DETERMINATION OF SEED NUMBER PER POD IN OILSEED RAPE

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INTRODUCTION

The number of seeds within a pod depends on the sequential determination of the number of ovules, the percentage of ovules which develop a fertile embryo sac, the rate of fertilisation and finally the percentage of fertilised embryo sacs which are retained until maturity. It is important to establish which of these successive stages are important in the determination of the final number of seeds, and how these stages respond to external and internal factors.

DEVELOPMENTAL ASPECT OF THE DETERMINATION OF SEED NUMBER PER POD

Introduction

Important abortion seems to occur at the open-flower stage when many ovules fail to resume growth, while a second, and less important, series of abortions occurs during the following days (Pechan, 1983). Quantitative microscopic observations of ovules and embryo sacs at anthesis and during bud development have been made in order to establish the cause of the first series of abortions.

Materials and methods

Oilseed rape plants (cv Haplona) were grown under greenhouse conditions. Flowers were sampled from basal and apical positions on the terminal raceme at flower opening and one and two days thereafter. Buds at various developmental stages were also collected before flower opening. The stage of development of these buds was assessed morphologically by bud length. The tissues were fixed in CRAFIII, and either cleared in Herr's fluid or wax embedded, sectioned and stained.

Results

At the open flower stage, about two thirds of the ovules contained a complete embryo sac, a few ovules had an immature embryo sac and in other ovules no embryo sac could be found. Two days after flower opening, most embryo sacs had been fertilised and were developing actively. The number of fertilised embryo sacs was furthermore similar to the number of seeds at maturity (Fig 1). Thus, the failure to develop an embryo sac by a number of ovules (ovule sterility) appears to be the main factor limiting the number of seeds that develop within an ovary.

The number of ovules containing an embryo sac at flower opening was not signficantly different from the number of ovules containing a uninucleate megaspore when the buds were 4 mm in length (Table 1). Semiquantitative observations on meiosis suggested that failure of gametophytic development occurred mainly during late meiosis and/or the uninucleate megaspore stage i.e. when the buds were between 2.5 and 4 mm in length.

PHYSIOLOGICAL ASPECTS OF THE DETERMINATION OF SEED NUMBER PER POD: INFLUENCE OF LIGHT AND TEMPERATURE

Introduction

The results reported above suggest that the late meiosis/early megaspore differentiation stage is critical in the determination of the number of seeds within a pod. The influence of various factors on this process was studied and in particular those of weather conditions.

Materials and Methods

Plants were grown in individual pots in a growth cabinet under the following conditions: 0.35 Einstein/m^2/s light intensity, 16 hours photoperiod, 18/14° C day/night Temperature. Low temperature treatments were imposed by transferring the plants into a similar cabinet with the same lighting conditions but with a temperature regime of 13/8° C; shading treatments were imposed with green plastic netting inside the cabinet which reduced the light intensity by 60-65%. The experimental plan is given in Table 2. The duration of the treatments was 10 days. At the beginning and at the end of the treatments, the stage of development of every fifth bud/flower/pod of the terminal raceme was recorded. Flowers were sampled from a basal and apical position on the terminal raceme for histological analysis. At harvest, the terminal raceme of each plant was divided into the flower node groups 1 to 10, 11 to 20, 21 to 30, 31 to 40 and above. For each sample, the number of pods, seeds and the seed dry weight were determined. The results were analysed by analysis of variance with 6 plant replicates per treatment.

Results

On a plant basis, cold treatments led to reductions in seed number per pod, which were significant at the flowering stage (Fig 2). Shading however had no effect. A detailed analysis of the terminal raceme indicated that the decreases in seed number per pod were localised: on the plants treated at the bud stage, pods at nodes 1-15 were affected, but to a small extent. On the plants treated at the open-flower stage, seed numbers were severely reduced in those pods located at nodes 20-35 (Fig 3), and this effect was accompanied by pod abortion; small or aborted pods were also observed in the basal region of the lateral racemes of these plants. Cold treatments at both stages led to a larger region of the terminal raceme being affected when the low temperatures were accompanied by low rather than high light intensity.

