CHARACTERISTICS OF SOME ANTHER-DERIVED DOUBLED HAPLOID LINES OF WINTER RAPESEED /BRASSICA NAPUS L./

Teresa Cegielska, Jan Krzymański

Plant Breeding and Acclimatization Institute
Gorzów Wlkp., Poland.

Anther culture not only offers increased efficiency in the production of homozygous lines but also may provide breeders with a tool to reveal the genetic variation enclosed in a population.

In previous papers we reported on the production of dihaploid rapeseed lines by anther culture /Nałęczyńska, Cegielska, 1984/. The subsequent task is testing of anther-derived doubled haploids in field experiments for uniformity and variability of their traits.

First results of field tests reported in this paper concern the androgenetic dihaploid /DH/ lines obtained from winter rapeseed cultivars and hybrids.

Materials and methods

Sixty-six anther-derived doubled haploids from the following winter rapeseed material: Jet Neuf cv.-42 DHs, Janpol cv.-10 DHs 592 F_1 hybrid-12 DHs, 4-L F_1 hybrid-2DHs. have been tested in the field experiments. Details and results of anther culture procedure and responsiveness of the anther taken from this material as well as method of colchicine treatment for doubling chromosomes have been described recently /Nałęczyńska and Cegielska, 1984/.
For this study seeds of androgenetic dihaploid lines were sown in autumn of 1984, in an experimental field in Poznań. Every DH line was sown in 5 rows, each two meters long with 7 plants, which resulted in 35 plants per line.
The following observations and measurements were done on each plant from the line: seed yield, thousand seed weight /TSW/, plant height, height to the first branch, number of branches. Fat content in seeds and glucosinolates content in meal were also estimated.

The spring sowing of dihaploid winter rapeseed lines was used as a test for homozygosity /Nałęczyńska and Krzymański 1985/.

For this purpose observation of uniformity or differences in appearance among the plants within each DH line were performed. A particular attention was given to flower-shoot formation by plants. The seeds of sixty-six studied DHs were sown on May 20th,1986, and the plants were observed till the end of August 1986.

Results and discussion

It is of a very great importance to check whether the obtained doubled haploids lines are fully homozygous. Morphological and physiological uniformity or differences among plants within DH lines as well as between DH lines were especially evident in an experiment with spring sowing of these winter rapeseeds. Plants within lines of studied dihaploids appeared to be highly homozygous. As it is seen in Table 1 out of 66 tested DH lines 51 DHs were uniform and developed flower-shoot, 13 DHs were uniform and remained at rosette stage. Only two lines from Jet Neuf cv. segregated, 60% of the plants developed flower - shoot and 40% of the plants did not. In parent cultivars Jet Neuf and Janpol 76% and 74% plants respectively developed flower-shoot. The spring sowing experiment showed that a clone, derived from microspore of 4-L F_1 hybrid, was not uniform. Namely, two lines of the clone DH 4-L/1, DH 4-L/2 were very similar in studied traits /see: Table 2/ but DH 4-L/2 required cold to induce flowering and DH 4-L/1 did not /see: Table 1/. Unexpected differentation of the two doubled haploids, was heritable, indicated spontaneous variation, which took place during in vitro culture. This phenomenon is described as a somaclonal variation /Larkin and Scowcroft, 1981/ and was observed in another clone of the study. 354G clone, obtained from microspore of Jet Neuf cv. through secondary embryos, consisted of four homozygous lines: DH Ga, DH Gd, DH Ge_1, DH Ge_3. Three of them: DH Gd, DH Ge_1, DH Ge_3 were of win-

ter type with high level of glucosinolates while the fourth line: DH Ga was a spring type with low level of glucosinolates, small seeds and high content of fat /Table 3/. The somaclonal variation was checked by other authors /Hoffmann et al.,1982; Wenzel et al., 1985/, and it seems to be a source of new variability creating new genotypes of rapeseed.

As it was expected, the studied androgenetic lines showed a great differentation between themselves for the traits checked. Among 42 DHs from Jet Neuf cv. a portion of DH lines was superior, another was statistically at par with parent and the rest was inferior /Table 4/. Similarly, in 12 doubled haploids from 592 F_1 hybrids, a great distribution in the studied characters was observed/Table 5/.

