

COMPOSITION AND CORRELATION STUDIES OF FATTY ACIDS IN SEED OIL OF YELLOW SARSON (*BRASSICA CAMPESTRIS* L.) CULTIVARS AND BACKCROSS DERIVED ZERO ERUCIC ACID YELLOW SARSON POPULATIONS

M.H. RAHMAN^{1*}, O. STØLEN¹, L. RAHMAN² and M.M. RAHMAN²

¹*Department of Crop Science, Royal Veterinary and Agricultural University, DK-1871 Frederiksberg-C, Denmark.*

²*Department of Genetics and Plant Breeding, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh.*

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Abstract: The Yellow Sarson (*Brassica campestris*) cultivars of Bangladesh origin contain a high level (54%) of erucic fatty acid (C_{22:1}). In the seeds of backcross derived zero-erucic acid Yellow Sarson populations, absence of erucic acid caused a simultaneous reduction in the content of arachidic (C_{20:0}), eicosenoic (C_{20:1}) and behenic (C_{22:0}) fatty acids, with an increase in palmitic (C_{16:0}), oleic (C_{18:1}), linoleic (C_{18:2}) and linolenic (C_{18:3}) acids. The correlations of C₁₆ (palmitic) and C₁₈ (oleic and linoleic) fatty acids with all other C₂₀ (arachidic and eicosenoic) and C₂₂ (behenic and erucic) fatty acids were negative. Palmitic acid had positive correlations with oleic and linoleic acids. Among the C₁₈ fatty acids, oleic acid was positively correlated with linoleic acid and linoleic acid was positively correlated with linolenic acid. On the other hand, all C₂₀ and C₂₂ fatty acids were positively correlated with each other.

Key words: Backcross, *Brassica campestris*, fatty acids, yellow sarson oils

INTRODUCTION

The seed oil of *Brassica* cultivars, including the recently released Yellow Sarson (*B. campestris* L.), grown in Bangladesh, contain a high level of erucic fatty acid^{1,2}. The presence of erucic acid in oil is nutritionally undesirable for edible purposes, because it was observed that high content of erucic acid had adverse effects on the body of the experimental animals through changes in heart tissue, deposits of fatty acids in the muscles and myocardial lesions³. A backcross breeding programme was launched to develop erucic acid free Yellow Sarson varieties⁴. This programme resulted in a number of zero erucic acid Yellow Sarson lines⁵.

Correlation between fatty acids could be different in rape varieties adapted to different geographical locations. In Asian varieties, linolenic acid appears to be independently variable from other fatty acids¹⁰. Many authors have studied correlation between different fatty acids in *Brassica* seed oil⁶⁻¹¹. These studies were mainly done on *B. napus*. No study on Yellow Sarson *B. campestris*, including all major and minor fatty acids, could be found in available literature. Therefore

* Present address: Danisco Seed, 14 Højbygårdvej, DK-4960 Holeby, Denmark.

the present study was conducted to study the correlation between different fatty acids, using the mean data of Yellow Sarson parents and zero erucic acid selfed progeny of first backcross generation (BC_1S_1) and acid selfed progeny of second backcross generation (BC_2S_1) seed populations. Fatty acid composition of zero erucic acid Canadian cultivar Tobin is given for comparison.

METHODS AND MATERIALS

The materials used in the study were two high erucic acid Yellow Sarson *B. campestris* cultivars Sampad and the breeding line M-91, one brown-seeded zero-erucic Canadian *B. campestris* cultivar Tobin and four of each zero-erucic BC_1S_1 and BC_2S_1 seed populations. The following breeding technique was followed for the development of the zero erucic acid BC_1S_1 and BC_2S_1 seed populations.

Initial crosses were made between Sampad x Tobin and M-91 x Tobin. F_1 plants were reciprocally backcrossed with the respective Yellow Sarson parent and the following four backcross populations were generated:

(Sampad x Tobin) x Sampad
 Sampad x (Sampad x Tobin)
 (M-91 x Tobin) x M-91
 M-91 x (M-91 x Tobin)

First backcross (BC_1) seeds heterozygous for erucic acid alleles were identified and plants were grown. The BC_1 plants were selfed to obtain BC_1S_1 seeds as well as backcrossed with the respective Yellow Sarson parent to obtain BC_2 seeds. The erucic heterozygous BC_2 seeds were identified and plants were grown to obtain BC_2S_1 seeds. Zero erucic acid seeds were selected from both BC_1S_1 and BC_2S_1 generations of the above-mentioned four backcrosses. Generation means of the four backcrosses for the two generations (BC_1S_1 and BC_2S_1) were used in the study. The details of the above-mentioned breeding technique is described elsewhere⁵.

