SYMPOSIUM: RAPESEED MARKETING AND BREEDING

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Influence of Genetics, Environment and Admixtures on Low Erucic Acid Rapeseed in Canada[1,2]

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ABSTRACT

Single plant isolates in the *Brassica napus* and *Brassica campestris* species of rapeseed yielded glyceride oil containing small amounts of erucic acid. Agronomically suitable varieties were grown commercially in 1971 as the first phase in a changeover of Canadian rapeseed production from varieties with erucic contents of 20-45% to low erucic acid varieties. A program to monitor the erucic content by gas chromatographic analysis in the stages of production, handling and transportation from seed to export shipment was undertaken to evaluate the effects of genetics, environment and admixture. The individual increase in erucic content ranged from 0.5 to 1.0, resulting in total increases of 1-2%.

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[2]One of six papers presented in the symposium "Rapeseed Marketing and Breeding," AOCS Meeting, Ottawa, September 1972.

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INTRODUCTION

Rapeseed production in Canada has gone through a number of growth and utilization phases since the introduction of the crop in 1942. The initial production was to supply rapeseed oil as a lubricant for reciprocating steam engines. The termination of hostilities and postwar conversion from steam to diesel power brought about the demise of this market. Interest was then turned to the domestic vegetable oil market to replace some of the imported oils used for production of cooking and salad oils, shortenings and margarine. Surplus wheat production favored the development of rapeseed as an alternate crop. The successful initial experiences in rapeseed production in Western Canada indicated that acreage could be increased to serve both a domestic and an export market. The history of rapeseed production and utilization in Canada has been reviewed in previous publications (1,2).

Nutritional research on rapeseed oil in the 1950's with experimental animals indicated reduced food consumption and growth rate when the oil was used as the sole dietary source of fat (3-6). These effects were attributed to erucic acid and, along with other observations, led to decisions to conduct further research on nutrition and at the same time to study the variation of erucic acid in rapeseed with a view to the possible production of oils that had lower erucic acid contents. Subsequent nutritional studies showed that growth and food intake were more related to the low level of saturated acids than to erucic content (7). The plant breeding and selection program led to the isolation in 1961

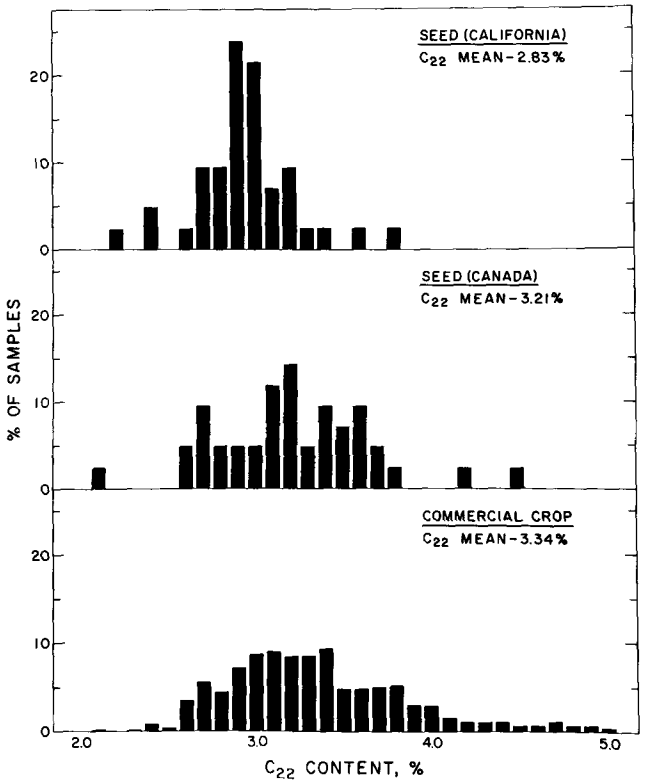

FIG. 1. Comparison of distribution of C_{22} fatty acid contents of oil from seeds of Span LEAR variety including California parent seed, Canadian seed produced in 1971 and 1971 commercial crop.

of rapeseed plants from *B. napus*, yielding an oil with essentially no erucic or eicosenoic acid (8), and in 1964 of similar plants from *B. campestris* (9). This opened the way for studies on inheritance of erucic acid in the *B. napus* species (10) and the development of suitable varieties. Further nutritional evaluations of rapeseed oil during the late 1960's in Holland, France and Canada (11-14) indicated that erucic acid accumulated in the hearts of weanling experimental animals. These considerations led to a decision to change the Canadian rapeseed from the varieties in production to varieties with oil containing low levels of erucic acid.

