Breeding for Lipid Composition in Corn

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ABSTRACT

Large genetic diversity is available in corn (*Zea mays* L.) for alteration of its lipid composition. Mass selection has produced strains ranging from 0.4-17% oil with genetic variability still existing in the high oil version. High oil hybrids (7-8% oil) with yields equal to those of commercial hybrids (4% oil) have recently been developed. In feeding trials at the University of Minnesota, pigs made more efficient gains on high oil corn. Oleic and linoleic acids usually make up 80-90% of the fatty acids of corn oil. Screening of corn lines has revealed a range for linoleic acid of 25-71% and for oleic acid of 20-60%. Although the genetics of fatty acid synthesis have not been completely elucidated, breeding of corn with selected unsaturation should be possible. Fatty acid placement within the triglyceride molecule also may be subject to genetic modification in corn.

INTRODUCTION

Corn (*Zea mays* L.) usually is not classified as an oilseed, but with its large acreage in the United States, corn annually produces three-fourths as much oil as soybeans. The corn oil is largely hidden, because only 8.5% of the crop is processed (1). Most of the crop is fed to livestock.

Breeding to improve the chemical composition of corn has been emphasized increasingly in recent years. Several new developments have increased the possibilities for changing the lipid composition of corn.

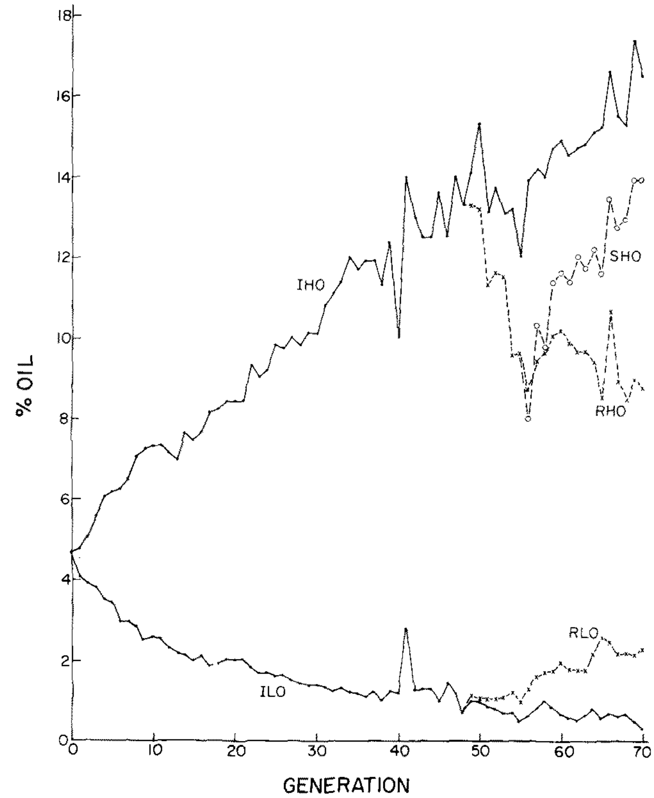

FIG. 1. Mean percentage of oil for Illinois High Oil (IHO), Illinois Low Oil (ILO), Reverse High Oil (RHO), Reverse Low Oil (RLO), and Switchback High Oil (SHO) strains of corn plotted against generations (3).

OIL CONTENT

A classic plant breeding experiment was started at the University of Illinois by C.G. Hopkins in 1896 (2). His objective was to determine whether the percentages of oil and of protein in corn grain could be modified by selection. It is now the longest continuous selection experiment that has been conducted in higher plants (3). The progress attained for high and low oil in 70 generations is shown in Fig. 1. The original Burr's White ears averaged 4.7% oil. After 70 generations of mass selection (selection of ears with lowest and highest oil contents), the Illinois Low Oil strain had 0.4% oil and the Illinois High Oil, 17% (3). The experiment established that oil content is heritable in corn.

The reverse selections, Reverse Low Oil and Reverse High Oil, were initiated after 48 generations, to determine whether genetic variation remained. Switchback High Oil was started from Reverse High Oil after 12 generations of reverse selection. These experiments indicate that the long continued selection has not resulted in fixation of the genes governing high or low oil content. Even in the Illinois High Oil strain, oil should increase further, if selection is continued. At 17% oil, corn approaches the range of oil in oilseeds such as soybean, which has 17-22% oil. The problem with the Illinois High Oil strain is that it is low in yield, perhaps 30% that of good hybrids.

