Effects of Storage Condition on Oil Analysis of Milled Corn Fractions by Gas-Liquid Chromatography[1]

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ABSTRACT

Storage of various oil-bearing agricultural commodities usually causes a drop in oil content as measured by extraction with petroleum ether. The phenomenon involved in this change, and conditions which affect it, have been explored to a limited extent. A gas-chromatographic method, developed at this Laboratory for determining the oil content of dry-milled corn fractions, was used to examine the effects of heat, particle size, and type of atmosphere on changes in extractable oil content. Ground and unground corn grits were held for various periods under atmospheres of air, oxygen, nitrogen, carbon dioxide, helium, and vacuum and at temperatures ranging from -25 to 160 C. These experiments demonstrated that loss of soluble oil was directly related to storage time, temperature, particle size, and oxygen content of the atmosphere.

Numerous investigators have observed a decrease in petroleum ether-extractable oil during storage of various oil-bearing agricultural materials (1-6). These reports indicate that the phenomenon occurs in most ground foods or feedstuffs, including cottonseed, rice, mustard seed, wheat, and corn. We observed that when finely ground corn fractions were stored at room temperature, their soluble oil content was significantly lower after 6 months.

Several explanations are possible for the decrease in soluble oil content during storage of milled corn fractions. The predominant one is exposure to oxygen, which may cause autoxidation of unsaturated fatty acids or other reactions, or both (7,8). Formation of polar, oxygen-containing compounds and polymers decreases the solubility of triglycerides in the nonpolar solvent normally used in the oil determination procedure (2). Another possible explanation for loss of soluble oil during storage concerns associative forces (oil fixation) between the glyceride and the meal or flour portion of the corn fraction (5,9). This oil fixation also reduces the amount of solvent-extractable oil.

Until recently the petroleum ether extraction method (10) has been the primary means for determining the oil content of milled corn fractions. The adverse effects of storage were difficult to determine accurately by this method. With the advent of gas-liquid chromatography (GLC), we developed a more precise method for analysis of oil in corn fractions (11). The GLC method for determining oil content does not detect unsaturated oil that has possibly become oxidized or bound in the meal. After examining a number of U.S.- produced corn oils, Beadle and co-workers (12) reported that the content of the saturated C_{16} methyl ester is generally between 11 and 12%. Therefore, when the content of the C_{16} climbs above the 12% level as indicated by GLC, it can be assumed that some loss of the unsaturated portion of the oil has occurred.

A series of experiments was made to determine the factors that cause a decrease of soluble oil in milled corn fractions and the degree that each factor contributes to the decrease. Samples were stored at various temperatures for periods up to 12 months. Influence of atmospheric oxygen was also determined.

MATERIALS AND METHODS

The corn fractions used in these experiments were produced during experimental dry-milling of yellow dent hybrid corn. These samples included large grits fractions and -25-mesh degerminator fines. For storage tests where ground material was used, the samples were passed through a Mikrosampl mill fitted with a 60-mesh screen. Oil content and the relative proportion of saturated C_{16} and unsaturated components in the oil for all samples were determined by our newly developed gas-chromatographic method (11). The effects of time, temperature, particle size, and atmosphere during storage of the fractions were also examined.

GLC Analysis of Oil

The GLC method is more reliable than the solvent extraction method for determining oil content at low levels, e.g. 1%. In the GLC method, the ground sample is extracted for 16 hr. at room temperature with a mixture of benzene, methanol (containing anhydrous hydrogen chloride gas), and dimethoxypropane. This solvent mixture does not extract the oil but rather transesterifies it into methyl esters. The transesterified mixture is injected directly into a chromatograph equipped with an SE-30 column. The methyl esters are separated by chain length into C_{16} (methyl palmitate) and the saturated and unsaturated C_{18} group. (An actual chromatogram of the eluted esters is shown in Fig. 7.) After a base line is ruled under each peak in the chromatogram, the peaks are cut out and weighed to $\pm$ 0.05 mg. Samples of refined corn oil treated identically were used to standardize the method. The percentage of oil in a sample is calculated by comparison of the total peak weight of the peaks from the sample with those from the standard.

A comparison of oil analysis is made in Table I between the standard petroleum ether extraction and the GLC method for a series of degerminator fractions.

The loss of soluble oil as determined by GLC was always accompanied by a corresponding loss in petroleum ether extractables.

Effect of Time and Temperature on Storage

To study the effects of both time and temperature, several samples of ground grits were stored in sealed bottles under air at various temperatures. Ground samples stored at $100°C$. were analyzed for their oil content at intervals during a 16-hr. period. Samples stored at $80°C$. were analyzed over a period of 8 days at 24-hr. intervals. Samples held at $-25°$, $25°$, and $45°C$. were analyzed at various intervals up to 12 months. To determine the maximum drying temperature that a sample could withstand without loss of soluble oil, equivalent samples of whole grits were held at $20°$ to $160°C$. in air for 16 hr. and analyzed.

TABLE I.   COMPARISON OF GLC METHOD WITH PETROLEUM ETHER EXTRACTION
ON FRACTIONS FROM CORN DEGERMINATOR STOCK

Fraction	Petroleum Ether Extraction %	GLC Method %	Increased Oil Found by GLC %
$-3\frac{1}{2}+4$ Grits	0.70	0.82	17.1
$-4+6$ Grits	0.56	0.68	21.4
$-6+8$ Grits	0.58	0.71	22.4
$-16+25$ Fines	2.14	2.51	17.3
$-25+Pan$ fines	5.32	6.06	13.9
Hull	2.99	3.27	9.4

Effect of Grinding on Storage

Samples of grits, both "as-is" and ground through 60-mesh, were stored in air at room temperature and analyzed periodically during a 10-month interval. To study the effect of storage under accelerated conditions, samples of grits, both as-is and ground, were stored at 160°C. and analyzed for their oil content after 1, 2, 4, 8, and 16 hr.

