

SYMPOSIUM:
MODIFICATION OF OILSEEDS THROUGH
PLANT BREEDING

conducted by The American Oil Chemists' Society

at its 41st Fall Meeting, Chicago, Illinois

October 15-18, 1967

C. G. YOUNGS, Program Chairman

Breeding Rapeseed for Oil and Meal Quality^{1,2}

R. K. DOWNEY, Research Station, Canada Agriculture, University Campus, Saskatoon, Saskatchewan, Canada,
B. M. CRAIG and C. G. YOUNGS, Prairie Regional Laboratory of National Research Council, Saskatoon,
Saskatchewan, Canada

Abstract

Significant variation in fatty acid composition occurs within the seed oils of the *Brassica* genus, which includes the mustards and rapeseed. Research into the inheritance and biosynthesis of fatty acids has shown that at least two biosynthetic pathways exist in the developing rapeseed and some of the steps are under direct genetic control. The plant breeder has the basic knowledge in this oilseed crop to produce seed oils with defined fatty acid composition, and a practical example is the commercial development of Canbra oil, the rapeseed oil from which erucic acid has been eliminated. *Brassica* seed meals contain thioglucosides which may cause metabolic disturbances when fed to certain classes of livestock. The major thioglucosides in rapeseed meal are those giving rise to 3-butenyl and 4-pentenyl isothiocyanate and 5-vinyl-2-oxazolidinethione. Partial success in eliminating these compounds has been achieved by breeding strains of turnip rape (*B. campestris*) which do not contain the glucosides of 4-pentenyl isothiocyanate and oxazolidinethione, and the identification of a *B. napus* variety with very low levels of all three glucosides. These findings suggest that complete removal of these sulfur compounds may be possible through plant breeding.

Introduction

Plant breeding is moving at an accelerated pace, in step with other sciences. A few short years ago it took 15 to 20 years to develop a new variety. Today this can be shortened to 7 to 8 years, which still includes at least 3 years of extensive field tests. The success of a plant breeding program depends on the variation that is present or that can be induced in the crop and its close relatives, and accurate means of identifying the characteristic of interest. These principles apply to quality as well as quantity.

The application of gas chromatographs, integrators, computers, controlled environments and visible, ultraviolet, infrared and NMR spectroscopy to crop quality breeding has resulted in an extremely rapid genetic advance. Fortunately in oilseed crops many of the quality factors of major economic importance are relatively simply inherited. These developments have tended to make breeding for quality a more efficient process than breeding for quantity or yield. Much credit for the present advance must be given to the chemist, for without the new analytical techniques the breeder would be faced with an impossible task.

Rapeseed (*Brassica campestris* and *B. napus*) is Canada's most important oilseed crop, occupying 1.7 million acres in Western Canada. More rapeseed is exported from Canada than all other countries combined. Canadian utilization of rapeseed oil and meal is surpassed only by soybeans. To make rapeseed even more valuable and versatile requires that the chemistry of the plant be changed. This type of breeding is possible because of the new chemical techniques developed and the great chemical and morphological variation present in the *Brassica* genus.

INDEX

- 121-123 BREEDING RAPESEED FOR OIL AND MEAL QUALITY, by R. K. Downey, B. M. Craig and C. G. Youngs
- 124-125 SOME PROBLEMS IN ALTERING THE GOSSYPOL CONTENT OF COTTONSEED THROUGH BREEDING, by Joshua A. Lee
- 126-129 VARIATION IN COMPOSITION OF SUNFLOWER OIL FROM COMPOSITE SAMPLES AND SINGLE SEEDS OF VARIETIES AND INBRED LINES, by E. D. Putt, B. M. Craig and R. B. Carson
- 130-132 MODIFICATION OF QUANTITY AND QUALITY OF SAFFLOWER OIL THROUGH PLANT BREEDING, by P. F. Knowles

¹ Contribution No. 309, Research Station, Canada Department of Agriculture, Saskatoon, Saskatchewan, Canada.

² Presented at the AOCs Meeting, Chicago, October 1967.

TABLE I

Per Cent Fatty Acid Composition of the Standard Rapeseed Varieties Tanka and Echo, and Selections and Varieties With Extreme Values

Species and variety	Carbon chain length and degree of saturation								
	16:0	16:1	18:0	18:1	18:2	18:3	20:0	20:1	22:1
<i>B. napus</i>									
Tanka	3.7	0.4	1.2	15.7	15.2	9.3	0.9	9.2	44.4
Nugget	3.3	0.2	1.5	22.5	12.2	5.4	t	14.2	40.4
Oro	4.8	0.5	2.4	63.1	19.4	9.0	t	0.8	0
<i>B. campestris</i>									
Echo	4.5	0.3	1.3	33.3	20.4	7.6	0.5	9.4	23.0
Yellow									
Sarson	1.8	0.4	0.8	11.7	10.5	8.3	t	5.9	61.0
Selection	3.0	0.3	1.1	54.8	31.1	9.7	0	0	0

Included in this genus are the crop plants: cabbage, kale, turnips, mustards, and forage and oilseed rape. Plants of most of these species readily intercross and characteristics of interest can be transferred.