Histological analysis of the basal flowers indicated that the lower seed numbers induced by low temperatures were due to a significant reduction in ovule fertility; seed retention was, on the other hand, increased (Fig 4). Similar trends, though not significant, were observed in the apical flowers sampled from a region where seed numbers per pod were not severely affected. For both treatments, the pods with a reduced seed number per pod were those which had arisen from buds which were 4-5 mm in length and undergoing early megaspore differentiation at the time of treatment.

In addition, some compensatory mechanisms for the lower number of seeds per pod were observed: increased production of branches, flowers and pods for the bud stage treatments but extension of flower production on the upper racemes and higher seed mean weight for the flower stage treatments (Table 3). Seed yields were not significantly affected overall, except for the B2 treatment (low temperature, high light at the bud stage), which gave a higher yield.

GENERAL DISCUSSION

The results presented here suggest strongly that: (1) the number of seeds within a pod is mainly determined by the percentage of ovules which develop an embryo sac (ovule fertility) rather than by the processes of fertilisation and seed development themselves, and (2) the stage around late meiosis and/or early megaspore development is critical in the determination of ovule fertility.

The early megaspore stage appeared to be very sensitive to low temperatures, particularly after plant anthesis. It remains unclear, however, how the temperature effect was elicited. This could be through a direct effect of low temperatures or an indirect effect through nutritional (e.g. C-assimilate and/or nitrogen supplies) or hormonal factors (e.g. ABA and/or Cytokinin metabolism). The great effects of low temperatures at the open flower stage may be because more buds were at a sensitive stage and/or because of the physiological changes induced by the presence of flowers and pods.

The absence of an effect of shading at normal temperatures (which is in agreement with Tayo and Morgan (1979), Inanaga and Kumura (1987) and Ancha (1988)), suggests that C-assimilate supply has little influence on ovule fertility and therefore that the flux of carbohydrates towards the buds is determined by sink demand. The greater impact of low temperatures under shaded conditions could further indicate that the sink strength of the flower buds depends on temperature.

Since the lowering of temperature seems too small to cause a real stress, ovule sterility is unlikely to be a stress response but to participate in the normal physiological regulation of seed number per pod. This result has some fundamental significance since only stress-induced sterility has been reported so far, e.g. in cereals and oilseed rape (Evans et al., 1975; Polowick and Sawhney, 1988). Applied implications include a better understanding of yield determination under field conditions and possibly breeding for less sensitive varieties.

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Table 1. Comparison of buds at the stage 4 mm in length (uninucleate *megagametophyte*) and open flowers. Mean ± standard error. NS: statistically non significant at 5%.

	Buds 4mm	Flowers	Difference of the means
No of ovules	24.6±0.3	25.4±0.4	0.8NS
No of megagametophytes	14.4±0.7	14.4±0.7	0.1NS

Table 2. Experimental planning and key to the treatments.

Stage of treatment	Treatment light temperature	normal normal	shaded normal	normal low	shaded low
Stem elongation		C	B1	B2	B3
Flowering		C	F1	F3	F3

Table 3. Influence of the light and temperature regime on yield and yield components on a plant basis.