The commercial varieties of rapeseed grown in the producing areas of Western Canada up to 1970 yielded glyceride oils with erucic acid contents of 25-45%; varieties of *Brassica campestris* ranged from 25 to 35% and those of *Brassica napus* from 40 to 45%. Studies were carried out in 1959 and 1961 (15,16) on the fatty acid composition of varieties grown at different locations in Western Canada, and data from the 1961 study are summarized in Table I. The maximum variation for individual fatty acids occurred with oleic and erucic acids for both varietal and station means. In the case of erucic acid the variation for varietal means was 18% and the station or environmental variation was one-third of that or ca. 6%. The variations by station were similar for both species of rapeseed. Accordingly some variation might be anticipated for low erucic acid rapeseed varieties (LEAR varieties) if the erucic contents were above zero, e.g., 2-5%. In addition, varietal trials are carried out under controlled experimental production, whereas commercial production may provide the opportunity for cross-pollination and admixture due to "volunteer" crop, which could increase the variation in erucic content of the harvested seed.

No similar study has been carried out with LEAR varieties. However some seed of a *B. napus* selection was multiplied in the early stages of varietal development to provide rapeseed oil for laboratory and commercial evaluation. The stages of development by years were as follows: 1961–first report of rapeseed plants yielding oil with essentially no erucic acid; 1964–experimental plot production of 55 kilos of seed to produce oil for laboratory scale evaluation; 1965–commercial production of 136,000 kilos of seed to provide oil for commercial evaluation; 1966–expanded commercial production to produce five tank cars of oil for commercial evaluation; 1967–expanded commercial production to produce 60 tank cars of oil for commercial evaluation.

The rapeseed was grown under contract with selected farmers who were able to provide clean land and to isolate the fields from the other varieties of rapeseed. These conditions were imposed to minimize admixture from the volunteer "normal" rapeseed and reduce the effects of cross-pollination. The rapeseed was processed in one plant, with precautions taken to flush out the extraction system using the zero erucic acid rapeseed. Samples of oil were analyzed by gas chromatography (GC) to monitor the system, and data presented in Table II on the 1966 production indicate the increase in erucic acid content of the oil.

The erucic acid content of the seed remained constant during the seed multiplication in 1965, and the commercial crop produced in 1966 under controlled conditions showed a very modest increase. There were further increases of erucic acid in the extracted oil and in the tank car shipments, which had to be due to inadvertent admixture in the extraction plant, which was processing and handling the regular rapeseed oil with erucic contents ca. 30%.

The varieties of rapeseed grown in the producing areas of Western Canada for over 25 years had yielded oils with erucic acid contents of 20-45%. The new low erucic acid rapeseed (LEAR) varieties were not distinguishable on the basis of the plants in the field or the seed harvested from the plants. Accordingly, any changeover to production of LEAR rapeseed required controlled production and a monitoring system. Seed would be supplied under contract to the farmers and some isolation would be required to

TABLE I

Data[a] Showing Variation in Fatty Acid Composition Due to Genetics and Environment

	Fatty acid (% of total)						
Need heading	16:0	18:0	18:1	18:2	18:3	20:1	22:1
Varietal means							
Highest value	3.8	1.6	32.9	17.5	9.0	12.4	42.4
Lowest value	2.7	1.4	16.9	13.7	8.7	11.2	24.2
Difference	1.1	0.2	16.0	3.8	0.3	1.2	18.2
Station means							
Highest value	3.7	1.8	26.2	16.3	10.1	13.3	36.8
Lowest value	2.9	1.3	21.2	14.4	7.4	10.7	31.2
Difference	0.8	0.5	5.0	1.9	2.7	2.6	5.6

[a]Selected from a study of six varieties of rapeseed grown at 22 stations in 1958 varietal trials (16).

