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Light and electron microscopic observations in connection with the developing pistil and seed-appendix (caruncle) of *Ricinus communis* L.

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Abstract

The development of the particularly organized stigma and obturator, their fine structure, their function as well as the histological differentiation and the role of the seed-appendix i.e. the caruncle of *Ricinus communis* L. have been investigated in several developmental phases from the so called "gynoecial primordium" state to the ripe state, using the terminology introduced by Sattler (1974). The stigma cells are characterized by dens cytoplasm, numerous vesicles mostly of ER origin, ribosomes and negatively stained mitochondria. Dilatation of ER, the appearance of electron opaque substances in it and between the plasmamembran and cell wall are frequent. The degenerating process of some stigma cells will start before the pollination because of autolysis. In the cells of the obturator and young caruncle however dictyosomes can be found more frequently than in stigma cells and the starch content of the plastids is remarkable. The thickening of the cell wall is connected with the function of these tissues.

INTRODUCTION

According to Heslop-Harrison and Shivanna (1977), *Euphorbiaceae* show the dry stigma type.

In connection with the organization of the pistil we wish to refer to the fact that the transmitting tissue is sometimes of heterogenous origin (Sattler, 1974). Under such circumstances the obturator of presumably placentic origin (regressing at seed development) is involved in guiding and growth of pollen tube.

At the time of pollination and fertilization considerable fine structural changes can be observed in stigma cells. (Kadej et al. 1977). Among the signs of the cell differentiation in *Ricinus* stigma the autolytical vacuolization too may be mentioned as in the seed-appendix cells of some monocotyledon plant (Komar, 1976) and in trichoblast (Harris, 1979).

Plate I

Fig. 1. Young stigma branches (SEM). × 600
Fig. 2. Ripe stigma with multiseriate papillae (SEM). × 200
Fig. 3. A detail of the "gynoecial primordium". × 9000
Abbreviations:
C — caruncle; CW — cell wall; D — dictyosome; ER — endoplasmic reticulum; IS — intercellular space; LD — lipid drops; M — mitochondrium; N — nucleus; OB — obturator; P — pit; PL — plastids; PM — plasma membrane (plasmalemma); R — ribosome; S — starch grain; SS — secretory substance; ST — multiseriate stigma papillae; T — tonoplast; V — vacuole; VE — vesicle.

As the pollination takes place by the mediation of gland tissues and these can be regarded as a type of nuptial nectaries remarks concerning nectary of Schnepf (1974) and Fahn (1979) among others are very valuable. There are some similarities between the ultrastructure of *Ricinus* extrafloral nectaries (Baker et al., 1978) and stigmatic "papillae" too.