Oil Quality

The fatty acid composition of rape and other *Brassica* seed oils differs from other vegetable oils in having substantial amounts of the fatty acids eicosenoic (6–15%) and erucic (20–61%). These acids are not undesirable in human nutrition but their presence may limit the usefulness of the oil in some products. Inbreeding and selection for low erucic acid values resulted in strains of *B. napus* and *B. campestris* which did not contain erucic acid in their seed oil (Table I) (5,10). Rape oils essentially free of erucic have been named Canbra oil, a contraction of the words Canada and Brassica. Reciprocal crosses between plants containing no erucic and normal high erucic acid types demonstrated that the fatty acid composition was controlled by the genetic constitution of the developing embryo, rather than the maternal parent (8). Genetic investigations supported the hypothesis that erucic acid content of the seed oil of *B. napus* is controlled by two genes and in *B. campestris* by one gene (4,9). In both species gene action was additive with no dominance. The different inheritance pattern in the two species was expected since *B. napus* is an amphidiploid and *B. campestris* is a diploid.

The isolation of plants containing no erucic acid in their seed oil gave simultaneous selection for low eicosenoic acid. As the genetic capacity for erucic and eicosenoic acids decreased there was an increase in the percentage of oleic acid, with no decrease in total oil content. Evidence from oil analysis following

pod injections with radioactive sodium acetate supported the hypothesis that in the normal embryo an acetate is added to the carboxylic end of oleic to form eicosenoic and a second acetate is added to eicosenoic to form erucic acid (7).

A backcross breeding program was initiated to introduce the zero erucic acid character into strains adapted to Western Canada. By 1963 agronomically acceptable strains had been selected. These were used in experimental field increases undertaken by the Saskatchewan Wheat Pool, Vegetable Oil Division, in cooperation with the Canada Department of Agriculture, to determine the production feasibility and market potential of this new kind of oil. Canada Packers Co. Ltd. evaluated the Canbra oil obtained and concluded that in the manufacture of partially hydrogenated, winterized salad oils, Canbra had substantially lower stearin losses than normal rapeseed or soybean oils (11). In addition, Canbra was found to be equal to these oils in the manufacture of margarines and shortenings. Canbra oil from over 8,000 acres will be utilized in the Canadian domestic market in 1967–68.

Rapeseed oil, such as that from Indian yellow Sarson varieties, contains a high level of erucic acid (Table I), and may have an industrial market. Selected strains of yellow Sarson would appear to be a more economical source of erucic acid than *Crambe* since the problems associated with seed transportation and meal utilization are considerably less. Other rapeseed fatty acids may also be varied by selection. It is possible through plant breeding to vary the level of palmitic acid from 2.8% to 10.0%, of oleic from 7.8% to 78.0%, and of linoleic from 9.3% to 35.5%, and to reduce linolenic acid to approximately 4%. Economically the most important change that could be brought about in rapeseed oil for edible uses is the reduction of linolenic acid to less than 3%. This is presently under investigation (6).

Meal Quality

Rapeseed meal is extensively used as a high protein feed supplement and as such is an important by-product of the rapeseed crushing industry. In the development of nearly every oilseed meal, nutritional problems have been encountered. Soybean has a trypsin inhibitor, cotton seed has gossypol, and flax has a cyanogen factor. Rapeseed meal is no exception. The literature is well documented with the metabolic upsets resulting when improperly processed rapeseed

TABLE II

Normal Ranges of Thioglucosides in *Brassica* Seeds Expressed as Milligram per Gram Oil-Free Meal of Their Hydrolysis Products, p-OH Benzyl, Allyl, 3-Butenyl and 4-Pentenyl Isothiocyanates and Oxazolindimethione (OZT)

Brassica species	Crop name	Isothiocyanate				
		p-OH	allyl	butenyl	pentenyl	OZT
<i>B. hirta</i> ^a	Yellow mustard	12–20	0	0	0	0
<i>B. arvensis</i> ^a	Wild mustard					
<i>B. juncea</i>	Brown or leaf mustard	0	7–15	0	0	0
<i>B. nigra</i>	Black mustard					
<i>B. carinata</i>	Abyssinian mustard					
<i>B. oleracea</i> ^b	Kale, cabbage, etc.	0	1–9	0–4	0	0–7
<i>B. napus</i>	Rape	0	0	1–4	0.5–1.5	4–12
<i>B. campestris</i>	Turnip rape	0	0	1–3	1–3	0.5–3
	Indian yellow Sarson	0	0	8–16	0	0

^a Personal communication from R. L. Wetter, Prairie Regional Laboratory of National Research Council, University of Saskatchewan, Saskatoon, Sask.