TABLE II

Increases in Erucic Acid Content in First Commercial Production and Processing of Low Erucic Acid Rapeseed in Canada

Product	Fatty acid composition by chain length, % w/w			
	C_{16}	C_{18}	C_{20}	C_{22}[a]
1964 seed plot	4.8	93.7	1.8	0.3
1965 seed increase CDA	4.5	93.7	1.6	0.2
1965 seed increase (comm)	4.8	93.3	1.7	0.2
1966 commercial crop				
Seed, six farms	4.8	92.6	2.0	0.6
Crude oil	4.6	92.2	2.2	1.0
Degummed oil	4.6	92.1	2.2	0.9
Tank cars, three	4.5	91.8	2.2	1.5

[a]Erucic + behenic (behenic ~ 0.2 - 0.5%).

TABLE III

Mean Values for C_{22} Content (% of Total Fatty Acids) of LEAR Rapeseed at Different Stages of Production, Handling and Transportation

Seed source	Variety of rapeseed	
	Span	Oro, Zephyr
Parent seed	2.80	---
Certified seed, 1650[a]	3.22	0.42[b]
Farm sample, 3461[a]	3.36	---
Carlot sample, 1100[a]	3.86	1.10[c]
Cargo sample, 14[a]	4.13[d]	1.51[c]
	4.87[e]	

[a]Numbers of samples analyzed.
[b]Oro and Zephyr.
[c]Mainly Oro.
[d]Thunder Bay.
[e]Vancouver.

minimize cross-pollination, the latter being more extensive with *B. campestris* than with *B. napus*. The harvested rapeseed would be stored in identified storage by the farmer and then handled through the normal grain handling system for final shipment to domestic crushers and to export markets. The elements of the Canadian handling and transportation involve the country elevator, transfer to identified railway boxcars, shipment to terminal elevators, transfer, cleaning to remove weed seeds and other foreign materials, storage and transfer to cargo vessels. This handling and transportation system would be handling the "normal" rapeseed at the same time, and controls were necessary to minimize admixture with the high erucic rapeseed. Since the rapeseed could not be distinguished on the basis of seed characteristics, a monitoring system involving gas liquid chromatographic analysis of the fatty acids in a representative sample of the rapeseed was proposed. This study, involving analysis of the initial seed, farm production, carload and cargo lots, was undertaken to indicate the extent of admixture at the various stages of the production, handling and transportation system and to provide background information for the proposed complete turnover of Canadian rapeseed production to the LEAR varieties.

MATERIALS AND METHODS

B. napus Varieties

The variety Oro was licensed and released for production in 1968, and seed produced by select seed growers was available for commercial production in the 1971 crop year. A limited quantity of seed of a new variety, Zephyr, was produced in 1970, but seed for commercial planting was extremely limited. Thus nearly all of the low erucic acid