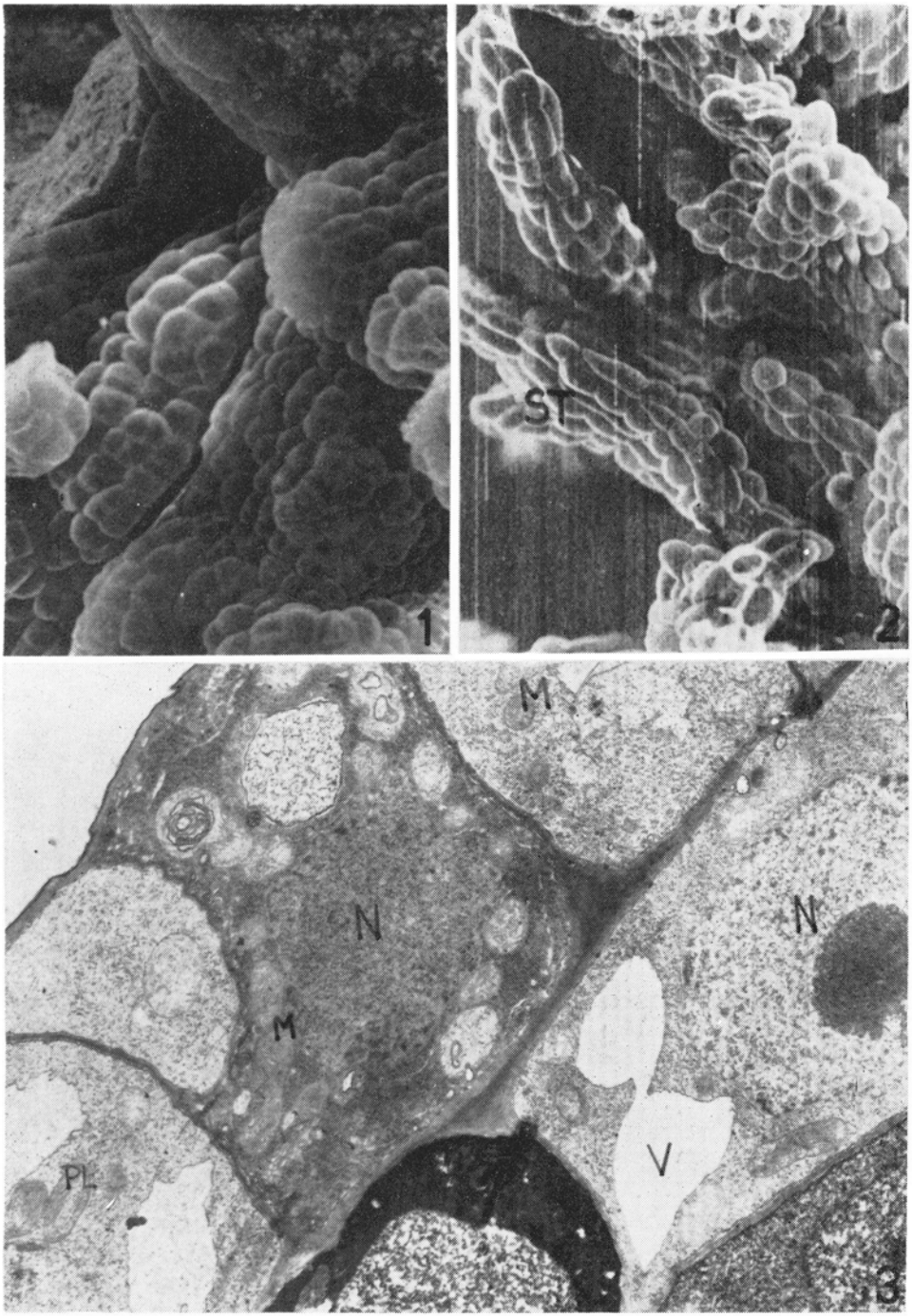
MATERIAL AND METHODS

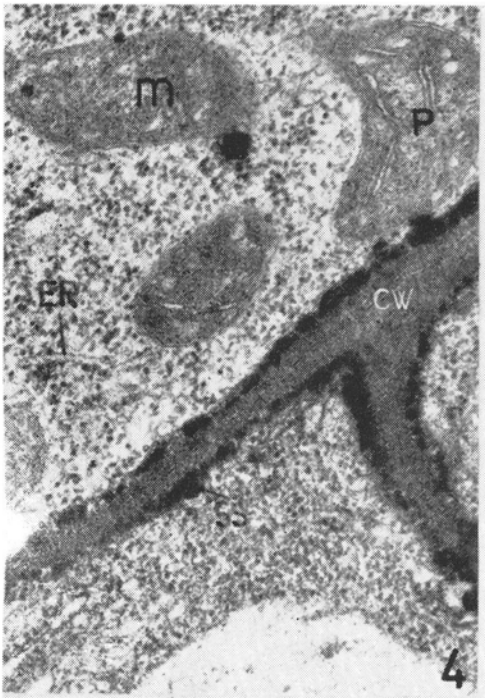
Examinations were performed by light microscope in serial paraffin sections, fixed with Bouin-solution and in half ultrathin Durcupan sections, stained with aniline-blue and fuchsin "B" (LM), by transmission electron microscope (Tesla BS 500) in ultrathin sections fixed with 4.5% GA in 0.1 M phosphate buffer, pH 7.2, at room temperature. The material was postfixed with 1% OsO_4 and stained with uranyl acetate and lead citrate, and by scanning electron microscope (Jeol LTD) in materials (SEM) having been fixed in 5% GA solution and dried using the critical point method (CO_2).

