Cyto-morphological Studies on Male Sterility in *Brassica campestris* L.

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Male sterile mutants were observed in two varieties of *Sarson* (*Brassica campestris* L.). In brown *sarson* it was a case of functional male sterility where the anthers were normal looking with viable and healthy pollen grains but there was no dehiscence. The anthers of male sterile plants in yellow *sarson* were reduced in size and contained non-viable pollen. Studies on the mode of inheritance revealed that in each case male sterility was recessive to normal condition and showed a monogenic type of control (Chowdhury and Das 1966). The present work was taken up to determine the causes leading to either failure of normal dehiscence or degeneration of pollen grains in the male sterile plants. The results obtained are discussed in this paper.

Materials and methods

To find out if any meiotic abnormality during pollen development is the cause of male sterility, anther smears were prepared in aceto-carmin from male-sterile as well as normal plants. A few good slides were made permanent by the method suggested by Swaminathan, Magoon and Mehra (1954).

Studies were made on the development of tapetum in both the cases to determine if there is any difference between the two types. In the case of non-dehiscent anthers which contained otherwise viable pollen grains, a microscopic study of the anther wall was also taken up.

Results and discussion

Cytology. Male sterility has been found in several cases to be associated with abnormality at some stage of meiosis in the process of formation of pollen grains. The degeneration may take place just in pollen mother cells before the actual division has started (Fabergé 1937, Rick 1944) while in other cases it may be during prophase I (Childers 1952, Bernardo 1957). In yet other cases breakdown may be at metaphase (Fabergé 1937, Dodds and Simmonds 1946) or anaphase (Valmyor, Mendoza and Millare 1956, Bosemark 1957). There are other types of abnormalities reported which cause degeneration of tetrads formed after normal meiosis. There may be prolonged coherence of the four cells of a quartet until they finally abort (Rhoades 1934, Rick 1948), failure to form exine (Frankel 1940) and degeneration of pollen nuclei (Nakamura 1943, Fukasawa 1956, and Ustinova 1961).

In the present material a comparative study of meiosis in the male-sterile

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and normal plants in yellow *sarson* showed no difference. There were 10 bivalents present at metaphase (Fig. 1) and regular separation of chromosomes was seen at anaphase I (Fig. 2) which led to the formation of normal tetrads (Fig. 3). It was after the formation of tetrads that further development was inhibited and spores were seen as crumpled mass of thin walled cells with very little cytoplasm which took either very light or no stain at all (Fig. 4).

Development of tapetal layer. Changes in the development of tapetal layer cause the degeneration of tetrads (Childers 1952, Childers and McLennan 1960, and Pandey 1961). However, it is also true that in some cases no

Figs. 1-4. Meiosis in male-sterile plants of *Brassica campestris* L. var. yellow *sarson* 1, metaphase I showing 10 II. 2, anaphase I showing regular separation of chromosomes. 3, normal tetrads. 4, degenerated pollen grains.

irregularity is found in male-sterile plants (Soost 1951, Fukasawa 1956, and Pandey 1961). A study on the development of tapetum in yellow *sarson* was rewarding. A conspicuous difference was observed between the tapetal cells of normal and male sterile plants in later stages of microsporogenesis. There was normal development of tapetum in both the types till post-meiotic stages. But then the tapetal cells in the normal plants started degenerating and finally disappeared (Fig. 5). On other hand, in male-sterile plants this layer showed no sign of degeneration even after the disappearance of pollen

grains (Fig. 6). Similar observations have been reported in *Cucurbita maxima* (Singh and Rhodes 1961) and *Sorghum* (Singh and Hadley 1961).

Degeneration of microspore in the present material may be due to the

Figs. 5-9. T. S. of anthers from normal and male-sterile plants. 5, T. S. of anthers from normal plants of *B. campestris* var. yellow *sarson* showing degeneration of tapetal layer. 6, T. S. of anthers from male-sterile plants of *B. campestris* showing intact tapetal layer even after the degeneration of pollen grains. 7, T. S. of anther from male-sterile plants of *B. campestris* var. brown *sarson* (non-dehiscent type). 8, enlarged portion of the anther wall from normal plants of *B. campestris* var. brown *sarson* showing regular, wall developed cells. 9, enlarged portion of the anther wall from male-sterile plants of *B. campestris* var. brown *sarson* (non-dehiscent type) showing thick and irregular cells in the form of a compact layer.

tapetal layer not functioning as a nutritive layer as has been reported by other workers also (Artschwager 1947, Singh and Hadley 1961 and Singh and

Rhodes 1961). Other reasons have been suggested for the degeneration of microspores like growth of hypertrophied and swollen cells of tapetal layer (Artschwager 1947, Childers 1952 and Pandey 1961) or sudden release of metabolic wastes (Artschwager 1947). No such observation could be made in the present case.

Studies on the structure of anther wall. Non-dehiscence of anthers have been reported in various crops such as grapes (Olmo 1943), *Trifolium repens* (Atwood 1944), maize (Sprague 1939), tomato (Roever 1948, Larson 1949), brinjal (Jasmin 1954) etc. The failure in the normal dehiscences may be due to hard anther wall, insufficient pressure within the anther or absence of anther pore.

In the present case of non-dehiscence in brown *sarson* a microscopic study of the structure of anther wall was undertaken. There was no difference observed between the anther walls of the two types till the formation of tetrads. The wall in each case consisted of two layered epidermis and a dark stained single layer of endothecium. Degenerating tapetal cells could be seen in both the cases. But in the later stages, the cells in anther wall of non-dehiscent type enlarged and appeared as a thick, compact layer (Figs. 7 and 9), while no marked change was observed in the wall of normal anthers. The cells were regular, oval shaped and thin walled (Fig. 8). In the non-dehiscent type the anther wall remained intact even after 2-3 days of flowers opening with clear differentiation of cells till the anthers dried up. The present study, therefore, indicates that anthers failed to dehisce due to the presence of hard and compact anther wall.

Summary

Cyto-morphological studies were conducted on male sterility in yellow and brown *sarson*. In the post-meiotic stages, the tapetum did not seem to function as a nutritive layer in yellow *sarson* as it persisted long after degeneration of tetrads. The non-dehiscence of anthers in brown *sarson* appeared to be due to the presence of large, irregular cells of anther wall in the form of a thick, compact layer. Meiotic studies on these two types of male sterility did not show any abnormality

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