Fatty acid composition of seed oil was determined following gas chromatographic analysis of methyl ester of fatty acids. The details of the method are described elsewhere^{2,5,12}.

RESULTS AND DISCUSSION

Fatty acid composition

Erucic acid ($C_{22:1}$) content of Sampad and M-91 was approximately 54%, while that of the Canadian cultivar Tobin was almost zero. Compared to Tobin, the Yellow Sarson parents had a higher percentage of stearic ($C_{18:0}$), arachidic ($C_{20:0}$), eicosenoic ($C_{20:1}$) and behenic ($C_{22:0}$) acids, but a lower quantity of palmitic ($C_{16:0}$), oleic ($C_{18:1}$), linoleic ($C_{18:2}$) and linolenic ($C_{18:3}$) acids (Table 1 and 2).

Table 1: Fatty acids in the seed oil of parents and zero erucic acid BC₁S₁ and BC₂S₁ seeds of the cross between Sampad and Tobin.

Parents and backcross generation	Fatty acid (%)									
	Palmitic C _{16:0}	Stearic C _{18:0}	Oleic C _{18:1}	Linoleic C _{18:2}	Linolenic C _{18:3}	Arachidic C _{20:0}	Eicosenoic C _{20:1}	Behenic C _{22:0}	Erucic C _{22:1}	
Sampad (S)	1.69	1.58	14.50	10.36	7.85	1.59	7.40	1.52	53.51	
Tobin (T)	2.87	0.93	61.62	20.59	12.35	0.20	1.30	0.10	0.00	
BC ₁ S ₁ : (SxT) xS	2.67	1.59	66.34	16.79	10.91	0.50	1.07	0.20	0.00	
BC ₁ S ₁ : Sx(SxT)	2.25	1.54	69.60	14.89	9.74	0.54	0.33	1.11	0.00	
BC ₂ S ₁ : ((SxT)xS)xS	2.43	1.95	71.15	13.55	9.39	0.41	1.10	0.02	0.00	
BC ₂ S ₁ : Sx(Sx(SxT))	2.49	1.61	68.86	15.70	10.04	0.22	1.08	0.00	0.00	

Note:- BC₁S₁ = Selfed progeny of first backcross generation,
 BC₂S₁ = Selfed progeny of second backcross generation.

Table 2 : Fatty acids in the seed oil of parents and zero erucic acid BC_1S_1 and BC_2S_1 seeds of the cross between M-91 and Tobin.

Parents and backcross generation	Fatty acid (%)									
	Palmitic $C_{16:0}$	Stearic $C_{18:0}$	Oleic $C_{18:1}$	Linoleic $C_{18:2}$	Linolenic $C_{18:3}$	Arachidic $C_{20:0}$	Eicosenoic $C_{20:1}$	Behenic $C_{22:0}$	Erucic $C_{22:1}$	
M-91(M)	1.71	1.64	13.50	11.44	7.56	1.49	6.49	1.82	54.35	
Tobin (T)	2.87	0.93	61.62	20.59	12.35	0.20	1.30	0.10	0.00	
$BC_1S_1 : (M \times T) \times M$	2.68	2.06	68.57	15.54	8.68	0.70	1.29	0.50	0.00	
$BC_1S_1 : M \times (M \times T)$	2.41	1.57	70.01	14.59	9.36	0.57	1.31	0.18	0.00	
$BC_2S_1 : ((M \times T) \times M) \times M$	2.74	0.02	73.63	13.01	6.91	0.41	1.15	0.09	0.00	
$BC_2S_1 : M \times (M \times (M \times T))$	2.59	1.81	71.27	14.41	8.23	0.41	1.28	0.00	0.00	

Note :- BC_1S_1 = Selfed progeny of first backcross generation,
 BC_2S_1 = Selfed progeny of second backcross generation.