RESULTS AND DISCUSSION

At the beginning of the organisation of the pistil when the carpels are still homogeneous meristematic protuberances, the neighbouring cells in the protoderm and in the two-three layers underneath considerably differ from one another as to density and in the quantity of osmiophylic substances (Fig. 3). In the process of fine structural changes the density of the cytoplasm, the quantity of ribosomes and polysomes increases. The surface of the nucleus will often become lobed. The cytoplasm around the vacuoles forms an electron-translucent halo which might be due to vacuolar enlargment and/or autolysis. The plastids are of irregular shape, with a stronger density than the cytoplasm, their stroma membranes can be clearly recognized but the grana hardly. The number of dictyosomes is small. The thickened plasmamembrane becomes waved.

PLATE I

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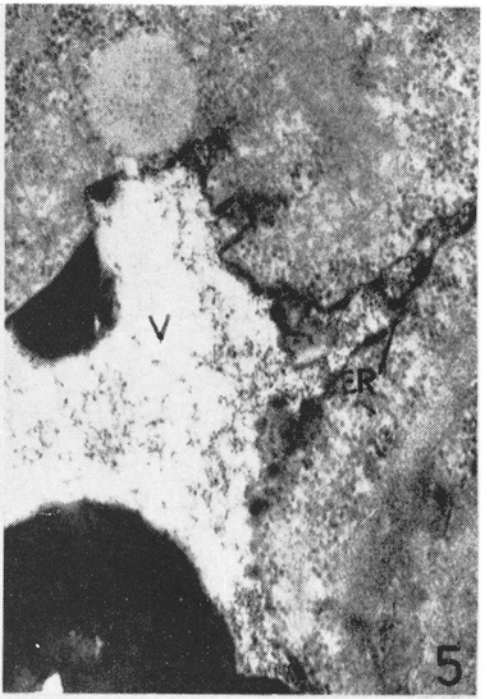
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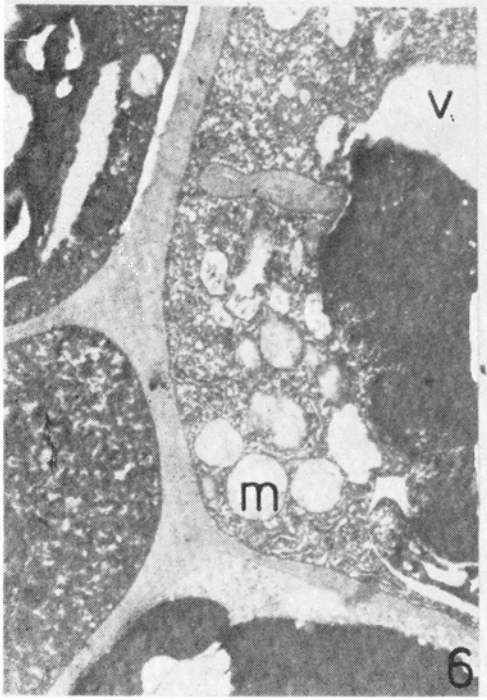
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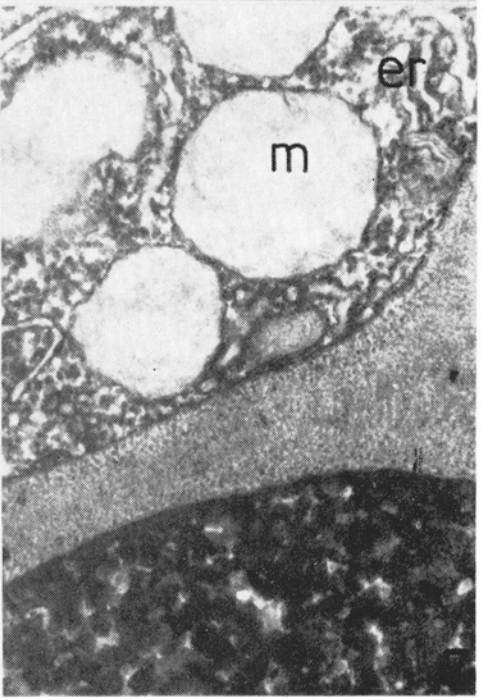
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Plate II