Treatment	C	B1	B2	B3	F1	F2	F3	LSD (5%)
Raceme No	3.1	3.5	3.5	3.5	3.3	2.7	2.7	0.6
Flower No	94	108	129	113	99	108	117	13
Pod No	73	84	91	84	74	80	83	9
Seed No	660	710	750	660	660	600	620	85
Seed No/pod	9.1	8.6	8.3	7.9	9	7.4	7.5	0.9
Seed Mean Wt	3.8	3.8	3.7	3.8	3.7	3.95	4.0	0.25
Seed Yld (g)	2.5	2.7	2.8	2.5	2.5	2.4	2.5	0.3

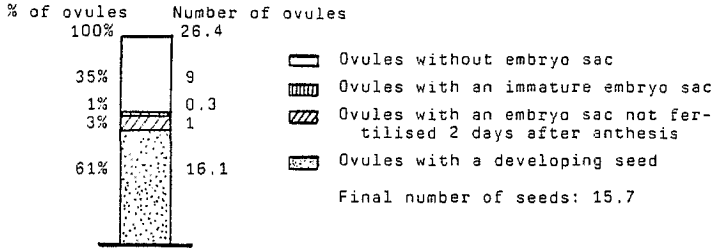

Fig 1. Contribution of the processes of ovule and embryo sac development, maturation and fertilisation to the number of seeds of a pod.

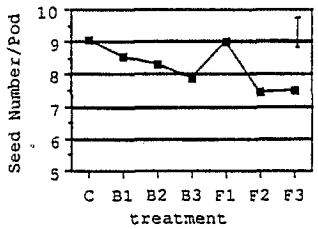

Fig 2. Influence of the light and temperature regime on seed number per pod. Plant basis.

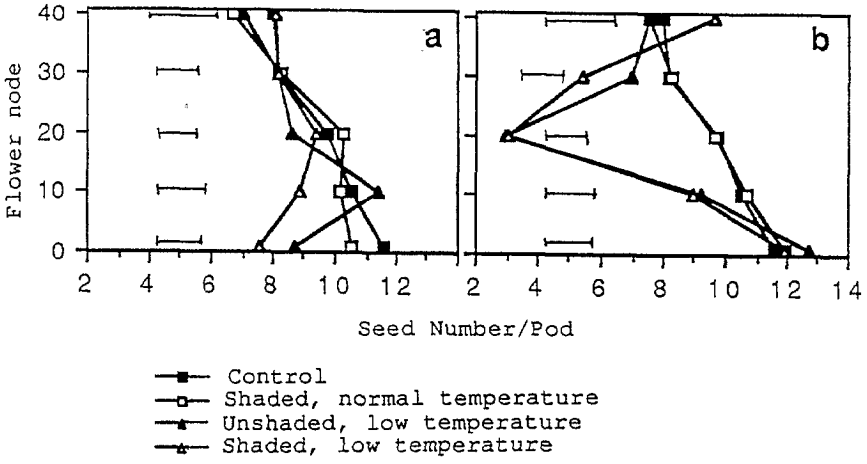

Fig. 3. Influence of the light and temperature regime on seed number per pod on the terminal raceme for: (a) plants treated during stem elongation; (b) plants treated during flowering

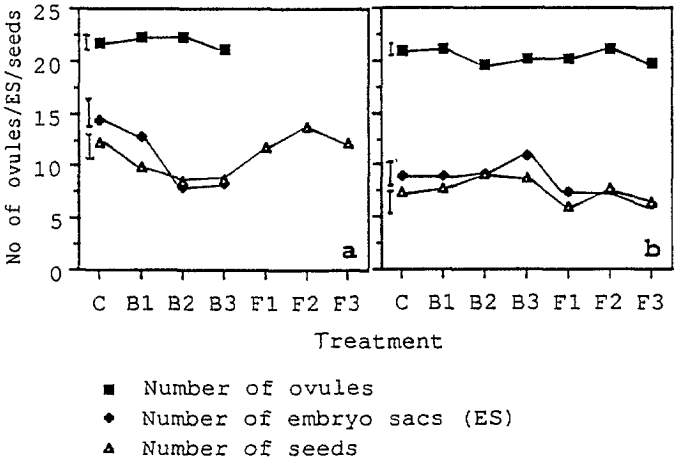

Fig. 4. Influence of the light and temperature regime on ovule, embryo sac and seed development in flowers sampled on the terminal raceme at the nodes: (a) 3±1; (b) 38±1