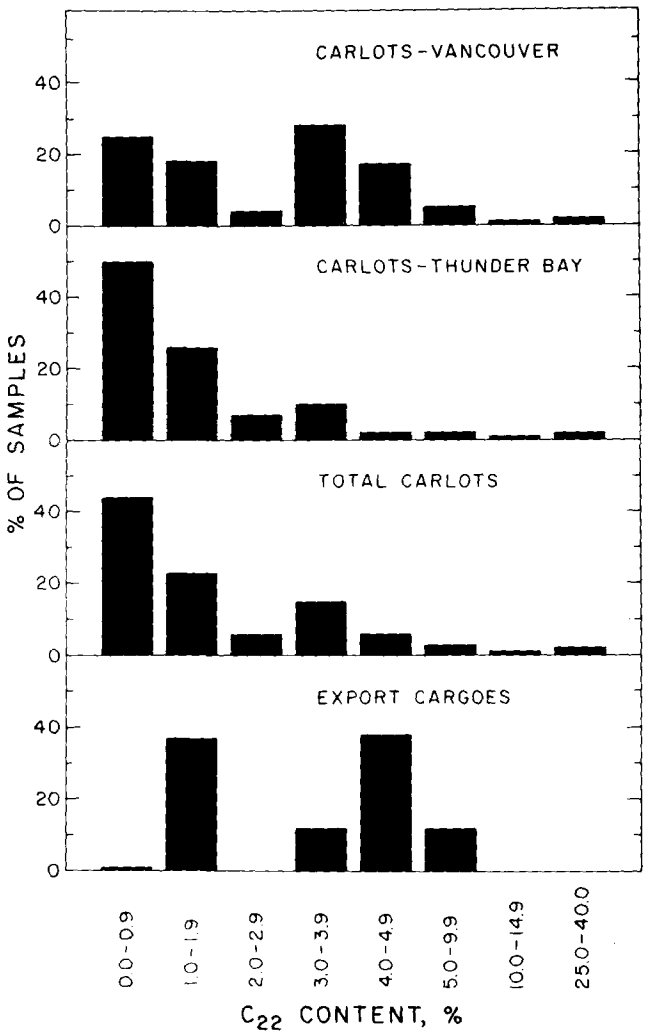

FIG. 2. Comparison of distribution of C_{22} fatty acids in carlot deliveries of LEAR variety to Vancouver and Thunder Bay, total carlots and export cargoes.

seed of this species moving in the commercial channels in 1971-72 was of the Oro variety.

B. campestris Varieties

The plant breeding program to develop a LEAR variety of *B. campestris* was not as far advanced as the *B. napus* program. A limited quantity of a selection given the varietal name Span, having a low erucic acid content, was increased in the Imperial Valley, California, to provide seed supplies for commercial planting in 1971. The variety had not been tested over a range of environmental conditions.

The 1971 seed increase was undertaken by select seed growers in Western Canada. Commercial production was carried out by contracts with farmers. Identified seed was provided and the following stages monitored for the Span and Oro varieties: (a) Span seed grown at one location. GC analyses by Analytical Services Section, Production and Marketing Branch, Canada Dept. of Agriculture, Ottawa, Ont. (b) Seed multiplication in Imperial Valley, California. Seed lots of Span analyzed by Analytical Services Section. (c) Seed production by select seed growers in 1971. Analyses by Analytical Services Section, Saskatchewan Wheat Pool and Prairie Regional Lab. (d) Commercial farm production by contract growers in 1971. Analyses on farm stored lots by Prairie Regional Lab. and Saskatchewan Wheat Pool. (e) Commercial rapeseed. Carlot shipments to terminal elevators at Vancouver, B.C., and Thunder Bay, Ont. Analyses by Grain Research Lab., Canadian Grain Commission, Winnipeg, Man. (f) Commercial rapeseed,

export cargoes. Analyses by Grain Research Lab.

The four laboratories conducted a preliminary study using prepared fatty acid standards and samples of rapeseed to compare inter- and intralaboratory variation. Each laboratory used its own procedures to obtain a representative sample seed, extract the oil, convert glycerides to methyl esters and GC analyses. Satisfactory agreement was achieved, and a large sample of rapeseed from the 1971 crop was used for a daily check on methodology and performance of GC equipment.

RESULTS AND DISCUSSION

The C_{22} content of the Span seed used to plant the winter increase in the Imperial Valley of California was 2.2%. The seed harvested from this planting was returned to Canada in a number of lots, cleaned to seed standards, and the different lots were analyzed for C_{22} content. The C_{22} contents of these lots, shown as a bar graph (Fig. 1), varied from 2.2 to 3.8% with a mean value of 2.83%. Approximately 81% of the samples ranged from 2.7 to 3.2%. Rapeseed had not been grown previously on a commercial scale in the selected areas in California. Thus it is probable that the variation was due to environmental effects, since crop management varied from field to field and contamination from outside sources was nonexistent. The weather during the growing season was not completely favorable and the seed was grown under irrigation, which made comparison to original seed difficult.

The analysis of certified seed produced by Canadian seed growers in 1971 is represented in the middle section of Figure 1. The C_{22} contents varied from 2.1 to 4.5% with a mean of 3.21%. About 5% of the seed samples had a content above 4.0%; 3% had 2.1%; and the remaining 92% ranged from 2.6 to 3.8%. The shift in the mean value from the California seed production could be attributed to environment, admixture from volunteer rapeseed or to cross pollination with volunteer plants in the production field or with plants of other varieties of *Brassica campestris* in commercial production, beyond the mandatory 100 yd of isolation.

