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# Megasporocyte formation in *Pisum sativum* L. against the background of bud development

## WANDA WOJCIECHOWSKA\*, TERESA MACKIEWICZ \*\*

\* Laboratory of Interspecific Hybrids, Institute of Plant Genetics, Polish Academy of Sciences, ul. Strzeszyńska 30/36 60-479 Poznań, Poland

\*\* Institute of Genetics and Plant Breeding, Academy of Agriculture, ul. Wojska Polskiego 71c, 60-625 Poznań, Poland

#### Abstract

Megasporocyte formation in *Pisum sativum* L. takes place at the stages of bud development, when the vexillum surpasses the lower androecium whorl slightly at the beginning, and markedly in further stages. Under the epidermis of an ovule, a multicellular archesporium is formed. Mitotic division of these cells gives rise to a two-layered parietal tissue and to a group of megaspore mother cells (MMCs). The tetrad is formed from one megasporocyte. In the bud development of *Pisum sativum* the carpel walls accrete to each other relatively early.

## INTRODUCTION

Megaspore formation in *Pisum sativum* was described by R e m b e r t (1969). In the present paper an attempt is made to analyse megasporocyte formation with reference to simultaneous events proceeding in a developing bud.

## MATERIAL AND METHODS

Buds of *Pisum sativum* L., cv. Folger were examined using the parrafin microtome technique. The preparations were stained with iron hematoxylin and fast green.

## RESULTS AND DISCUSSION

At a very early stage of bud development when the highest corolla petal — the vexillum — is located below the lower androecium whorl, the pistil has no ovules and the stamens are at a very early stage of differentiation (Plate I, Photos 1-3). In a somewhat older bud, when the

## Pisum sativum L.

- Photo 1. Longitudinal section of bud with vexillum (arrow) below the lower androecium whorl;  $\times$  35.
- Photo. 2. Longitudinal section of the same bud as in Photo 1; there are yet no ovules in the pistil ovary; × 35.
- Photo 3. Longitudinal section of anther from the bud presented in Photos 1 and 2; the anther is differentiated into meristematic tissue surrounded by protoderm; × 440.
- Photo 4. Longitudinal section of bud with vexillum (arrow) slightly surpassing the lower androecium whorl; the ovary contains ovule primordia; X 35.
- Photo 5. Transverse section of bud at the same stage as the bud in Photo 4; the pistil walls are closely contiguous, but not accreted;  $\times$  100.
- Photo 6. Longitudinal anther section from the bud of the upper and roecium whorl presented in Photo 4; under the epidermis two layers of cells;  $\times$  440.
- Photo 7. Fragment of longitudinal section of bud with vexillum markedly surpassing the lower androecium whorl; telophase II in anthers, see Photo 9; in the ovules are megasporocytes, see Plate II, Photo 18; × 35.
- Photo 8. Transverse section of bud at a somewhat later stage than in Photo 7; the pistil walls are clearly accreted;  $\times$  100.

Photo 9. Longitudinal section of anther at telophase II with tapetum;  $\times$  440.

vexillum slightly surpasses the stamens of the lower whorl, small protuberances of ovules begin to appear in the ovary, and the anthers of the upper whorl have two layers of the anther wall under the epidermis. At that time the cross-section of the middle part of the bud shows that the pistil walls are already contiguous, i.e. conduplicate folding of the carpel has been completed (Plate I, Photos 4-6; Plate II, Photo 1).

At the stages of bud development, when the vexillum initially slightly, and at the further developmental stages markedly, surpasses the lower androecium whorl, a process of complete megasporocyte formation takes place. This begins with the appearance of hypodermal cells with enlarged nuclei and lasts until the time of advanced prophase I in a functional megasporocyte (Plate I, Photo 7). Plate I, Photo 8 shows a somewhat older bud with ovules having megasporocytes at the stage of metaphase I, the pistil walls of the middle part of the ovary are already accreted.

The process of megasporocyte formation in pea proceeds as follows: under the epidermis of an ovule, the hypodermal cells with large nuclei and dense cytoplasm are formed. According to R u t i s h a u s e r's (1969) terminology these are archespore mother cells (AMCs). Under the epidermis, these cells divide to produce parietal tissue and below a group of archespore cells (ACs). At the beginning of meiosis these cells become megaspore mother cells (MMCs), i.e. megasporocytes. Only the top megasporocyte and, therefore, the youngest one gives rise to a tetrad. The PLATE I





## Early megasporogenesis in pea

- Photo 10. Protuberances of ovules on placenta in the bud presented in Plate I, Photo 4. The upper ovule has double, and the lower a single layer of cells under the epidermis; × 440.
- Photos 11-12. Ovules with archespore mother cells (AMCs);  $\times$  440.
- Photos 13-15. Ovules with archesporial cells (ACs) and a single layer of subepidermal cells;  $\times$  440.
- Photos 16-17. Megaspore mother cells (MMCs). Prophase I is more advanced in the top MMC;  $\times$  440

Photo 18. Ovule with a single megasporocyte in late prophase I;  $\times$  440.

development of the remaining megasporocytes is inhibited at the beginning of prophase I (Plate II, Photos 10, 11).

It is difficult to give a clear description of megasporocyte formation in angiosperms because of the different conceptions of this process proposed in the literature (Warming, 1878; Schnarf, 1929; Rutishauser, 1969; Poddubnaya-Arnoldi, 1976) with different terminology used by authors to describe the same phenomena. The megasporocyte development in pea, here, may be reffered to Warming's (l.c.) type of dichlamyde or to Schnarf's (l.c.) developmental type I or II, as well as to developmental type I according to Poddubnaya-Arnoldi (l.c.). The occurence of megasporocytes in pea has already been described earlier (Roy, 1933; Cooper, 1938). Roy (l.c.) believed that AMC in pea, called by him an archesporial cell, functions as a megasporocyte without previous division. As it appears from our description his observations do not to be correct. The difference between Cooper's (l.c.) observations and ours is that in our material, as a rule, several AMCs were observed, while in Cooper's (l.c.) only one. The present description extends a previous one (Wojciechowska, 1978) in relation to the earliest stages of pea development. Mitchell (1975) described sporogenesis and early stages of bud development in Vicia sativa L. His results agree in many details with the course of bud development in Pisum sativum L. Howewer, there are differences, the most significant of which is a subhypodermal, not hypodermal, as in Pisum, formation of AMCs.

The accretion of the pistil walls in *Pisum sativum* is in clear contrast to the slow accretion rate of the ovary walls in *Trifolium repens* L. where the ovules begin their development before the completion of the conduplicate folding process in the ovary (R e m b e r t, 1977a). Since in self-pollinating *Glycine max*, and pea, the carpel conduplication takes place very early in floral development (R e m b e r t, 1977b), it may be suggested that this process is faster in self-pollinating plants and slower in cross-pollinating ones.

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