The question of relative yield of standard hybrids and higher oil hybrids has not been fulled resolved, although it is central to wide scale production of higher oil types. Oil gives 9 kcal/g, compared to 4 kcal/g for protein or carbohydrate. If high oil hybrids yielded as much grain per acre as low oil varieties, the high oil types would produce more calories per acre. Production of more calories per acre appears to be an important goal for agriculture in the future. Our experience at the Illinois Experiment Station leads us to believe that higher oil hybrids, perhaps as high as 7-8% oil, are not intrinsically lower yielding than those in the 4-5% range. Table I shows the performance of some newly developed, higher oil hybrids. A standard hybrid, Mo17 x N28, had 3.3% oil and yielded 188 bu/acre. Four commercial hybrids with 4.2% oil averaged 172 bu/acre when grown at Urbana, IL, in 1973. The higher oil hybrids had acceptable yields, 154 to 175 bu/acre, with oil contents from 6.7-8.5%. The higher protein levels of the higher oil varieties may be due to increased germ size in higher oil kernels (3).

Hybrids containing as much as 8% oil are now being commercially produced on a limited basis. Development of

TABLE I

Performance of High Oil and Standard Corn Hybrids, Urbana, Illinois, 1973

Hybrid	Yield[a] bu/A	Water[b] %	Oil[c] %	Protein[c] %
R802A x R806	172	23	8.5	12.7
R802A x R805	154	22	8.3	12.5
$R801^{HT}$ x 10120-8	171	22	7.0	12.8
R806 x N28	175	26	6.7	10.3
Mo17 x N28	188	25	3.3	11.1
Four commercial hybrids[d]	172	24	4.2	10.4

[a]Least significant difference = 18 bu/acre.
[b]Moisture percentage at harvest.
[c]Oil and protein percentages are on a dry-wt basis.
[d]Mean of 4 popular commercial hybrids.

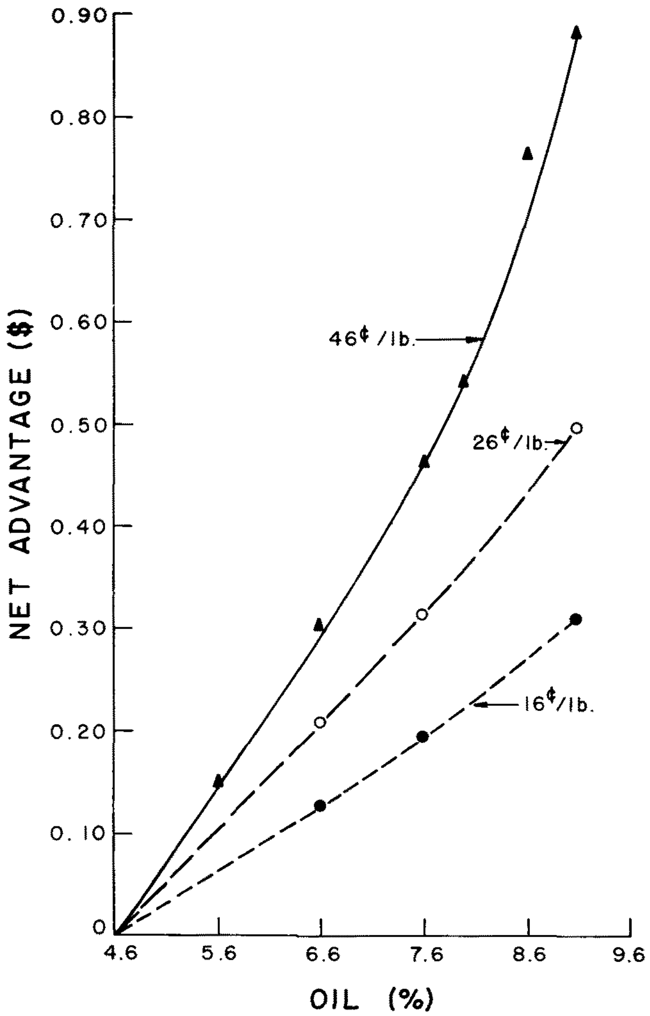

FIG. 2. Increase in value (dollars per bushel) for corn of different oil contents at 3 oil price levels (assuming 85% oil recovery, starch at 7.1¢/lb, 15.5% moisture).

these hybrids was made possible because several unrelated high oil gene pools had been created earlier by selection. Selection in other strains by more efficient schemes than in the Burr's White experiment have produced varieties at the Illinois Station in 10 generations with as much as 12% oil. High oil lines isolated from one gene pool can be tested in combinations with inbreds isolated from an unrelated pool or with high oil backcross recoveries of widely used lines.

