

II. REPORTS FROM COOPERATORS

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1. Further studies on the occurrence of twin seedlings in *Coix aquatica*.

The occurrence of twin seedlings in *Coix aquatica* is believed to be the result of two ovaries, consequently two seeds, developing within the same spathe (MNL 43:4-6, 1969). Further observations on this aspect are made and reported here.

325 spathes each possessing multiple stigmas instead of a normal bifid stigma were found in 1968 plantings. On closer scrutiny, in 148 of these, multiple branches of the stigma emerged from a single style and a random sample of 25 spathes on breaking open have been found to contain only one well developed ovary. The rest of the 177 spathes had two separate bifid styles and on closer examination, 25 random spathes of this group possessed two well developed ovaries, each of which in turn had a style of its own. Since the spathes with two bifid styles are potentially able to form two seeds at maturity, the remaining 152 were pollinated and bagged. 108 of these turned hard and black (the appearance of normal healthy spathes possessing seed) while the rest were chalky white and soft and usually sterile (the result of failure of fertilization in both the ovules). Of the 108 well developed spathes, 50 were broken open. 18 of these had two seeds while 32 had only one seed in each (possibly due to failure of fertilization in one or the other of the two ovules). Of the remaining 58 spathes, when sown in June, 1969, 46 germinated and among these 10 produced double seedlings (in one case triple seedlings). 5 of the spathes giving rise to multiple seedlings were broken open carefully and in all cases except one each of the seedlings originated from a separate seed. In the exceptional spathe, both seedlings arose from the same seed. The occurrence of this exception and of triple seedlings from a spathe where only two seeds are possible is a clear indication of polyembryony. A cytological check of the 21 plants that arose as double seedlings revealed a diploid chromosome number in all.

In an embryological study of spathes with multiple as well as single styles, some interesting observations were recorded in a low proportion of cases. In the ovule the nucellus is partitioned vertically into two halves by a wall reaching well up to the funicle; in one half there is a well developed embryo sac and in the other there is a suggestion of an embryo sac developing. If the two embryo sacs give rise to two embryos and they in turn to two seedlings, then both the seedlings would come from the same seed. The occurrence of several four nucleate unreduced embryo sacs in each ovule (MNL 40:164, 1966) of the pistillate inflorescences produced towards the end of the season also provides an opportunity for the occurrence of more than one embryo in a seed. Thus it is borne out by the embryological study that cases where twin seedlings arise from the same seed may be due to the occurrence of two embryo sacs in an ovule rather than to one arising from the fertilized egg and the other from a haploid nucleus of the same embryo sac. The diploid nature of the twins as observed affords further proof for this assumption. When two embryo sacs occur in an ovule, whether both are sexual, one sexual and one unreduced, or both unreduced is, however, not known. Further studies are underway to determine the origin and nature of multiple embryo sacs occurring in an ovule. It appears that a search for monoploids by screening of twin seedlings is not of much help in Coix.

The frequency of double seeds in a random lot of spathes is quite low (0.0045%, MNL 43:4-6, 1969). However, the frequency is much higher when spathes with multiple styles were chosen (more than 15%). Therefore, it appears that only such spathes and ovules with twin embryo sacs contribute to double and triple seedlings in the population. Although spathes with multiple styles are less frequent in the general population, particular lines show a greater frequency of such spathes. For example, all the 325 spathes in the present study came from seven plants of the same line although not all spathes on any one plant are multistyled. Of the 21 plants that arose as double seedlings only 8 produced spathes with multiple styles of varying frequencies. Whether the multistyled (double seeded) condition is a mutant character with incomplete penetrance and

variable expression is not known. The genetic nature of multistyly is under study.

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2. Similar mutants in corn and Coix

Some of the segregants located in experimental populations of Coix lacryma-jobi and C. aquatica have a close resemblance to some of the well described mutants in corn, although the nature of their inheritance in Coix is yet to be studied. They are listed in the table below.

Mutants in <u>Coix</u>	Similar to mutants in corn
<u>Coix lacryma-jobi</u>	
1. ' <u>Virescent</u> ' - seedling yellow, turns green slowly.	Virescent $\frac{v_2, v_4}{\text{and } v_{16}}$
2. ' <u>Luteus</u> ' - lethal yellow seedling, seedling dies at 2 or 3 leaf stage.	Luteus $\frac{1}{2}$
3. ' <u>Knotted leaf</u> ' - seedling leaves show a knotted appearance, plant leaves normal.	Knotted <u>Kn</u>
4. ' <u>Crinkled leaf</u> ' - plant more or less short, leaves wrinkled.	Crinkled leaf <u>cr</u>₁
5. ' <u>Adherent</u> ' - first seedling leaves stick together, plant leaves normal.	Adherent <u>ad</u>₁
6. ' <u>Gold stripe</u> ' - yellowish longitudinal stripes, often broad, on margins and blades of leaf throughout the life of the plant.	Old gold stripe <u>Og</u>
7. ' <u>Pygmy</u> ' - leaves short, broad and pointed, plant more or less short.	Pygmy <u>py</u>
8. ' <u>Styleless</u> ' - styles not produced, ovules abort, young spathes wrinkled.	Silkless <u>sk</u>
<u>C. aquatica</u>	
9. ' <u>Luteus</u> ' seedling and plant yellowish green.	Luteus $\frac{1}{7}$
10. ' <u>Yellow stripe</u> ' - leaves with yellow tissue between leaf veins.	Yellow stripe <u>ys</u>₁