In zero-erucic BC_1S_1 and BC_2S_1 seeds, the percentage of palmitic, oleic, eicosenoic and behenic acids were comparable to Tobin, while stearic acid percentage was comparable to Yellow Sarson. The percentage of linoleic and arachidic acids were intermediate of Yellow Sarson and Tobin. Thus, an absence of erucic acid in BC_1S_1 and BC_2S_1 seeds caused a simultaneous reduction in arachidic, eicosenoic and behenic acids and an increase in palmitic, oleic and linoleic acids. Linolenic acid content in BC_1S_1 and BC_2S_1 seeds of Sampad x Tobin crosses lay in between the values of the two parents, while for M-91 x Tobin cross it closely corresponds to the value of M-91.

Correlation between fatty acids

Coefficients of correlation of C_{16} and C_{18} fatty acids (palmitic, stearic, oleic, linoleic and linolenic) with all C_{20} and C_{22} fatty acids (arachidic, eicosenoic, behenic and erucic) were negative, while it was significant only for palmitic, oleic and linoleic vs. all C_{20} and C_{22} fatty acids (Table 3). The $C_{16:0}$ palmitic fatty acid also had significant positive correlation with $C_{18:1}$ and $C_{18:2}$ fatty acids (oleic and linoleic). A positive correlation of palmitic acid with oleic acid was reported in *B. napus*¹⁰. However, a negative correlation between these two fatty acids was reported elsewhere⁹. Negative correlations of oleic acid with eicosenoic and erucic acid have also been reported⁶⁻⁸.

Among the C_{18} fatty acids (viz. stearic, oleic, linoleic and linolenic), significant positive correlations were found only for oleic vs. linolenic and linoleic vs. linolenic acids. The positive correlation between linoleic and linolenic acid indicates that it would be difficult to reduce the level of linolenic acids alone by conventional breeding. Positive correlation between linoleic and linolenic acids have also been reported⁷⁻¹¹. However, a mutant *B. napus* line with 2-20% linolenic acid was selected by chemical mutagenesis¹³. Low linolenic acid *B. napus* line (3.4%) was developed from interspecific cross of *B. juncea* x *B. napus*¹⁴.

All C_{20} and C_{22} fatty acids (viz. arachidic, eicosenoic, behenic and erucic), are positively correlated with each other. Significant positive correlation between eicosenoic and erucic acid in *B. napus* has also been reported⁸.

In summary, the results show that correlation of oleic acid ($C_{18:1}$) with all other C_{20} fatty acids is negative, but it is positive with $C_{18:2}$ fatty acid. On the other hand, $C_{18:2}$ fatty acid is positively correlated with $C_{18:3}$. All C_{20} and C_{22} fatty acids are positively correlated with each other. These results are in agreement with the biosynthetic pathway of the major fatty acids from oleic acid as proposed by many workers^{6,15,16}: that eicosenoic ($C_{20:1}$) and erucic ($C_{22:1}$) acids are formed from oleic acid by chain lengthening process and linoleic ($C_{18:2}$) and linolenic ($C_{18:3}$) acids are formed by successive desaturation of oleic acid.

Table 3: Correlation coefficients between different fatty acids involving Yellow Sarson parents and zero erucic acid BC₁S₁ and BC₂S₁ populations of the crosses of Sampad x Tobin and M-91 x Tobin of *B. campestris*.

Fatty acid	Fatty acid (Correlation coefficient [†])							
	Stearic C _{18:0}	Oleic C _{18:1}	Linoleic C _{18:2}	Linolenic C _{18:3}	Arachidic C _{20:0}	Eicosenoic C _{20:1}	Behenic C _{22:0}	Erucic C _{22:1}
Palmitic, C _{16:0}	0.519	0.921**	0.782**	0.314	-0.884	-0.887**	-0.890**	-0.922**
Stearic, C _{18:0}		0.366	0.023	-0.425	-0.277	-0.284	-0.400	-0.328
Oleic, C _{18:1}			0.789**	0.432	-0.964**	-0.985	-0.874**	-0.997**
Linoleic, C _{18:2}				0.765**	-0.780**	-0.842**	-0.658*	-0.834**
Linolenic, C _{18:3}					-0.480	-0.515	-0.355	-0.494
Arachidic, C _{20:0}						0.957**	0.901**	0.962**
Eicosenoic, C _{20:1}							0.805**	0.989**
Behenic, C _{22:0}								0.867**

[†]df=8, *p<0.05, **p<0.01

Note :- BC₁S₁ = Selfed progeny of first backcross generation, BC₂S₁ = Selfed progeny of second backcross generation.

