Sporoderm infrastructural and cytochemical modifications in cytoplasmic male sterile broad-bean (*Vicia faba* L.)*

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Abstract

A comparative study of mature sporoderms of sterile and fertile pollen grains was performed using electron microscopic techniques. In sterile pollen grains, intine is lacking; ectexine sculpture is reduced and tectum is overlaid by membranous systems. Infratectal texture is compact and a sporopollenin granulous mass is obturing the aperture central region. Endexine reacts with proteins and acidic carbohydrates tests.

INTRODUCTION

Cytoplasmic male sterility is used in various crops to produce functional female plants for hybrid seed production. Several studies conducted at light and electron microscope have established the timing of the cytological events leading to anther sterility (Edwardson, 1970; Harvey et al., 1972; Heslop-Harrison, 1972; Laser, Lerssten, 1972).

In *Vicia faba* L. this sterility is indicated by the genesis of abnormal shaped pollen grains with a damaged cytoplasm and a highly modified sporoderm, the latter is the object of this article.

MATERIALS AND METHODS

Normal and cytoplasmic male sterile *Vicia faba* anthers grown in the "station d'amélioration des plantes de Dijon (France)" were fixed by glutaraldehyde, post-fixed by osmium tetroxide and embedded in araldite (Glaubert, 1978). Ultra thin sections were stained either with usual uranyl acetate followed by lead citrate (Berlyn, Micksche, 1976) or with PATAg for the neutral carbohydrates detection (Thiey, 1967), with P. T. A. HCl and P. T. A. chromic acid for the acidic carbohydrate identification (Pears, 1968), and P. T. A. acetone for the proteins localization (Pears, 1968).
RESULTS

Compared with the fertile pollen grain sporoderm, those of sterile pollen grains are different both in their infrastructural configuration (Fig. 1 A-D) and in their cytochemical reactivities (Fig. 2 A-H).

Sporoderm cytochemical modifications

Among the most typical facts we shall mention: 1) intine absence; 2) exine sculpture reduction; 3) membranous systems overlaid on the tectum; 4) compactness of the infratectal zone where the footlayer is difficult to see; 5) obturation by a sporopollenin granulous mass of the central region of the aperture the general configuration of which is greatly modified.

Sporoderm infrastructural transformations

In the fertile pollen, intine strongly reacts with P. T. A. acetone (Fig. 2A) and very lightly with PATAg (Fig. 2D). Endexine is not contrasted by cytochemical tests (Fig. 2 A-D); whereas, ectexine becomes electron dense after P. T. A. chrome acid (Fig. 2 C) and PATAg (Fig. 2 D).

In the sterile pollen, ectexine reaction to the used cytochemical tests is not modified (Fig. 2 E-H) but endexine is vigorously contrasted by P. T. A. acetone (Fig. 2 E) and P. T. A. HCl (Fig. 2 F):

CONCLUSIONS

When they exist (De Vrije, 1e, 1970), the structural modifications of sterile pollen sporoderm are associated with: 1) a more or less precocious interruption of the pollen wall development owing to the sporal cytoplasm destruction along with a juvenile aspect of the sporoderm (Cheng et al, 1979; Horner, Rogers, 1974; Horner, 1977); 2) a more or less accentuated disorder of the cytobiological mechanisms participating in the exine and intine genesis: in that case, the achieved sporoderm presents a new texture (Albersten, Palmer, 1979). Thus in Vicia faba, these two modalities simultaneously participate in the sporoderm structural anomaly as it occurs in wheat (Young et al., 1979).

The specific cytochemical reactivity of the sterile pollen endexine is an original and important datum: it demonstrates that the sporopollenin biosynthetic events are principally disturbed through sporal cytoplasmic way leading to a modified endexinic chemical composition. Thus, cytoplasmic male-sterile plants could be interesting materials to analyse the biosynthesis processes and the chemical composition of the sporopollenin.
Fig. 1 A-D. Sporoderm infrastructural modifications (T.E.M., glut/OsO₄, Uranyl acetate-lead citrate).

A: extra-apertural fertile pollen exine (× 24,000); B: extra-apertural sterile pollen exine (× 24,000); C: fertile pollen aperture (× 12,500); D: sterile pollen aperture (× 12,000).

Fig. 2 A-H. Sporoderm cytochemical modifications (T.E.M., glut/OsO₄, P.T.A. acetone: A and E; P.T.A. HCl: B and F; P.T.A. chromic acid: C and G; PATAg: D and H).

A-D: extra-apertural and apertural fertile pollen exines (× 20,000); E-H: extra-apertural sterile pollen exines (× 21,000).

Abbreviations as in Fig. 1.
Acknowledgments

This work was supported by R.C.P. 575.

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