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Isoperoxidases and microspore differentiation in *Daucus carota* and *Foeniculum vulgare*

(Key words : *Daucus carota*/*Foeniculum vulgare*/isoperoxidases/microspore differentiation)

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Abstract

The paper contains data on the isoperoxidase profile at different stages of microsporogenesis and male gametogenesis of *Daucus carota* and *Foeniculum vulgare*. The results highlight that development of microspores and male gametophyte is accompanied by an increase in the activity of specific isoperoxidases.

Introduction

Reproduction is an attribute of all living beings It is accomplished through the sexual and asexual means. The former is not only a method of sexual reproduction but also a method for generating variability because it involves gametic recombination. The basic physiological processes involved in the differentiation of gametes in plants, is a problem which is engaging a number of workers. Some of those studies have pointed out that, among other factors, the male gamete development is under the control of isoperoxidases[1-7]. In this laboratory as well, it has been recorded that the activity (expressed in terms of the quantity and diversity) of isoperoxidase increase with the onset of pmc meiosis and it declines after the meiosis has been completed[8,9]. The present work is in continuation of the same on two umbellifer species having different umbel organisation.

Materials and Methods

Extracts from floral buds of *Daucus carota* and *Foeniculum vulgare* were analyzed at different developmental stages for isoperoxidases as described earlier[9]. The isozymes were extracted by crushing flower buds in phosphate buffer maintained at pH 7.3 (30 mg per 0.15 ml). Separations were achieved on 7.5% polyacrylamide gels, using Tris-glycine (pH 8.3) as the electrode buffer. The isoperoxidases were detected by the Benzidine-H_2O_2 reaction.

Results

(i) *Daucus carota* : The peroxidase profiles of the different stages of microsporogenesis and male gametophyte development in the male and bisexual flowers have been illustrated in Fig. 1. The peroxidase patterns of the male and bisexual flowers match

at all stages of microsporogenesis and male gametogenesis. In both cases, the peroxidase activity is maximum during pmc meiosis. With the completion of meiosis, the diversity is reduced, with the result that, only three peroxidases are present at the time the pollen grains differentiate. At the time of anthesis the number is further reduced to two bands.

