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Results of investigations on the possibility of CMSpol utilization for winter rapeseed hybrid varieties development

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Gene-cytoplasmic male sterility - CMS*pol* found in rapeseed spring variety Polima in China (Fu 1981) is one of CMS forms which can be used for creation of hybrid varieties of oilseed rape. The factor limiting the possibility of utilization of CMS*pol* for hybrid seeds production is the instability of male sterility expression as well as low frequency of restoring and maintaining genotypes in *Brassica napus* species.
Heterosis effect gained in hybrids with CMS*pol* is also discussed (McVetty, Scarth, Rimmer 1991).

Material and methods

In Oil Crop Department in Poland up to now there have been selected:
- male sterile plants with *polima* cytoplasm in genotype of double improved winter oilseed rape,
- two maintaining double improved winter lines of rapeseed - PN 410/88 and PN 4315/89 assuring in CMS*pol* over 90 per cent of thermostable male sterile plants,
- two sources of restorer genes:
-double low winter rapeseed line PN 5297/86 - determination of male fertility restoration by two pairs of additive alleles,
- forms selected from partially male fertile plants (pMF) - determination by one dominant gene.

These materials as well as traditional spring variety Italy discovered as possesing dominant restorer gene of male fertility for CMS*pol* (Fang, McVetty 1987) were the objects of investigations.

Restorer genes from mentioned sources were introduced into genotype of double low winter rapeseed by crosses with different lines of this type of rapeseed or with double improved CMS*pol* lines. Inbred lines have been obtained from the both maintaining lines. From the maintaining line PN 410/88 by androgenesis in anther cultures have been obtained doubled haploids (DH). Progeny A_2 of doubled haploids was tested in hybrids with
male sterile lines for ability to maintain male sterile forms with stable expression of this trait during whole flowering period.

Observations of male sterility expression were made in greenhouse and field conditions. In greenhouse observations were carried out on three plants from each line and in the field on 20 to 30 plants from each line.
Heterosis effect in progeny F_1 of hybrids between male sterile lines and 20 restoring lines possesing in their genotype restorer genes originating from line PN 5297/86 was evaluated in the field trial in completely rendomized blocks design in four repetitions.
In F_1 hybrids and their parental forms the content of alkenyl glucosinolates in seeds has been examined using gas chromatography (Byczynska, Krzymanski 1981).

Results and discussion

There have been obtained maintaining and restoring lines of winter oilseed rape without erucic acid and with very low glucosinolate content by succesive backcrosses with different double low winter lines of rapeseed (Table 1). The oil content in seeds of maintaining lines and with restorer genes originating from PN 5297/86 is on the same level as in seeds of double improved varieties. Some lines with restorer genes originating from partially male fertile plants (pMF) and variety Italy are characterized by low oil content in seeds. This character has been introduced with genotype of spring varieties Italy and Polima. The increase of oil content is possible by multiple backcrosses with the best lines of double low winter oilseed rape.

The stability of male sterility expression in different environmental conditions is necessary for the use of the CMS system in hybrid seeds production.

In the case of structural forms of male sterility, such as CMS*pol*, in some environmental conditions, e.g. high temperature, a convertion to partial male fertility occurs (Bartkowiak-Broda 1991; Fan and Stefansson 1986).Therefore the maintaining lines containing a set of analogous recessive genes determining male sterility in homozygous stage are indispensable to obtain stable male sterile lines.

Earlier observations of PN 410/88 line ability to maintain CMS*pol* revealed that with the increase of homozygocity by inbreeding this line with CMS*pol* lines gave progeny with higher stability of male sterility expression (Bartkowiak-Broda 1991). Therefore from this line doubled haploids (DH) were obtained by androgenesis. Out of the investigated 33 DH lines (A_2 progeny) in hybrids with CMS*pol* 21 have been selected as complete maintainers (Table 2). These observations have been confirmed both in greenhouse and field conditions.

Hybrids of F_1 progeny between 20 different restoring lines and CMS*pol* lines revealed heterosis effect measured in comparison to the parents mean yield, ranged from 8 to 239 per cent. Over the standard double low variety yielded 14 hybrid combinations, in range from 0,7 to 50,5 per cent (Fig.1).

Results concerned with estimation of sterile cytoplasm *polima* value are controvertial up to now. It is known that the metabolic processes and their quickness in sterile cytoplasm are discriminated. This fact has negative influence on heterosis effect. The biological cost associated with the use of CMS system depends on the origine of CMS forms as well as on genotype introduced to sterile cytoplasm.

