A PRELIMINARY STUDY OF INSECT POLLINATION OF MALE-STERILE OIL-SEED RAPE (*Brassica napus*) IN 'MIXED' CULTURES WITH VARIABLE PROPORTIONS OF POLLINISING PLANTS.

Renard M.*, Mesquidà J.**

* INRA. Station d'Amélioration des Plantes. B.P. 29. 35650 Le Rheu. FRANCE.

** INRA, Laboratoire de Zoologie. B.P. 29. 35650 Le Rheu. FRANCE.

INTRODUCTION.

Promising performances of winter and spring oilseed rape have drawn attention to the interest of F1 hybrid varieties (Lefort-Buson & Datte 1985; Sernyk 1983). Amongst different sources of male cytoplasmic sterility currently under study (CMS, Rousselle 1981) is a CMS derived from radish (Ogura 1968; Bannerot et al 1983), improved by protoplast fusion (cybrids obtained by Pelletier et al 1983). However, because of difficulties encountered in selection of restorer lines (Pellan-Delourme 1986) the possibility of exploiting male sterility without fertility restoration in the hybrid F1 is being considered. One way of doing this is to use a 'mixed' variety, in which F1 hybrid male-steriles obtained from cybrids mentioned above are exploited as 'females', with male-fertile pollinisers, in a given proportion. Yield in such a mixture would be assured by the hybrid F1, the 'male' plants serving as pollen source. However, by-passing the problem of fertility restoration in cybrids in this way raises fresh problems. These make it necessary to examine the significance of factors affecting pollination, notably the role of bees, and of those involved in competition, in order to determine the type of male-fertile oilseed to be used, and the most appropriate proportion of fertile plants in the mixtures. As a first step work has begun using a same line of spring rape, in its male-fertile ('male') and male-sterile ('female') forms. Results of tests undertaken in 1985 are described here.

MATERIALS AND METHODS.

Alloplasmic male-sterile Brutor lines were selected by successive backcrossing of Brutor on cybrids FU 27, FU 58 and FU 118, obtained previously by protoplast fusion (Pelletier et al 1983.)

The 'mixed' varieties were composed of these three male-sterile cybrid lines as 'females' with Brutor line plants as 'male' pollinisers .

Mixtures of 5%, 10%, 20% and 30% Brutor were established for each cybrid, seeds being mixed mechanically, taking account of germination rate and weight of 1000 seeds. Proportions of male and female plants in field plots were checked systematically at flowering. They were usually slightly, but not significantly (chi square) lower than those expected (6.5% for 5% treatment, 8.4% for 10%; 15% for 20% and 26% for 30%.

Trials were run on four plots in 1985, isolated from one another and any other oilseed crop by several hundred metres. Each plot had 'female' lines with a same proportion of 'males'. Plots one to four thus corresponded to mixtures 5% to 30%. Trial plots thus provided four replications with three mixed varieties (FU 27, FU 58 and FU 118 with x% Brutor), randomly allocated. Basic trial units all involved twenty 7 m rows, spaced at 0.25 m, giving a 5.60 m strip with plant density 60-80/m^2.

Pollination depended on local factors (insects and particularly honey bees together with wind).

Observations (see Mesquida & Renard 1979 for methods) were mainly concerned with phenology (flowering synchronisation, nectar secretion) and insect biology (pollinator identity, behaviour, preferences). Pollinisation was assessed in terms of pod set on main stems and some parameters of yield.

RESULTS.

In general 'male' flowering covered 'female' flowering well, although cybrid FU 118 was distinctly late. Male plants showed a clear reduction in leaf surface, and were less vigorous, from onset of flowering, compared with cybrids, notably for the 5% and 10% mixtures.

Nectar secretion was higher for FU 27 and FU 58 (70% and 60% of parent male levels) than for FU 118 (27% of parent male level).

All male and female lines were highly attractive to bees, but FU 27 and FU 58 like their parent males were more so than FU 118.

Mean pod set was generally very high (more than 87%). This was comparable to that of the parent male (about 95%) for mixtures at 10% or more pollinisers for FU 27 and FU 58. In FU 118 equivalence relative to the male parent was found for the 30% mixture (Table 1).

Mean seed number per pod, and mean weight of 1000 seeds (Tables 2A and 2B) were generally higher for cybrids than for corresponding control males. Differences were not, however, significant with the exception of seed weight for FU 58 compared with its parent male (means 13% to 21% higher, Table 2B). Seed number per pod was higher for the 20% treatment for cybrids (18.9 or 110% of control level for FU 27; 20.4 or 120% for FU 58; 19.3 or 107% for FU 118, Table 2A).