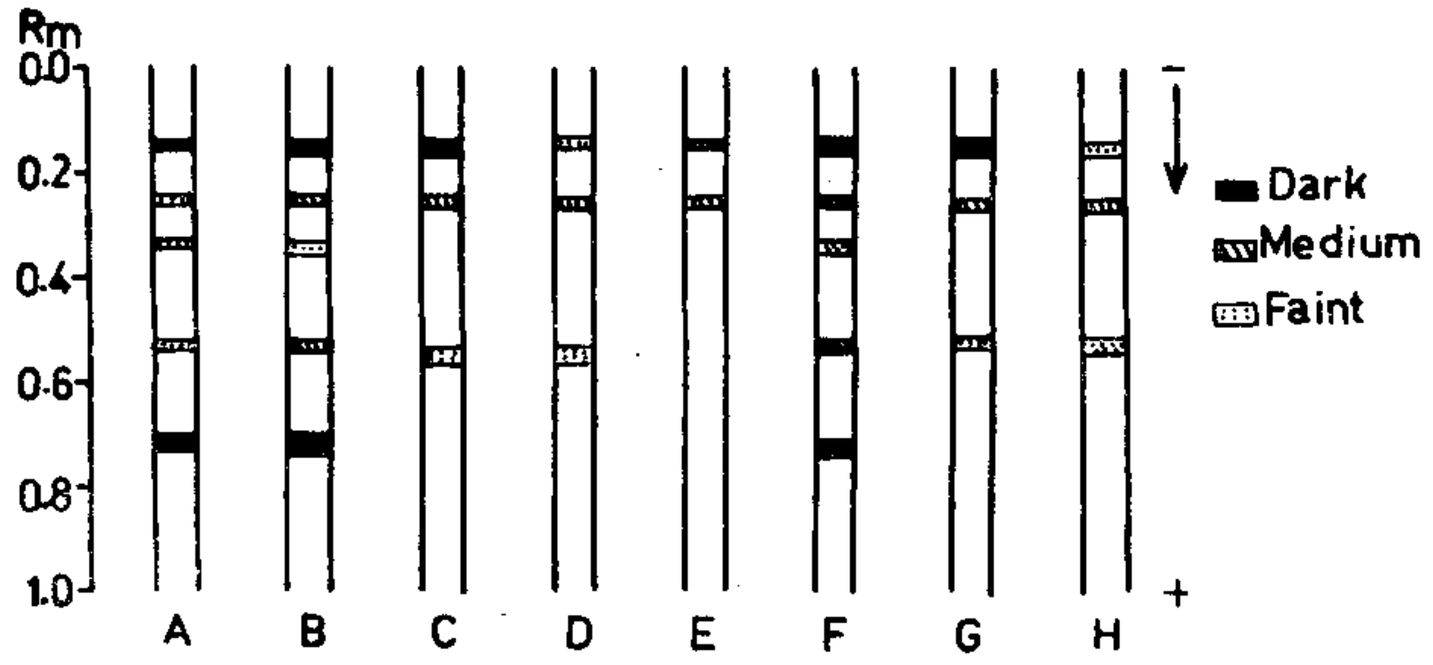

Fig. 1. Electrophoretograms representing isoperoxidases spectra at different stages of pmc meiosis in bisexual (A–E) and male (F–H) flowers of *Daucus carota*. A – Premeiotic stage, B & F – Meiotic stage, C & G – Pollen grain stage, D & H – Dehiscence stage, E – Post dehiscence stage.

Out of the five peroxidases recovered from the anthers, two, represented by band nos. 3 & 5 are exclusive to anthers at the pre-meiotic stages. These increase in quantity while meiosis is in progress, indicating their involvement in accomplishing this process. Reduction in intensity of band nos. 1 & 4 during the post–meiotic stages implicates them also in male meiosis.

(ii) *Foeniculum vulgare* : Fig. 2 summarises data on the isoperoxidases present in *F. vulgare* during microsporogenesis and male gamete development. In all, seven isoperoxidases have been separated from buds of different developmental stages.

Out of these, two, represented by band nos. 5 and 6, are specific to anthers undergoing meiosis. The peroxidases that separates as band no. 7, is present in anthers preparing for or undergoing meiosis, but it varies in quantity at the two stages; the band is faint during pre-meiotic phase but intensifies latter. Distribution pattern of the type suggests that, the three isoperoxidases are in some way involved in the pmc meiosis. Total absence of these bands from the electrophoretograms of the flowers deprived of their stamens through emasculation, support this conclusion further.