Fig. 4. Osmiophyl substances appear between the plasma membrane and cell wall. × 62000

Fig. 5. Dilatation of the ER is visible. Secreted substances will appear in the vacuole too. × 62000

Fig. 6. Cytoplasm is disorganized and vacuoles are mixed with it. × 10000

Fig. 7. A detail from the Figure 6 shows the swollen ER and the desintegration of mitochondria. × 38000

Abbreviations as in Plate I.

The differentiation of the stigma primordium is almost simultaneous with the development of the integuments. Most cells of the stigma protoderm and the second-third layer underneath can have secretory function only the level of differentiation are not the same. Enlargment of increasing number of plastids and mitochondria, the considerable dilatation of ER (Fig. 5) and the appearance of vesicles of probably ER origin are to be seen. Part of the secreted substances is localised between plasmalemma and cell wall by exocytosis (Fig. 4). The secreted substances will appear in the vacuole too (Fig. 5).

The beginning of stigma branching coincides with macrosporogenesis and its final development with macrogametogenesis (Figs 1, 2). In the former case the obturator above the micropyle can already be observed. The degenerating process of the secretory cells starts. The recognition of mitochondria and plastids becomes uncertain because of autolysis, the ribosomes cannot be identified either, the continuity of the tonoplast is lost (Figs 6, 7).

In the cells of the obturator with papillary surface (Figs 8, 9) being of similar density to one another the differentiation process is very rapid. Already in young state vacuolisation is intensive. The number and variety of cytoplasmic organelles is significant, numerous plastids with starch, mitochondria, dictyosomes can be found, the later more frequently than in the stigma. The frequency of rough ER is medium. On the ground of its structure the obturator cannot be regarded as a typical secretory tissue (F a h n, 1979). Between the relative thickness of the cell wall and the frequency of dictyosomes a correlation may be supposed. The further differentiation of cells can be related to the fact that the main function is the nourishing of the pollen tube. Later, with increased vacuolisation the starch content of the plastids will grow and during tube growth its mobilisation starts. The quantity of homogenous gray bodies (probably lipid drops) will increase. Intercellular spaces with small osmiophylic granuls are characteristic of the obturator (Fig. 10).

The primordium of the seed appendix (caruncle) consisting of closly connected cells is formed from the derivates of the apical cells of the outer integument after organisation of the obturator. Its differentiation, as opposed to the obturator is a relatively longer process. The beginning

Plate III

Fig. 8. Papillary surface of the obturator (SEM). × 650
Fig. 9. Longitudinal section of the obturator and the outher part of the caruncle (LM). × 300
Fig. 10. Papillae of the obturator in cross-section. × 6000
Fig. 11. Vesicles of ER origin can be seen in the caruncle-cell. × 16000
Abbreviations as in Plate I.

of its development is simultaneous with macrogametogenesis and it reaches its final form in the ripe seed. Its thin walled cells (in ripe flower) are almost completely meristematic, whereas an increase vacuolisation could only be found in the outer layers. Frequency of plastids, mitochondria and ribosomes is characteristic of its cytoplasm besides the large nucleus, but ER occur rarer. After fertilization the process of vacuolisation will start in the inner cell layers too, while the ER increases and in some plastids starch is formed. Later on vesicles are formed from the ER, mobilisation of starch begins, and the cell walls start to thicken. The caruncle finally have very thick walled cells, and pits can be seen in places. The thick cell wall consists mostly of cellulose but probably also hemicelluloses and mucilage are present, the former supplies nourishment for pollen germination, and tube growth the latter secures the water intake necessary for germination. In the last phase the cytoplasm becomes degenerated, and lipid drops can be observed (Fig. 11).

