Breeding of oilseed rape for new seed oil quality

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Introduction

Diversification of fatty acid composition is one of the main selection goals in oilseed rape (*B. napus* L.) breeding for new seed oil quality, due to the oil crop market demands. High oleic (HO, ≤ 79.5% of oleic acid in seed oil) as well as low linolenic (LL, ≤ 2.1% of linolenic acid) mutants have been obtained *via* chemical mutagenesis induced by ethyl metanesulphonate (EMS) treatment at the Plant Breeding and Acclimatization Institute—NRI in Poznan, Poland (Spasibionek 2006). New forms, HOLL have been developed lately, both by chemical mutagenesis and reciprocal crosses of HO and LL mutants.

The presented work comprises the development of new breeding forms and the improvement of their agronomic value by introducing them into a new genetic background as well as by selection in field experiments.

Material and Methods

HO, LL and HOLL mutant breeding forms were crossed with high-yielding varieties: Bojan, Cabriolet, Californium, Contact and Lisek. They were also included into hybrid breeding by crossing with *ogura* CMS and *Rfo* restorer lines. Field experiments were carried out in different environments and the seed yield, fatty acid composition, as well as oil content were estimated. The fatty acid composition of mature seeds was determined by gas liquid chromatography of the methyl esters (Byczynska and Krzymanski, 1969) using an Agillent Technologies 6890N gas chromatograph (DB 23 30 m, ID 025, 0.25 µm layer capillary column) equipped with a Chemstation integrator. The SNapShot analysis was used to identify the homozygous low linolenic genotypes (Mikolajczyk et al., 2010) and SCAR markers – to detect the *Rfo* restorer gene and the *ogura* CMS cytoplasm (Mikolajczyk et al., in press).

Results

F_{11}—F_4 recombinant inbred lines obtained as a result of crosses between the HO, LL and HOLL mutants and high-yielding varieties contained, on average: ≤ 81.7% of oleic (HO—type), ≤ 2.5% of linolenic acid (LL—type), as well as 71.0—81.7% of oleic and 3.5%—5.6% of linolenic acid (HOLL—type) in seed oil (Table 1).

Table 1. Mean values of fatty acid content (%) in seed oil of parental lines (mutants and high-yielding varieties) and F_{11}—F_4 recombinant inbred lines

Object	Oleic acid C18:1	Range	Linoleic acid C18:2	Range	Linolenic acid C18:3	Range
Parental lines						
M-10453	77.2		8.1		6.8	
M-10464	78.4		7.7		7.0	
M-681	64.2		24.9		2.8	
Bojan	60.3		21.8		10.1	
Cabriolet	64.1		18.4		8.9	
Californium	61.1		21.3		9.7	
Contact	72.2		10.8		9.8	
Lisek	63.6		19.0		9.5	
F_{11}—F_4 Recombinant inbred lines						
In total (n=129)	75.7	61.1-81.7	9.9	5.0-26.3	7.3	2.5-10.8
HO-type (n=98)	77.7	73.7-81.7	7.5	5.0-11.2	7.8	6.0-10.8
LL-type (n=7)	66.0	63.3-68.6	23.3	21.1-26.3	3.7	2.5-4.9
HOLL-type (n=8)	75.4	71.0-80.4	13.1	7.3-17.1	4.7	3.5-5.6

The mean values of seed yield of recombinant mutant lines ranged from 30.5 dt/ha to 41.4 dt/ha, as compared to reference varieties Castille (mean value of seed yield—46.8 dt/ha) and Chagall (46.0 dt/ha) and the mean values of seed oil content ranged from 46.1% to 49.4% (Chagall—48.2% and Castille—47.7%). The lines and strains selected as a result of recombinant breeding were characterized by high seed oil content—above 48% and altered seed oil fatty acid composition. They revealed high content of oleic acid, 75—79%, and 1:1 ratio of linoleic to linolenic acid content, 8% and 7.7%, respectively, which significantly improves the nutrition value of oil. Alkene glucosinolates content was very low, below 5 μmol g^{-1} seeds (Table 2).

Table 2. Comparison of seed quality and yield of some F_{11}—F_4 recombinant inbred lines of *Brassica napus*, with respect to Castille and Chagall varieties investigated in field trials in 2010