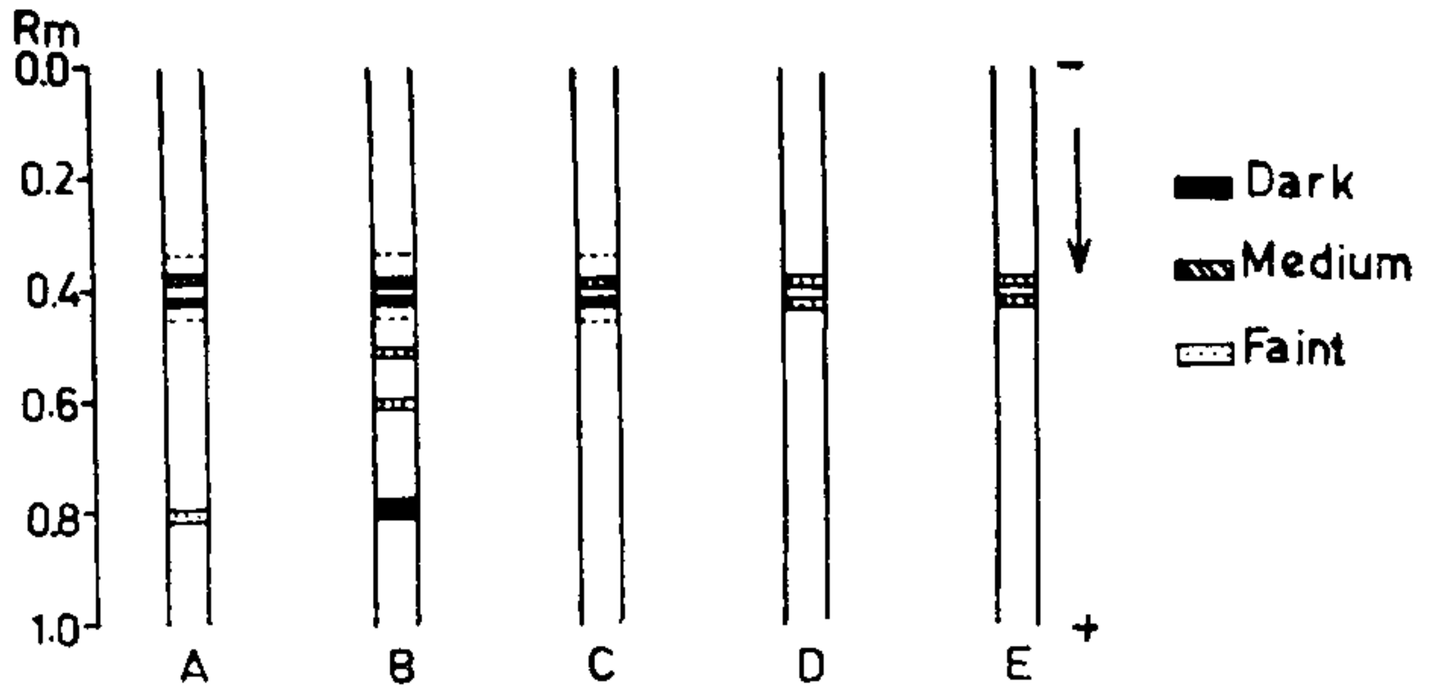

Fig. 2. Electrophoretograms representing isoperoxidase spectra at different stages of pmc meiosis in *Foeniculum vulgare*.

A- Premeiotic stage, B- Meiotic stage, C- Pollen grain stage, D- Post dehiscence stage, E- Emasculated flowers.

Discussion

Of the various roles that isoperoxidases play in plant growth and morphogenesis, differentiation of gametes is one. This role of the irshas been brought to light only recently by Kahlem[1-3], Durand *et al.*[4], Koul and Bhargava[8], and Bhargava and Koul[9]. In order to validate this further, isoperoxidases assaying was undertaken at different stages of development of flowers of carrot and fennel which represent andromonoecious and monoecious systems respectively. The peroxidase patterns of the male and hermophrodite flowers match at all stages of microsporogenesis and male gametogenesis. The diversity of peroxidases is maximum during the pre-meiotic and meiotic stages of anther development. At the completion of meiosis the diversity is considerably reduced (Fig. 1). This type of distribution suggests some kind of relation between pmc meiosis and peroxidase activity. Out of the five peroxidases recovered from the anthers at different stages, the two constituting band nos. 3 & 5 are exclusive to the pre-meiotic and meiotic phases. These two peroxidases increase in quantity while meiosis is in progress. Two other peroxidases, which reduce in quantity after the completion of meiosis are those constituting band nos. 1 & 4.

The same situation exists in case of *Foeniculum vulgare* as well. Band nos. 5 & 6 are restricted to anthers which are undergoing meiosis. Moreover, band no. 7 is present both at pre-meiotic and meiotic stages but decereases in quantity thereafter. It is, therefore, obvious that these three peroxidases are in some way involved in pmc meiosis. Absence of these peroxidases from electrophoretograms prepared from emasculated flowers vindicates this conclusion further.

Fluctuation in the number of peroxidases, during pmc meiosis, has been observed (Observation of the authors) in some other species which include *Scandix pecten-veneris*

(andromonoecious), *Dioscorea composita* and *Carica papaya* (dioecious) and *Ricinus communis* (monoecious). Existence of male specific anodic peroxidases have been found in *Mercurialis annua*[1,2], *Morus nigra*[6] and *Coccinia indica*[5]. From the data in hand, it can be pointed out that irrespective of the method of reproduction all angiosperms have very high peroxidase activity during the course of pmc meiosis.

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