Our examination try to elucidate merely a few detalis but several other problems require further investigations.

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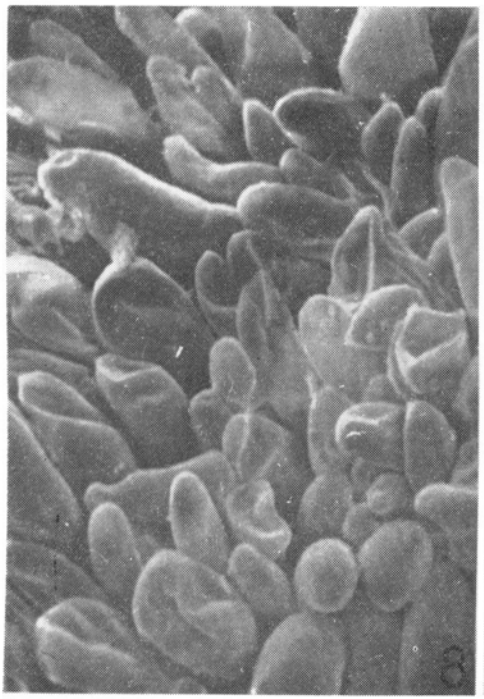
PLATE III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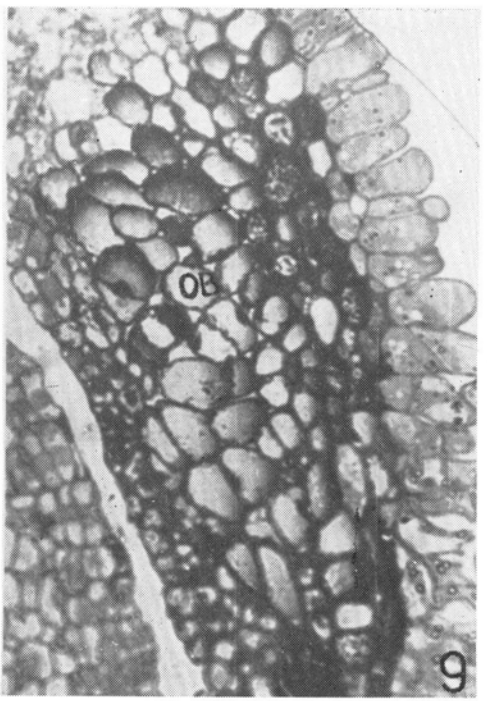

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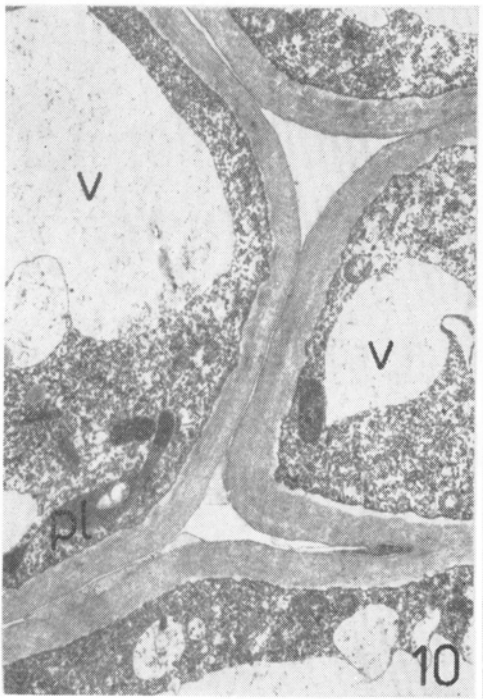

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