single rather than twin-embryo seeds, probably by haplo parthenogenesis.

Blank and Allison (1963) showed that the incidence of polyembryony was observed in a number of varieties and strains of Upland cotton (G. hirsutum L.), most of which have an Acala back ground.

Owings et al. (1964) mentioned that in the Sea Island cotton Z 101 and its hybrids with five marker lines, most of the twins consisted of a haploid and an amphidiploid, the haploid having small stomata, leaves, flowers and small pollen grains. They reported that the most likely supposition for the development of the polyembryonic haploids was the formation of an extra embryo from a cell of the embryo sac other than the egg-cell. Such haploid would probably develop from an unfertilized synergid, since the polar nuclei are needed for endosperm development and since the antipodals possibly degenerate prior to maturation of the egg apparatus. He mentioned other two possibilities for the occurrence of the haploids first that two embryo-sacs within a single ovule; the second a 16-nucleate embryo-sac with only one egg cell being fertilised. The most logical explanation for the moncembryonic haploids would be development without fertilization of a cell of the egg apparatus.

Tiranti (1965) reported that examination of a haploid plant, which was dwarfed, had small leaves and flowers and short branches with numerous ramifications, showed that it was sterile and that seeds set by

pollinating with pollen from normal plants aborted several weeks later.

Murcotte and Reaster (1969) in their studies on semigametic production of haploids in Pima cotton reported that semigamy could be employed to produce haploids at will. The original semigametic line had normal characteristics thus limiting haploid production via semigamy to marked stocks. The fact that semigamy was transmitted through the egg and pollen permitted the development of semigametic, marked stocks. The use of a semigametic, marked stock, as female could allow the production of haploids and their identification within any cotton strain.

2- The frequency of polyembryony and haploidy

Beasley and Richmond (1941) reported that in a population grown from seeds produced by pollinating normal flowers of Rogers Acala with X-irradiated pollen, one haploid was found. If this haploid was the result of X-irradiating pollen, the frequency of the haploids is too low to be of commercial value.

Bessly (1941) cited that haploid-diploid twins such as frequently occurred in G. barbadense L. had been found though extremely rarely in G. hirsutum L.

Roux (1958) reported that amongst 44,000 seeds of various Sea Island cotton examined at Stoneville, 141 pairs of twin embryos were found, of these, 112 appeared to consist of one diploid and one haploid. The percentage of haploids varied between 0.1 and 1.6 % amongst these varieties. In G. hirsutum L. cotton, the average frequency of twin embryos was one pair in 20,000 to 25,000 seeds but the proportions of diploids and haploids are not known.

Blank and Allison (1963) in examining 330,130 seedlings from a number of varieties and strains of Upland cotton, G. hirsutum L., 20 twin embryos were found (an over-all frequency of 1 : 16,506). The twin embryos were brought to flower and the seed increased in field plantings. Of major importance is the finding of a line yielding 23 twin embryos in 60,698 seedlings from the first increase seed. This line had a ratio of 1 : 2,639, a highly significant increase in the incidence of polyembryony.

Huette and Easter (1963) reported that the progenies of a doubled haploid line, D 57-4 of *G. perhadenza* Pina 3 (9) contained a high proportion of haploid plants. These plants, in contrast to those previously reported in cotton originated from single rather than twin-embryo seeds. The frequency of haploids was greater in greenhouse than in field plantings, probably because the environmental conditions during emergence and early growth were more hazardous for haploid than for diploid seedlings.

Owings et al. (1964) in their studies on Sea Island cotton 2 101 and its hybrids with five marker lines reported that the incidence of haploids (7.7 per 1000 emerged seedlings) closely paralleled that of twinning (9.3 pairs per 1000 germinated seeds).

Cytological identification of the haploids

Cytological studies were carried out on the haploid plants in cotton by several workers and the Florida review showed the most important work in this aspect.

Webber (1938) in his studies in a haploid-diploid pair which was obtained in G. barbadense found that the diploid members had regular meiosis except for occasional quadrivalents and univalents while the haploid had 26 I in 22 cells, 1 II + 24 I in one cell and 2 II + 22 I in two cells.

Beasley (1941) found that the haploids in G. hirsutum had a maximum of 5 bivalents. In (1942) he reported that the meiotic chromosome behaviour was regular in diploid species of Gossypium, but there were few irregularities in tetraploid species. The investigator also reported that in a haploid of tetraploid species a maximum of five pairs of chromosomes were found.

Endrizzi (1959) in his studies on the haploid of G. hirsutum found an average of 0.16 bivalents per cell. It was evident that most bivalents consisted of pairing between (A) and (D) chromosomes. One cell, however, was found with all 26 univalents oriented on the equatorial plane.

Sastry and Swaminathan (1960) in their studies on the haploid G. barbadense reported that in each of

the 100 P₁C's which he examined in a single haploid ($2n = 24$), the chromosomes were all univalents. A single microsporocyte had 26 II. Nondisjunction during premeiotic mitosis could have led to the formation of such a cell. At diakinesis and metaphase they observed specific secondary associations of the end to - end, side - to side and end to side types. The associations noted involved 1 large and 1 small univalent, 2 large and 1 small, 3 large and 1 small or 3 large univalents. The occurrence of secondary associations between large and small chromosomes was interpreted as being indicative of homology between a few of the (A) and (D) genome chromosomes, while the secondary association of large chromosomes only was regarded as providing support for the view that chromosomes belonging to genome (A) had undergone a higher degree of duplication than those of (D) as suggested by the observation of Kammacher et al. (1957).

Tiranti (1965) in his study on chromosome association and some characters of a haploid plant in D. hirsutum found an average of 0.128 II and 25.74 I as scored over 372 counts. He reported that in the haploid there was no evidence of intergenomic affinity since the number of bivalents was very low.