Cybrid seed weight per plant was usually distinctly greater than that of the corresponding male control (generally two to three times, Table 2C).

Table 3 shows that all mean yield values for mixed varieties (cybrids + control males) with FU 27 and FU 58 were greater than those for FU118 in corresponding treatments (6% to 22%). FU 118 pods had certain deformations.

DISCUSSION.

Results obtained show that cybrids FU 27 and FU 58 possess some interesting advantages relative to FU 118, namely:

- better synchronisation of flowering with that of the 'male' alloplasmic line (it may be noted that the slightly later flowering of FU 118 relative to that of the 'male' line may be set against slower evolution of flowering in the latter.
- higher level of nectar secretion.
- greater attractiveness to bees.
- pod set equivalent to that of 'male' controls for mixtures at 10% or more.
- increased yield.

In any event the cybrids have better performance than the old male sterile source line (3.22.3.7) which is chlorophyll and nectar defficient. (Mesquida & Renard 1979).

Nevertheless the present study also indicates some negative features attributable to competition factors.

Low yields of 'male' lines relative to those of cybrids, lower vigour and reduced leaf surface (particularly in 5% and 10% mixtures) may result from inter-varietal competion . This is known to be operative in oilseed rape (Buson 1979). Better development and yield of cybrids might be due to hybrid cytoplasm (recombined mitochondria, oilseed chloroplasts, Vedel et al 1983), or to floral biology of the male sterile plant. However all cybrids did not behave in the same way, as shown by low FU 118 yield, which may be a consequence of pod deformations

Although the results require confirmation, it seems reasonable to admit that in terms of lines, cybrids FU 27 and FU 58 are better than FU 118 and that the 10% mixture , and more particularly that at 20% pollinisers, are satisfactory.

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TABLE 1 - MEAN LEVELS (%) OF CYBRID AND PARENT 'MALE' POD SET.

ISOLATES	CYBRIDS			"Brutor"
	FU.27	FU.58	FU.118	
5 %	95,80	95,67	87,33	98,67
10 %	95,00	95,47	87,47	95,47
20 %	95,80	97,67	87,67	92,80
30 %	95,47	97,13	93,33	95,13

TABLE 3 - MEAN YELD (Qx/ha)

	Mixed varieties			Means	Differences
	FU.27 (1)	FU.58 (2)	FU.118 (3)		
5 %	16,0	18,1	14,8	16,3 (A)	S **
10 %	28,5	29,8	26,1	28,1 (B)	
20 %	19,3	19,3	17,6	18,7 (AC)	
30 %	19,5	21,8	18,4	19,9 (C)	
Means	20,8(AB)	22,2 (A)	19,2 (B)		
Differences	S *				

(1) : FU 27 + parent 'male'

(2) : FU 58 + " "

(3) : FU 118 + " "

* : S p 5%

** : S p 1%

Means signalled by a same letter do not differ significantly.

TABLE 2 - MEAN YIELD COMPONENT VALUES FOR CYBRIDS RELATIVE TO PARENT 'MALES' FOR EACH ISOLATE.

	FU.27		FU.58		FU.118	
	T 1	FU.27 (en % T 1)	T 2	FU.58 (en % T 2)	T 3	FU.118 (en % T 3)
2.A. - Number of seeds / pod .						
5 %	17,0	107	16,3	117	17,0	95
10 %	14,3	120	14,8	109	14,3	117
20 %	17,2	110	17,0	120	18,1	107
30 %	15,2	111	18,4	79	15,9	103
2.B. - Weight of 1000 seeds (g).						
5 %	3,49	111	3,36	121	3,35	119
10 %	4,02	100	3,51	113	3,60	103
20 %	3,61	115	3,25	117	3,56	101
30 %	3,33	92	3,37	120	3,44	95
2.C. - Total seed production/plant (g).						
5 %	2,80	178	1,90	289	2,48	216
10 %	4,15	216	3,79	244	4,14	220
20 %	3,54	232	2,67	299	3,69	210
30 %	3,44	184	3,05	214	3,21	179

T 1 : Cybrid FU 27 control 'male'

T 2 : Cybrid FU 58 control 'male'

T 3 : Cybrid FU 118 control 'male'