Corn oil is the most valuable commercial constituent of the grain. As oil content of corn increases, starch decreases. But because starch is less valuable than oil, high oil corns have a net advantage over normal corn. Figure 2 shows the increase in value of corn of different oil contents at 3 oil price levels. The milling of high oil corn appears to have compelling advantages, particularly to wet millers. Technical details of the milling process may need to be modified for orderly milling of these new hybrids. But if a shortage

TABLE II

Fatty Acid Compositions of Commercial Corn Oil (1,5)

Year	Fatty acid composition (wt-%) 16:0	18:0	18:1	18:2	18:3
1964	11.5	2.2	26.6	58.7	0.8
1968	11.1	2.0	24.1	61.9	0.7

of high quality cooking oil persists, considerable economic advantage appears likely in the milling of higher oil hybrids.

FATTY ACID COMPOSITION

Palmitic, oleic, and linoleic acids make up over 95% of the fatty acids found in corn oil. A low level of the triunsaturated linolenic acid (ca. 1%) makes corn oil less susceptible to oxidation and rancidity than soybean oil, which had ca. 7% linolenic acid (4).

Reiners and coworkers (1,5) determined the fatty acid composition of commercial oil processed from U.S. Midwest corn from December, 1962, to March, 1964, and again 4 years later in 1968 (Table II). For the samples taken before 1964, the average linoleic acid percentage was 58.7. By 1968, the linoleic value had increased over 3%, to 61.9%. Presumably, the introduction of new hybrids brought about this increase. It could easily have been a decrease, because most hybrid seed corn producers do not monitor fatty acid composition. We must be vigilant just to maintain the linoleic or polyunsaturation level in corn oil.

The Illinois High Oil strain has a low level of linoleic acid (48.9%) (Table III). This fact caused some concern that high oil content might be linked with low unsaturation. Recently, hybrids have been developed that have 5.6-7.7% oil that also have higher levels of linoleic acid (52.4-61.9%) (Table III). These data indicate that oil content and fatty acid composition are independent, and that higher oil hybrids can be produced with desired levels of polyunsaturation. Perhaps, however, the higher oil corns, particularly if used for livestock feeding, should be more saturated than normal corns.

In feeding trials at the University of Minnesota with growing-finishing pigs, Nordstrum and coworkers (6) fed high oil corn, which had been grown at Urbana. The high oil corn improved feed conversion efficiency. The amount of high oil corn required per pound of gain was 5-6% less than that of normal corn. The protein supplement needed was also substantially reduced, e.g., a 22% reduction in a 16% protein diet and a 41% reduction in a 13% protein diet.

Table IV shows the fatty acid compositions of the normal corn (59.1%, 18:2), the high oil corn (51.7%, 18:2) and corn oil (60.0%, 18:2) used in the pig diets. The high oil corn was lower in unsaturation than the normal corn or corn oil. The normal corn had 4% oil, and the high oil corn had 7% oil (Table V). Normal corn was supplemented with 3% commercial corn oil to give a comparable total oil content of 7%. A diet with 16% total oil was prepared by addition of 12% corn oil to normal corn. The higher oil

TABLE III

Fatty Acid Compositions of Oil from High Oil Corns

	Oil %	Fatty acid composition (wt-%) 16:0	18:0	18:1	18:2	18:3
Illinois High Oil	16.4	11.7	2.4	36.0	48.9	1.0
R802A x R805	7.7	12.8	1.9	31.4	52.4	1.0
SK85 x N28	5.9	10.3	2.6	34.3	51.5	0.8
SK85 x Mo17	5.6	11.1	2.2	28.8	57.0	0.4
SK85A2 x Oh43HO	5.6	12.2	2.1	22.6	61.9	1.0
Commercial Hybrid 1	3.6	12.7	1.4	26.0	57.9	1.6

TABLE IV

Fatty Acid Compositions of Corn Oil in Pig Diets (6)

	Fatty acid composition (wt-%)				
	16:0	18:0	18:1	18:2	18:3
Normal corn	12.4	2.0	24.1	59.1	2.4
High oil corn	12.4	2.4	31.6	51.7	1.4
Corn oil	12.8	2.0	23.8	60.0	1.4

to saturated fatty acids have been produced when the animals were fed polyunsaturated oils protected against microbial hydrogenation with a layer of formaldehyde treated casein (7,8). However, controversy has arisen over increasing the human consumption of polyunsaturated fats (9,10). Nutritionists have not yet determined the optimum levels of unsaturated and saturated fats for human diets. But whatever the outcome of the debate over unsaturated versus saturated fats, corn has an amazing diversity in fatty acid composition that can be used to meet demands that may arise.

