

INFLUENCE OF TEMPERATURE AND DAY-LENGTH ON THE FATTY
ACID COMPOSITION OF SUMMER RAPESEED (BRASSICA NAPUS)

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Introduction

In a paper of APPELQVIST (1971) on the influence of the environment on the fatty acid composition of *Brassica napus* seed oil, a "high linolenic" line is compared with a "low linolenic" line. The lines had been selected under greenhouse conditions and contained 15.6 and 6.1 % linolenic acid. When grown under natural conditions, the difference was reduced to only a few per cent. This example illustrates the difficulties that are being encountered in breeding for changes in fatty acid composition. The breeder is enforced to select under "greenhouse conditions", which are usually variable and, in addition, differ very much from the conditions under which his future varieties will have to perform. Other complicating factors are abnormalities as a consequence of underdeveloped seeds (RÖBBELEN, 1973).

In our laboratory a study into the inheritance of C18 fatty acids in erucic acid free material of summer rapeseed has been started. In the course of the development of the inbred lines to be used in this investigation it soon became clear, that more knowledge on genotype-environment interaction of the fatty acid composition was required. This report describes the results of our first trial on this subject. Earlier work has been published by CANVIN (1965) and APPELQVIST (1971), whereas RÖBBELEN (1973) and RAKOW and MCGREGOR (1973) have published some observations on changes in fatty acid composition in the course of a breeding programme.

Material and methods

Six inbred lines of summer rapeseed (*B. napus* forma annua), which differed in fatty acid composition, were grown in a greenhouse under long day conditions (16 hours) at about 17° C. Just before the start of flowering, the plants were transferred to the phytotron of the Institute of Horticulture at Wageningen. Each inbred line received the following six treatments (4 plants per treatment): three different, constant day/night temperatures (13, 21 and 25° C), whereas each temperature treatment was split into a 8 and a 16 hours daylength treatment. All plants were selfed by hand pollination at different stages of flowering. Ripe seeds were harvested and analysed for total oil content (Fosslet), seed weight, and fatty acid composition of the seed oil. Fatty acids were determined by GLC (column: 9 % BDS on Chrom. W 80/100, 200x0.2 cm). Light intensity was max. 48.000 erg/cm² sec, provided by Philips TL 40 W/57 tubes. The tubes were replaced after 2000 burning hours. Relative humidity was kept at 70 %.

Results

Of the six inbred lines four were zero-erucic and only the results obtained with these four lines will be dealt with. Furthermore, the treatment at 25° C and 8 hours day-length is omitted, as in this treatment too many plants died. In addition, the majority of the surviving plants showed poor male and female fertility.

From table 1 it is clear, that the 1000-seed weight decreases with increasing temperature and also with decreasing day-length. Line P32, however, did not respond to temperature and this indicates a possibility for selection.

Table 1: Influence of temperature and daylength on 1000-seeds weight (g)

Inbred line	Treatment (° C/h daylength)				
	13/8	21/8	13/16	21/16	25/16
ZZ21	4.3	2.6	4.7	3.1	2.8
P29	3.8	2.4	4.4	3.2	2.9
P32	2.4	2.5	3.3	3.1	3.2
P34	3.8	2.6	4.8	3.4	3.7
Average	3.6	2.5	4.3	3.2	3.2

1. s. d. between treatments for a given line: 0.6;

1. s. d. between lines for a given treatment: 0.6.

Table 2 summarizes how total oil content responded to the treatments. The determinations have been carried out with a "Foss-let" apparatus, but unfortunately the samples used were too small. Although this has caused a rather high variation, it is nevertheless clear that the total oil content decreases with increasing temperature and with decreasing day-length.

Table 2: Influence of temperature and daylength on total oil content (%)

Inbred line	Treatment (° C/h daylength)				
	13/8	21/8	13/16	21/16	25/16
ZZ21	32.5	29.1	34.3	31.2	25.9
P29	30.0	22.5	33.1	28.4	26.5
P32	28.0	27.6	37.2	31.9	28.2
P34	38.4	22.0	34.9	28.6	22.3
Average	30.7	25.3	34.9	30.0	25.7

1. s. d. between treatments for a given line: 3.7 (P=0.05);

1. s. d. between lines for a given treatment: 1.5 (P=0.05).

Table 3: Influence of temperature and daylength on the fatty acid composition of Brassica napus seed oil

Inbred line	Day and night temperature (°C)	Daylength (h)	Amount of oil per 100 seeds (mg)	Fatty acid composition				
				16:0	18:1	18:2	18:3	20:1
ZZ21	13	8	140	5.0	66.3	15.2	8.7	3.1
	21	8	76	5.5	70.9	13.8	5.7	2.8
	13	16	161	4.7	67.3	15.4	8.7	2.7
	21	16	97	4.4	75.6	10.4	4.3	2.6
	25	16	73	5.7	72.6	12.3	4.2	3.4
P29	13	8	114	7.0	51.7	26.6	10.9	3.0
	21	8	54	8.4	52.6	25.7	9.0	2.9
	13	16	146	6.4	52.9	25.1	11.7	2.8
	21	16	91	7.7	58.1	22.4	8.4	2.5
	25	16	77	8.2	66.5	16.0	5.0	3.1
P32	13	8	67	5.5	46.2	32.1	12.5	2.8
	21	8	69	6.4	60.9	21.8	7.3	2.6
	13	16	123	3.9	53.9	26.5	11.2	1.6
	21	16	99	5.9	65.7	18.9	6.4	2.3
	25	16	90	5.9	66.7	17.6	5.3	2.2
P34	13	8	108	6.6	46.7	29.0	14.2	2.6
	21	8	57	7.6	51.7	27.2	9.9	2.4
	13	16	168	6.3	50.4	26.7	13.0	2.6
	21	16	97	7.4	57.9	22.2	8.9	2.5
	25	16	83	6.5	66.9	17.4	5.3	2.7