The differences between dihaploids and their parent or from one parental source were due to different genotype of gamets that were segregating from hybrid male parent.

Referring to the data, we can discard the poorest DH lines from a further study, thus doubled haploid lines with desired characters can be incorporated in to rapeseed breeding program.

Conclusions

1. There were large differences among dihaploid lines for all studied characters but plants within lines appeared to be uniform.
2. Somaclonal variation, examples of which were noted in the study, can increased genetic variation of rapeseed.

References

Hoffmann F., E.Thomas, G.Wenzel, 1982. Anther culture as a breeding tool in rape.II. Progeny analyses of androgenetic lines and induced mutants from haploid culture. Theoretical and Applied Genetics, 61: 225-232.

Larkin P.J., W.P.Scowcroft, 1981. Somaclonal variation - a novel source of variability from cell cultures for plant improvement. Theoretical and Applied Genetics, 60: 197-214.

Nałęczyńska A., T.Cegielska, 1984. Dihaploids production in Brassica napus L. by in vitro androgenesis. Genetica Polonica, 25: 271-276.

Nałęczyńska A., J.Krzymanski, 1985. Siew wiosenny jako metoda testowania homozygotyczności dihaploidów rzepaku ozimego. Wyniki Badań nad Rzepakiem Ozimym, IHAR, 59-64.

Wenzel G., B.Froughi-Wehr, W.Friedt, F.Köhler, T.Oo, 1985. Cell and tissue culture as supplementary tool in plant breeding: Exemplified in potato, oilseed rape and barley. Hereditas Suppl. 3: 15-25.

Table 1. Results of experiment with spring sowing of androgenetic doubled haploids of winter rapeseed and their parent cultivars.

Dihaploids	Number of dihaploid lines			
	tested	with 100% plants with flower-shoot	with less than 80% plants with flower-shoot	no plants with flower-shoot
Jet Neuf DHs	42	40	2	-
Janpol DHs	10	10	-	-
592 hybrid DHs	12	-	-	12
4-L hybrid DHs	2	1	-	1

	Number of plants		
	tested	with flower-shoot	without flower-shoot
Jet Neuf	85	65	20
Janpol	30	22	8

Table 2. Two doubled haploids originating from one embryo of 4-L F_1 hybrid were similar in studied traits.

DH	Fat content in seeds /%/	TSW /g/	Seeds yield per plant /g/	Plant height /cm/
4-L/1	48,18	3,67	41,91	1,35
4-L/2	49,15	3,83	51,33	1,30

Table 3. The mean values of measured traits of 354 G clone derived from microspore of Jet Neuf cv.

	DH designation	Glucosinolates content O low x high	Fat content in seeds /%/	TSW /g/
	Ga	O	/+/45,44	- 2,82
	Gd	x	/-/40,50	4,71
	Ge_1	x	/-/39,73	4,65
	Ge_3	x	/-/39,16	4,53
Jet Neuf cv.		x	41,83	4,61

+ : significantly superior to the parent cultivar-p=0,05

- : significantly inferior to the parent cultivar-p=0,05

Table 4. Relative performances of individual doubled haploid lines as compared to their parental cultivar Jet Neuf for six characters evaluated in an experiment.

Characters	Number of doubled-haploid lines		
	higher[x]	in the range of parent	lower[x]
Fat content in seeds	20	12	10
1000 seeds weight	9	25	8
seeds yield per plant	7	28	7
plant height	3	32	7
no. of branches	12	30	-
height to first branch	1	14	27

x - significantly different from the parent cultivar at p=0,05.

Table 5. Range of variation in six traits of doubled haploids of the 592 F_1 hybrid winter rapeseed.

	Fat content in seeds /%/	TSW /g/	Seed yield per plant /g/	Plant height /cm/	No. of branches	Height to first branch /cm/
12 DH lines	45,7-36,8	5,4-3,6	65,1-15,1	143-112	17,3-6,9	13,2-10,0