TABLE V

Fatty Acid Composition of Backfat of Pigs Fed High Oil Diets (6)

Oil in diet		Fatty acid composition (wt-%)						
Source	%	14:0	16:0	16:1	18:0	18:1	18:2	18:3
Normal corn	4	1.3	23.5	3.4	11.4	44.1	13.0	1.6
High oil corn	7	1.4	21.5	2.8	10.2	42.4	18.7	1.4
Normal corn + 3% corn oil	7	1.4	21.8	3.0	10.1	37.8	22.6	1.5
Normal corn + 12% corn oil	16	0.8	15.4	1.9	8.1	33.0	37.3	1.8

diets decreased the saturated fatty acids of the pig backfat and increased the deposition of linoleic acid (Table V). The higher level of linoleic acid in the commercial corn oil was reflected as a higher level of linoleic acid in the backfat. With the normal corn diet, the linoleic acid was 13%; with the high oil corn diet, 18.7%; and with the normal corn plus 3% corn oil, 22.6%. Backfat thickness did not increase significantly with the 7% oil diets. The carcasses, although slightly softer, were judged to be acceptable for conventional processing by meat packers. When the normal corn diet was supplemented with another 12% corn oil for a total of 16%, the linoleic acid of the backfat was increased to 37.3%, nearly 3 times the level from a normal corn diet. The carcasses from this diet were soft and oily, and would be unsatisfactory for conventional processing.

The ability to alter the fatty acid composition of animal fats through their diets offers exciting possibilities. In ruminants, meat and milk with higher ratios of polyunsaturated

TABLE VI

Fatty Acid Compositions of Oil from Inbred Lines of Corn

	Fatty acid composition (wt-%)				
Inbred line	16:0	18:0	18:1	18:2	18:3
C105	10.3	3.1	60.5	25.5	0.6
H21	15.1	2.7	37.8	43.4	1.0
M14	12.6	1.4	29.6	54.8	1.6
K6	10.3	1.3	22.8	63.9	1.7
NY16	6.6	1.5	20.3	70.7	0.9

Variations in fatty acid composition of 5 inbred lines are shown in Table VI. Linoleic acid ranges from 25.5%-70.7%. The low (25.5%) linoleic acid line has high oleic acid (60.5%). In most lines, linoleic and oleic acids comprise 80-90% of the total fatty acids and are negatively correlated. Jellum (11) found even greater genetic diversity in fatty acid composition in corn from countries other than the U.S. The ranges were 6-22% for palmitic acid, 14-64% for oleic acid, and 19-71% for linoleic acid. Because fatty acid composition varies greatly, and because composition is heritable, we should be able to breed corn with a selected fatty acid composition.

Only a few studies have been made of the inheritance of fatty acids in corn. Poneleit and Alexander (12) reported a dominant gene for low linoleic acid and high oleic acid in a cross between Illinois High Oil and a related inbred, R84. Evidence for a second gene locus and possible modifier genes was discovered when the genetic study was extended to unrelated strains (13). Poneleit (14) and Jellum (15) concluded that the levels of palmitic, oleic, and linoleic acids are predominantly controlled by loci with additive gene action. Breeding systems, such as mass selection or backcrossing, should be effective to change fatty acid composition. Analysis of the parents should enable satisfactory predictions of the fatty acid compositions of the hybrids.

Genotypic factors have much greater influence on fatty acid composition of corn oil than do environmental factors such as temperature, planting dates, or fertility (16). Inter-

TABLE VII

Stereospecific Analyses of Triglycerides from Three Inbreds of Corn and a Cross of Two of the Inbreds (23,24)

		Fatty acid distribution (mole-%)				
Strain	Position	16:0	18:0	18:1	18:2	18:3
H51	1	26.0	3.4	30.8	38.8	1.0
	2	1.5	0.1	26.8	70.6	1.0
	3	25.4	2.0	36.1	34.9	1.6
C103	1	22.4	4.2	41.2	31.7	0.5
	2	1.0	0.3	40.4	57.5	0.8
	3	14.2	2.1	47.5	35.6	0.6
C103 x NY16	1	21.3	3.2	30.4	44.3	0.8
	2	0.6	0.2	27.6	70.8	0.7
	3	9.0	1.5	38.4	50.1	1.0
NY16	1	15.6	3.9	21.4	57.8	1.3
	2	0.7	0.2	21.6	76.6	0.8
	3	7.0	1.6	19.2	70.6	1.6

actions between genotypes and environmental factors generally are not significant. The opaque-2 locus has little effect on the fatty acid composition of the corn oil (17). Breeding studies are underway to combine improved protein quality, high oil content, and high unsaturation.