1. s. d. (P=0.05) between lines for a given treatment:
 1. s. d. (P=0.05) between treatments for a given line:

The fatty acid composition of the oil is given in table 3. In agreement with APPELQVIST's (1971) and CANVIN's (1965) results, the palmitic acid content increases somewhat with increasing temperature. This happens both at 8 and at 16 hours day-length. Oleic acid follows the same pattern as palmitic acid, but the effects are more pronounced. As could be expected from the increase in oleic acid content of the oil with increasing temperature, there is a corresponding decrease in the percentage of the more unsaturated fatty acids; both linoleic and linolenic acid content decrease with increasing temperature and also to some extent with increasing day-length.

In this trial no significant differences were found between the fatty acid composition of the samples that originated from different pollination times on the same plant.

Discussion

An extensive discussion on the data obtained seems premature, as many factors may play a role in fat synthesis and only two have been studied incompletely in this trial.

In the foregoing section only fatty acids as percentages of the total amount of fatty acids have been mentioned. However, the total amount of oil may be effected very much (see tables 1 and 2) and, consequently, one should be careful in drawing conclusions as regards "increases" or "decreases" of fatty acids. In table 4 the average values of oleic acid are summarized for different methods of calculation. When expressed as percentage of total fatty acids it increases with increasing temperature. It decreases sharply, however, when expressed in mg per 100 seeds and its decrease is doubtful when expressed as percentage of the whole seed.

Table 4: Amount of oleic acid calculated in different ways

Treatment (°C/h daylength	Method of calculation		
	mg/100 seeds	% on seed	% of the oil
13/8	54.1	15.0	52.7
21/8	36.2	14.5	59.0
13/16	80.8	18.8	56.1
21/16	59.2	18.5	64.3
25/16	53.0	16.6	68.2

Another example is provided by linoleic acid; when calculated as percentage of total fatty acids, the short-day treatment gives higher values than the long-day treatment. The reverse is true when the amount of linoleic acid is calculated as percentage of the seed.

Seed size also seems to influence fatty acid composition. It has already

been mentioned by RÖBBELEN (1973) that oils of underdeveloped seeds tend to possess a high percentage of linoleic acid. This is confirmed by our data from table 5 in which the relationship between seed size and linoleic content of the oil is given for seeds that originated from a field trial. Table 5 also gives the overall relationship between linoleic acid content and seed size for the seeds of the present phytotron trial. Contrary to the field trial the correlation is negative now. Naturally, more variables were included in the phytotron trial, but the two correlations together suggest that without the decrease in seed size in the phytotron, the effect of temperature and/or day-length on the linoleic acid content might have been even more pronounced. Apart from the factors mentioned above it remains to be seen whether seed size, as a genetically controlled factor as such, also influences fatty acid composition. If so, another complication in inheritance studies would arise.

Table 5: Relationship between seed size and linoleic acid content of the oil

Treatment (°C/h daylength)	Phytotron trial (average values of 4 strains)		Field trial (average values of 10 strains)	
	1000-Seed weight (g)	Linoleic acid (%)	1000-Seed weight (g)	Linoleic acid (%)
13/8	3.6	25.7	5.2	21.2
21/8	2.5	22.1	3.5	22.8
13/16	4.3	23.4	2.3	24.5
21/16	3.2	18.4	1.4	26.9
25/16	3.2	15.8		

The response of seed oil content to temperature in CANVIN's (1965) and in our material was stronger than in that of APPELQVIST (1971). At first sight one is inclined to describe this to the low night temperature that, contrary to CANVIN's (1965) and our trial, has been applied by APPELQVIST. On the other hand we cannot exclude that the difference found was due to differences in response between genotypes.

APPELQVIST (1968) has grown *B. campestris* in Sweden and in Turkey. Linoleic acid contents differed hardly in the two climates, but in the phytotron trial (APPELQVIST, 1971) the highest linoleic acid content was clearly obtained at the highest day temperature. The linolenic acid content remained remarkably constant in this trial. The pattern found by us and by CANVIN deviates from these results. As it seems acceptable to assume that cool nights compensate for the linoleic acid decreasing effect of high day temperatures, one is again inclined to describe the difference to the cool nights of the treatments of APPELQVIST. However, it is difficult to accept that cool nights overcompensate, or in other words: if a constant temperature of 25° C gives lower amounts of linoleic acid than a constant temperature of 15° C, why then should the combination 25° C at day/10° C at night give higher amounts of linoleic acid than the combination 15° C at day/10° C at night? In our opinion it seems more likely that the

differences are caused by differences in genotype. Anyhow it seems unlikely that they are caused by differences in humidity as has been suggested by APPELQVIST (1971); in our trial the relative humidity was also kept constant.

It seems clear that the results obtained so far are insufficient to permit the most effective selection of rapeseed for fatty acid composition. The same applies to inheritance studies. In view of the scarcity of phytotrons and the many factors involved, somewhat more cooperation in this field might be worthwhile.

Acknowledgement

The authors wish to thank Prof. J. Doorenbos, Institute of Horticulture, Wageningen, for providing facilities and Mr. K. Steensma of that Institute for his excellent care of the plants. Furthermore, our thanks are due to Mr. A. J. Schakelaar for statistical analysis and to Messrs. G. J. Beutick and H. W. Brinkman for chemical analysis.

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