Investigations carried out in Canada by McVetty, Scarth and Rimmer (1991) showed that hybrids with cytoplasm *polima* display lower heterosis effect in comparison with hybrids based on *napus* cytoplasm. But there was stated that different genotypes have different level of tolerance for the defficiencies of *polima* cytoplasm. In this regard results of hybrid breeding program depend from the performance of the material generated.

In some investigated hybrids heterosis effect in alkenyl glucosinolate content has been found (Fig.2). The increase of glucosinolate content occured in each combination with high heterosis effect in seed yield. The highest heterosis effect in glucosinolate content was 59 perc cent in comparison to the parents mean glucosinolate content.

This results showed that it is necessary to hold strict selection concerning the glucosinolate content in seeds of hybrids componenets. In hybrids with high heterosis effect in yield also high heterosis effect in glucosinolate content can be expected.

Table 1.

Characteristics of restoring and maintaining lines

Lines	Number of lines	Oil [%]			Erucic acid [%]			Total glucosinolate content [μM/g f.f.d.m.]		
		$\bar{x}$	min	max	$\bar{x}$	min	max	x	min	max
Restoring										
pMF	18	45,1	36,7	48,7	0,0	0,0	0,2	4,39	0,6	10,8
Italy	21	42,5	34,5	48,2	0,0	0,0	0,3	7,86	2,9	12,2
PN 5297/86	41	48,2	46,1	50,5	0,0	0,0	0,2	3,48	0,2	7,8
Maintaining										
PN 410/88	37	46,1	44,1	48,1	0,0	0,0	0,3	5,5	0,3	10,3
PN 4315/89	22	48,6	45,5	50,4	0,0	0,0	0,2	4,4	0,7	8,2

Table 2.

Ability for maintaining of CMS pol by doubled haploids obtained from line PN 410/88

Number of DH line	Type of plants obtained in progeny with line CMS *pol*
PN 5756/4	MS,pMS
/5	MS
PN 5792/1	MS
PN 5793/1	MS
/2	MS
/6	MS
/7	MS, pMS
/10	MS
PN 5794/3	MS
/4	MS
/6	MS, pMS
/7	MS,pMS
/8	MS
/9	MS, pMS
/11	MS
/13	MS
/15	MS, pMS
PN 5795/1	MS, pMF
/4	MS, pMS, pMF
PN 5796/1	MS, pMS
/3	MS
/4	MS, pMS, pMF
/6	MS
/7	MS
PN 5756/9	MS
/15	MS
PN 5797/8	MS, pMF
/10	MS
/11	MS
/12	MS
/13	MS
/14	MS
/15	MS, pMF

MS - male sterile plants
pMF - partially male fertile plants
pMS - partially male sterile plants

Figure 1.