^b After Ettlinger, M. G., and C. P. Thompson, "Studies of mustard oil glucosides (II)," Final Rept. Contr. DA 19-129-QM-1689, Office of Technical Services, U.S. Dept. Commerce (1962).

TABLE III

Thioglucoside Content of Standard Rapeseed Varieties and Selections Expressed as Milligram per Gram Oil-Free Meal of Their Hydrolysis Products, 3-Butenyl and 4-Pentenyl Isothiocyanate and Oxazolidinethione (OZT)

Species and variety	Isothiocyanate		OZT
	Butenyl	Pentenyl	
<i>B. napus</i>			
Tanka	2.5	0.6	11.5
Nugget	1.7	0.3	7.8
Bronowski	0.3	t	0.3
<i>B. campestris</i>			
Echo and Arlo	2.0	1.5	1.7
Sel. OZT	2.4	1.6	0
Sel. pentenyl	1.1	0	1.0
Sel. pentenyl + OZT	2.1	0	0

meal is fed to poultry and swine (3). The undesirable principles in rapeseed meal are derived mainly from the thioglucosides which yield isothiocyanates, oxazolidinethiones and nitriles upon enzyme hydrolysis. These compounds when fed to nonruminants cause enlarged thyroid and adversely affect growth and reproduction. Although over 40 thioglucosides have been reported in plants only three major ones have been identified in the two rapeseed species: gluconapin, glucobrassicinapin and progoitrin, which on hydrolysis with the enzyme myrosinase give rise to 3-butenyl and 4-pentenyl isothiocyanates and 5-vinyl-2-oxazolidinethione, respectively.

Intact thioglucosides are relatively harmless (1,2). Thus inactivating the enzyme during seed processing largely overcomes the nutritional problem. However, there is always the danger of reintroducing the enzyme with other feeds. The ultimate solution is therefore to eliminate the thioglucosides by plant breeding or by a combination of plant breeding and processing methods.

Brassica seeds vary markedly in the kind and amount of thioglucoside present (Table II). This suggests that elimination of the thioglucosides in rapeseed might be achieved either by interspecific crosses between mustard and rape or by selection and inbreeding within the two rape species. The development of a fast, accurate method for quantitative and

qualitative determination of isothiocyanates and oxazolidinethione by Youngs and Wetter (12) made such a breeding program feasible. Results from the interspecific crosses are not yet available but selection has produced strains with extremely low levels of thioglucosides in the seed. Within the *B. napus* species, plants of the variety Bronowski were found to have extremely low levels of all three major compounds (Table III). In the *B. campestris* species, selection within the varieties Echo and Arlo, adapted to Western Canada, resulted in isolation of plants with seed free of the thioglucosides, giving rise to 4-pentenyl isothiocyanate, oxazolidinethione or both (Table III). Genetic investigation on these two compounds in *B. campestris* showed that they are independently inherited and that their absence is recessive in character. Results to date suggest that complete removal of all thioglucosides from both rapeseed species may be possible.

Crop improvement is a never-ending cycle. Much has been done to improve rapeseed but much remains to be accomplished. Cooperation of researchers in several fields has been and will continue to be the major ingredient for success. Future prospects for major improvement appear bright. If the overall program achieves success, rapeseed could mean to Canadian agriculture what the soybean crop has meant to the United States.

REFERENCES

1. Bell, J. M., *J. Animal Sci.* **24**, 1147-1151 (1965).
2. Belzile, R., J. M. Bell and L. R. Wetter, *Can. J. Animal Sci.* **43**, 169-173 (1963).
3. Bowland, J. P., D. R. C. Clandinin and L. R. Wetter, *Can. Dept. Agr. Publ.* 1257 (1965).
4. Dorrell, D. G., and R. K. Downey, *Can. J. Plant Sci.* **44**, 499-504 (1964).
5. Downey, R. K., *Can. J. Plant Sci.* **44**, 295 (1964).
6. Downey, R. K., *Qualitas Plantarum et Materiae Vegetabiles* **9**, 171-180 (1966).
7. Downey, R. K., and B. M. Craig, *JAOS* **41**, 475-478 (1964).
8. Downey, R. K., and B. L. Harvey, *Can. J. Plant Sci.* **43**, 271-275 (1963).
9. Harvey, B. L., and R. K. Downey, *Ibid.* **44**, 104-111 (1964).
10. Stefansson, B. R., F. W. Hougen and R. K. Downey, *Ibid.* **41**, 218-219 (1961).
11. Teasdale, B. F., (Canada Packers Ltd.), *Can. Pat.* 726,140 (1966).
12. Youngs, C. G., and L. R. Wetter, *JAOS* **44**, 551-554 (1967).

[Received May 28, 1968]