The analyses of the commercial rapeseed are shown in the bottom section of Figure 1. The C_{22} contents ranged from 2.1 to 5.1% with a mean value of 3.34%. Approximately 0.9% of total samples submitted for analyses had contents between 5.5 and 10%, and 0.6% were between 10 and 15%. These latter samples were obviously the result of admixture in the production and neither group is shown on the bar graph.

A comparison of the graphical data (Fig. 1) for the seed production in California and in Canada shows a similar range of C_{22} contents, except that ca. 5% of the 1971 seed crop had contents above 4.0%. It is interesting that both productions show some seed with erucic contents around the 2% level. Samples of commercial seed in the 2% range were also found, and the mean values for commercial and seed samples were remarkably similar. Since both commercial and seed production occurred in similar production areas, these could be considered to be produced under similar environmental conditions. The tendency in the commercial samples to erucic contents above 4% when compared to seed or California production would indicate the effects of cross-pollination and admixture from volunteer "normal" rapeseed. It should be noted that isolation techniques and clean field requirements were not as rigid for commercial production as for seed production.

The analyses of the Canadian seed production was carried out by the three laboratories, Plant Products, Saskatchewan Wheat Pool, Prairie Regional Laboratory and the commercial crop production by the latter two laboratories. Different numbers of samples were analyzed by each laboratory and a comparison of the laboratory results is given in Table II. These data show excellent agreement between the means for both seed and commercial production, even though there are differences in the number of samples analyzed and the distribution according to C_{22} content. The results on the commercial crop are of interest, since the samples analyzed by the Prairie Regional Laboratory were drawn from Alberta, Saskatchewan and Manitoba, whereas those analyzed by the Saskatchewan Wheat Pool were taken only from Saskatchewan.

The data on carlot and export cargo shipments are shown in Figure 2. Most of the carlot shipments were identified by variety and include the three varieties, Oro, Zephyr (*B. napus*) and Span (*B. campestris*). A few carlots were not identified by the shipper as to variety. The third section shows the distribution according to the total carlots. Approximately 3% had contents of 5.0-9.9%, 1% with 1.0-14.9% and 2% with 25.0-40.0% of erucic acid. The latter carlots were among those not identified as to variety. The samples with C_{22} contents between 5 and 15% were obviously the result of cross-pollination and admixtures. Over 60% of the carlot shipments were made up of the Oro and Zephyr varieties on the basis of C_{22} contents. The two top sections show the distribution of carlot shipments to the two Canadian terminals. Deliveries to the Vancouver terminals were evenly proportioned between the *B. napus* and *B. campestris* varieties, whereas deliveries to the Lakehead terminals were predominately the *B. napus* varieties. This would be expected since the *B. napus* varieties predominated in Manitoba, whereas Alberta and Saskatchewan production was more evenly divided according to species. The cargo shipments are shown in the last section. The cargoes in the 5.0-9.9% bracket were actually shipments that analyzed for 5.1 and 5.2%, respectively.

The mean values and distributions shown in Figures 1 and 2 provide some initial information on the success of the changeover program in 1971 from the erucic containing rapeseed varieties to the LEAR varieties. The data has been summarized in Table III as mean values for the two species at each monitored stage.

The increase of ca. 0.5% from the parent seed grown in California to the seed and commercial crop grown in Canada in 1971 may have been due to environment, although some increase due to admixture or cross-pollination cannot be ruled out on the basis of the tendency to increase at levels above 4.0% C_{22}. The distributions in Figure 1 indicate that the rigid control in seed production gave less progression to higher erucic content seed, which would support the view that a part of the increase of 0.5% may have been caused by cross-pollination and admixture with volunteer rapeseed. The increase of 0.5% from farm sample to carlot samples would be admixture in the transportation and handling systems carrying two kinds of rapeseed. The increase from carlot to cargo would be ascribed to admixture in the transportation and handling through the terminal elevators with two kinds of rapeseed and was again of the same magnitude.

It would seem reasonable to suggest that completion of the turnover of Canadian rapeseed production to the LEAR types can be accomplished by control over the seed used to produce each new crop. Since the Canadian farmers favor and advocate the use of good seed stock, this solution follows normal agricultural practice. The increases in erucic content due to admixture in the handling and transportation system would be expected to decrease as the relative proportion of LEAR varieties increases in the system.

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