FATTY ACID PLACEMENT

Fatty acids do not occur in corn oil as free fatty acids, but are bound in triglycerides. In a triglyceride, 3 fatty acids are esterified to a glycerol molecule. Over the years, there have been many theories as to how the fatty acids were distributed among these 3 positions. The simplest theory was the random distribution theory in which the fatty acid composition at each of the 3 positions would be identical. We had only theories for many years, because there was no way to determine the fatty acid composition at each position individually. In 1966, 2 procedures (18,19) were developed to analyze the fatty acids at each position. When animal triglycerides were analyzed (19,20,21), the fatty acids were found to be distributed completely nonrandomly, i.e., the fatty acid composition at each position was different. Corn oil, soybean oil, and several other vegetable oils were also stereochemically analyzed (22). In the vegetable oils, the fatty acid composition at the middle or 2 position was distinctly different from the fatty acids at the outer (1 and 3) positions, but the fatty acid compositions at 1 and 3 were very similar. There was still some question as to whether the distribution of fatty acids was random between the 1 and 3 positions of triglycerides from plants.

We noticed that all of these analyses had been done on commercial vegetable oils (Mazola for corn, for example). When we analyzed triglycerides from corn inbreds, we found significant differences in fatty acid distribution at all 3 positions (Table VII). The fatty acid distribution was clearly nonrandom at the 1, 2, and 3 positions (23). The saturated fatty acids, palmitic and stearic, were predominately esterified at the 1 position. The 2 position contained the most linoleic and the least saturated acids. The percentage of saturated fatty acids was higher in position 1 than in position 3, and the difference in position 3 was made up by oleic acid or linoleic or both.

The identification of a characteristic fatty acid pattern for each of the inbreds (Table VII) was the most interesting finding of these studies. For example, in H51 and C103, the percentage of oleic acid was higher in 3 than in 1, but in NY16, it was higher in 1. In H51, the level of linoleic acid was higher in 1 than in 3, whereas in the other 2 inbreds, it was higher in 3 than in 1.

Crosses of inbreds have indicated some heritability of fatty acid placement within the triglyceride molecule (24,25). Table VII shows the fatty acid patterns of C103 and NY16 and their cross, C103 x NY16. The major fatty acids at each position of the triglyceride of the cross were intermediate between the parents.

The specific location of fatty acids within the triglyceride molecule of vegetable oils is important for two very practical reasons. First, the placement of the fatty acids in the triglycerides affects their susceptibility to oxidation. Autoxidation, or the development of rancidity, is a major problem in the unsaturated vegetable oils. When Sahasrabudhe and Farn (26) heated corn oil, they found that the fatty acids in the 1 and 3 positions were more susceptible to oxidation than those in the 2 position. Raghuveer and Hammond (27) suggested that concentrating the unsatu rates at the 2 position may orient the triglyceride molecule to make it more stable against autoxidation. If, by breeding, the saturated fatty acids could be concentrated at the outer positions of the triglycerides and the polyunsaturated fatty acids at the middle position, oxidation should be reduced.

Also, high specificity for polyunsaturated acids at the 2 position is desirable nutritionally. When animals digest fats, pancreatic lipase in the intestine cleaves off the fatty acids at the outer, 1 and 3 positions (28). The free fatty acids are absorbed and then metabolized or reesterified. The fatty acid at the middle position remains bound. The resulting monoglyceride passes through the intestinal wall and is resynthesized into triglycerides or other lipids. Efficient use of the essential, polyunsaturated linoleic acid is ensured when it is bound in the middle position.

PROSPECTS

Oil content, fatty acid composition, and fatty acid placement are genetically controlled in corn. Conceivably, the lipid composition of corn can be varied by breeding in 3 ways; 1) quantity of oil, 2) fatty acid composition of the oil; and 3) positional distribution of the fatty acids within the triglyceride. The possibilities for increasing the value of corn for livestock feed and for milling and, perhaps, for development of oils for special purposes are greatly enlarged.

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