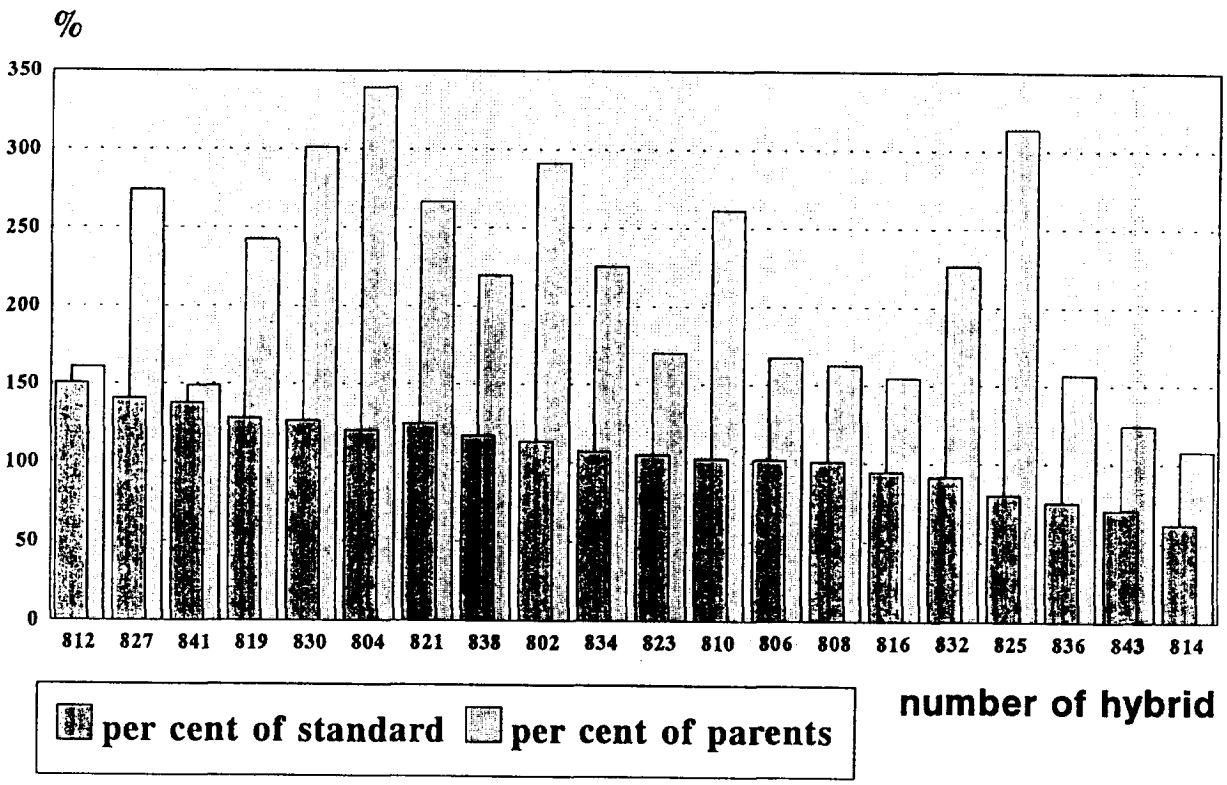
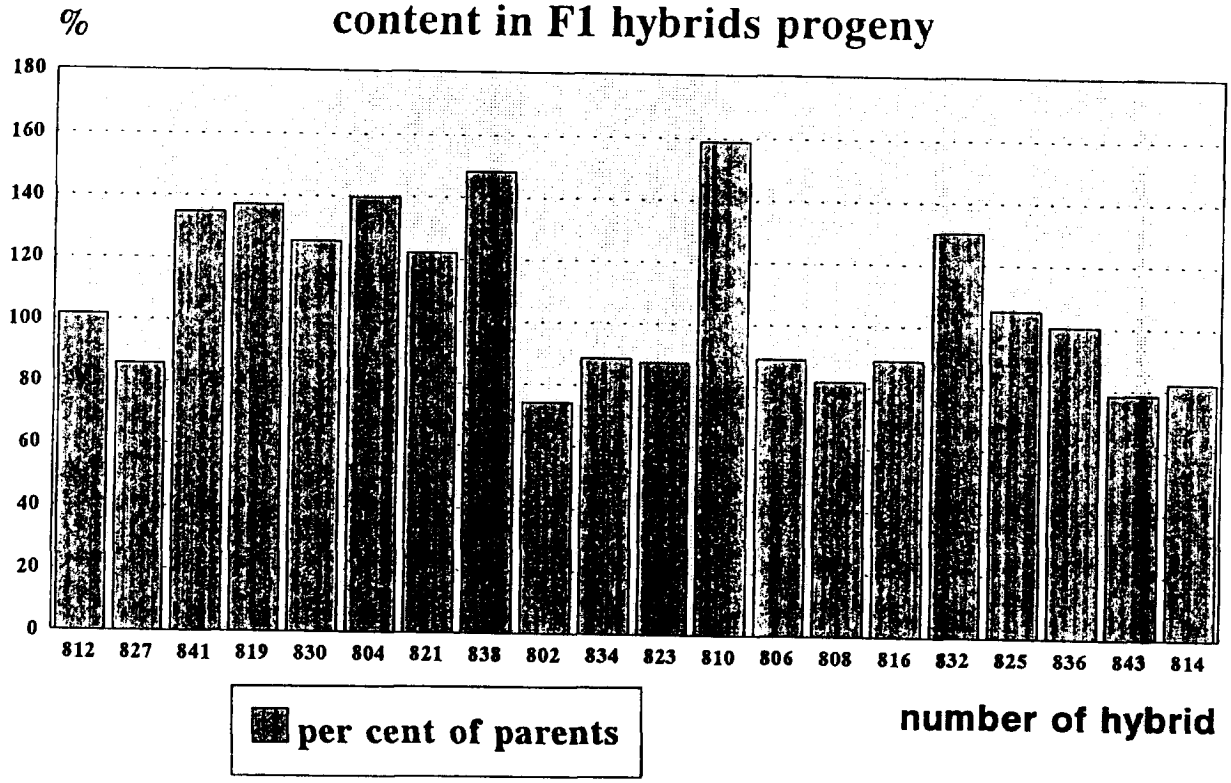


Figure 2.



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