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References

- 1 Rahman L. & Quddus M.A. (1981). Variability of oil content, fatty acid pattern and glucosinolate content in mustard and rapeseed. *Indian Journal of Agricultural Science* **51**: 23-26.
- 2 Møller P., Rahman M.H., Stølen O. & Sørensen H. (1985). Heredity of fatty acids and glucosinolates in oilseed rape. Possibilities for improvement of rape adapted for the growth conditions in tropical Asia. In: *Advances in the production and utilisation of cruciferous crops* (Ed. H. Sørensen). pp 286-300. Martinus Nijhoff Publ., Dordrecht.
- 3 Vles R.O. (1974). Nutritional aspects of rapeseed oils. *Proceedings of the 4th International Rapeseed Conference* pp 17-30. Giessen, W. Germany.
- 4 Downey R.K. & Harvey B.L. (1963). Methods of breeding for oil quality in rape. *Canadian Journal of Plant Science* **43**: 271-275.
- 5 Rahman M.H., Stølen O., Sørensen .H. & Rahman L. (1996). Recurrent backcross: a method for transferring erucic acid allele into *Brassica* oilseed cultivars, *B. campestris* L. Yellow Sarson as an example. *Acta Agriculturae Scandinavica, Sect. B, Soil and Plant Science* **46** : 68-73.
- 6 Downey R.K. & Craig B.M. (1964). Genetic control of fatty acid biosynthesis in rapeseed (*Brassica napus* L.). *Journal of the American Oil Chemists Society* **41**: 475-478.
- 7 Lööf B. & Appelqvist L-Å. (1964). Breeding work in rape, turnip rape and white mustard in connection with research on the composition of the fatty acids in their seeds. *Zeitschrift fuer Pflanzenzüchtung* **52**: 113-126.
- 8 Krzymanski J. & Downey R.K. (1969). Inheritance of fatty acid composition in winter forms of rapeseed, *Brassica napus*. *Canadian Journal of Plant Science* **49**: 313-319.
- 9 Stefansson B.R. & Storgaard A.K. (1969). Correlations involving oil and fatty acids in rapeseed. *Canadian Journal of Plant Science* **49**: 573-580.

- 10 Lee J.I., Takayanagi K. & Shiga T. (1974). Breeding for improvement of fatty acid composition in rapeseed, *Brassica napus* L. I. Fatty acid composition in rapeseed oil of Asian and European varieties. *Bulletin of National Institute of Agricultural Science Series D*, No. 25: 1-16 (In Japanese with English summary).
- 11 Kondra Z.P. & Thomas P.M. (1975). Inheritance of oleic, linoleic and linolenic acids in seed oil of rapeseed (*Brassica napus*). *Canadian Journal of Plant Science* 55: 205-210.
- 12 Rahman M.H., Rahman L., Stølen O. & Sørensen H. (1994). Inheritance of erucic acid content in yellow- and white-flowered Yellow Sarson x Canadian *Brassica campestris* L. *Acta Agricultural Scandinaviana Secretariate B, Soil and Plant Science* 44 : 94-97.
- 13 Röbbelen G. & Nitsch A. (1975). Genetical and physiological investigation on mutants of polyenoic fatty acids in rapeseed (*Brassica napus* L.). I. Selection and description of new mutants. *Zeitschrift fuer Pflanzenzeuchtung* 75 : 93-105.
- 14 Roy N.N. & Tarr A.W. (1986). Development of near-zero linolenic acid (18:3) lines of rapeseed (*Brassica napus*). *Zeitschrift fuer Pflanzenzeuchtung* 96:218-223.
- 15 Cherif A., Dubacq J.P., Mache R., Oursel A. & Tremolieres A. (1975). Biosynthesis of α -linolenic acid by desaturation of oleic and linoleic acids in several organs of higher and lower plants and in algae. *Phytochemistry*, 14: 703-706.
- 16 Agrawal V. P. & Stumpf P. K. (1985). Elongation systems involved in the biosynthesis of erucic acid from oleic acid in developing *Brassica juncea* seeds. *Lipids* 20: 361-366.