Object	Yield (dt/ha)	Fatty acids [%]			Glucosinolates (μmol g^{-1} seeds)	Fat content (%)
		$C_{18:1}$	**$C_{18:2}$**	**$C_{18:3}$**		
CASTILLE	46.8	64.4	18.1	9.8	14.2	47.8
CHAGALL	46.0	68.8	14.9	8.8	9.6	48.2
PN 841/1i/09	41.4	75.2	8.4	9.6	11.1	48.7
PN 833/3i/09	41.3	74.6	9.3	9.0	16.6	47.8
PN 835/2i/09	40.5	75.5	9.4	8.3	13.5	49.4
PN 860/4i/09	39.4	74.7	9.2	9.0	14.7	47.8
PN 1786/09	38.2	70.9	11.8	9.8	8.9	48.4
PN 840/2i/09	37.5	74.7	8.4	1.,1	11.7	48.6
PN 1914/09	37.4	75.7	9.6	8.0	8.8	49.2
PN 1821/09	37.4	75.5	9.3	7.8	10.1	47.4
PN 827/2i/09	37.1	75.1	8.5	8.5	17.7	47.8
PN 1795/09	36.6	75.8	8.2	8.7	9.4	47.2
PN 1884/09	36.6	74.5	9.8	8.2	6.5	47.4
PN 1769/09	36.0	75.7	9.0	8.5	8.1	47.2
PN 1792/09	35.9	77.3	7.7	8.0	7.8	48.4
PN 1767/09	35.9	74.8	8.8	8.8	6.8	47.5
F	**3.62****					**5.23****
NIR 0,05	**3.05**					**0.570**

The F_{11}—F_4 recombinant inbred lines of good agronomic value and high oleic (81.5—82.9%) and low linolenic acid (1.3%) content were then crossed with the *Rfo* restorer lines. As a result, recombinant restorer lines with high oleic acid content, HO—type (≤79.7%), as well as LL—type restorer lines (≤2.2% of linolenic acid) and HOLL-type restorer lines (72.4—80.8% of oleic acid and 2.5—5.8% of linolenic acid) were obtained (Table 3).

Table 3. Mean values of fatty acid content (%) in seed oil of parental lines (the F_{11}—F_4 recombinant inbred lines and restorer lines) and in F_6—F_4 recombinant restorer lines

Object	Oleic acid $C_{18:1}$	Range	Linoleic acid $C_{18:2}$	Range	Linolenic acid $C_{18:3}$	Range
Parental lines						
HO-type (n=3)	82.3	81.5-82.9	5.9	5.7-6.2	5.8	5.1-6.4
LL-type (n=2)	66.5	64.0-69.0	25.7	22.6-28.7	1.3	1.3-1.3
HOLL-type (n=3)	74.8	73.0-75.7	15.0	14.1-15.8	3.9	2.9-4.7
Restorer lines (n=4)	62.8	61.6-65.2	20.5	18.5-21.5	9.8	9.2-10.4
F_6—F_4 Recombinant restorer lines						
In total (n=88)	70.5	56.1-80.8	16.4	7.0-32.7	6.0	2.2-10.1
Type HO (n=46)	75.8	71.1-79.7	9.8	7.0-13.1	7.4	6.1-9.7
Type LL (n=33)	61.7	56.1-66.2	26.8	20.2-32.7	4.1	2.2-6.5
Type HOLL (n=7)	77.6	72.4-80.8	10.3	7.4-17.8	4.8	2,5-5,8

Backcrosses between the canola-type *ogura* male-sterile lines (59.7—65.8% of oleic acid, 18.4—21.5% of linoleic acid, 8.5—11.4% of linolenic acid and low glucosinolates content of about 9.9 μmol g^{-1} seeds) and M10453, M10464 i M681 mutant lines were performed and male-sterile BC_3 generations were obtained. HO—type *ogura* CMS lines (74.6—81.3% of oleic acid), LL-type *ogura*

CMS lines (≤2.2% of linolenic acid) and HOLL—type (71.8—74.9% of oleic acid and 2.3—3.0%) of linolenic acid were selected (Table 4).

Table 4. Mean values of fatty acid content (%) in seed oil of parental lines (mutant lines and the *ogura* CMS lines) and BC_3 *ogura* CMS lines

Object	Oleic acid $C_{18:1}$	Range	Linoleic acid $C_{18:2}$	Range	Linolenic acid $C_{18:3}$	Range
Parental lines						
M-10453	77.2		8.1		6.8	
M-10464	78.4		7.7		7.0	
M-681	64.2		24.9		2.8	
CMS *ogura* (n=3)	63.7	59.7-65.8	19.5	18.4-21.5	9.5	8.5-11.4
BC_3 *ogura* CMS lines						
In total (n=46)	71.3	57.6-81.3	14.7	6.0-28.2	6.2	2.2-11.3
Type HO (n=17)	78.1	74.6-81.3	7.4	6.0-9.6	7.4	5.6-8.8
Type LL(n=10)	66.3	59.8-69.5	22.5	18.9-28.2	3.2	2.2-3.9
Type HOLL (n=4)	73.8	71.8-74.9	15.9	15.0-18.0	2.7	2.3-3.0

Summary

As a result of crosses between the HO, LL and HOLL mutant lines and high-yielding varieties of oilseed rape, recombinant lines were obtained revealing good agronomic value and changed fatty acid composition.

F1 hybrid components, recombinant *Rfo* restorer and *ogura* CMS lines with differentiated seed oil fatty acid composition were developed.

The obtained breeding material will be evaluated in field trials in order to develop new cultivars and hybrid varieties.

Specific genetic markers were useful for monitoring the presence of the *Rfo* restorer gene, *ogura* male sterile cytoplasm and low linolenic mutant forms and they will be further applied for MAS in breeding programs.

References

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