Effect of Headspace Gas on Storage

Ground samples were stored at room temperature under atmospheres of air, oxygen, nitrogen, or under vacuum (less than 1.0 mm. Hg) and were analyzed for oil up to 7 months' storage. Samples under a headspace of air were subjected to temperatures at 20°C. intervals from 60° to 160°C. for 16 hr. and analyzed for oil. For other tests with oxygen-free atmospheres, samples of grits, ground to −60-mesh, were alternately evacuated and purged three times with atmospheres of carbon dioxide, nitrogen, or helium.

RESULTS AND DISCUSSION

Effect of Storage Temperatures

Periodic analysis of −25+pan degerminator fines (ground to −60-mesh) held at −25°C. showed no loss in soluble oil (reported as weight percent of sample, wet basis) after 12-month storage (Fig. 1). However, equivalent material stored at room temperature (25°C.) for the same period of time decreased approximately 25% in oil content. When the storage temperature was increased to 45°C., the loss of oil exceeded 50% at the end of 12 months. At 80°C. approximately 60% of the oil was lost in 3 days, but at 160°C. more than 60% of the oil content was lost in only 2 hr.

Figure 1 also gives the proportion of C_{16} in the oil. As the total oil decreases, the relative amount of C_{16} increases. For the −25° and 25°C. storage tests, the C_{16} value remained at about 12%. At the higher temperatures, however, the proportion of C_{16} began to rise at approximately the same time the total oil decreased.
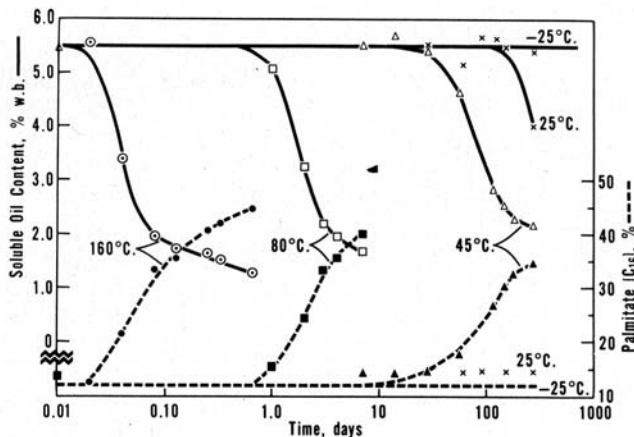

Fig. 1. Effect of storage time and temperature on soluble oil content (open symbols) of a high-oil corn fraction (−25+pan fines ground to −60-mesh). Effect on proportion of C_{16} in the oil is also shown.

This same series of storage experiments, when repeated on a low-oil fraction (−6+8 grits, ground to 60-mesh), gave quite similar results (Fig. 2).

The effect of storage temperature on soluble oil content of −25+pan fines (ground to −60-mesh) was shown in a different manner by holding comparable samples at 20° to 160°C. for 16 hr. (Fig. 3). Since water loss normally becomes rapid between 90° and 100°C. and the greatest decrease in oil also occurred within this temperature range, consideration was given to the possibility that perhaps water was shielding the loss of oil at lower temperatures. To determine whether moisture might inhibit loss of oil, grits (ground to −60-mesh) were vacuum-dried at 60°C. to less than 1% moisture and stored at temperatures between 20° and 160°C. Even after removal of most of the moisture, the oil content again fell off drastically at temperatures just below 100°C. Apparently between 0 and 15% levels, moisture content had little effect on storage stability of this grits fraction.
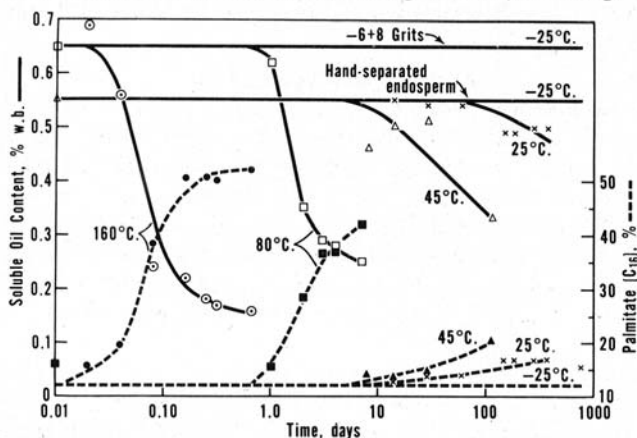

Fig. 2. Effect of storage time and temperature on soluble oil content of a low-oil corn fraction (−6+8 grits and hand-separated endosperm—each ground to −60-mesh).
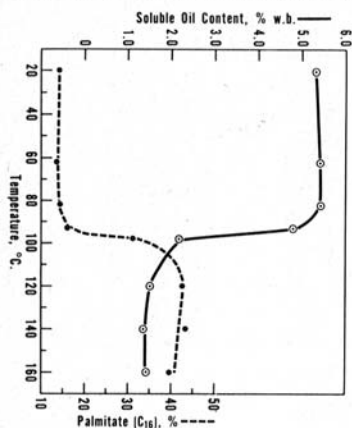

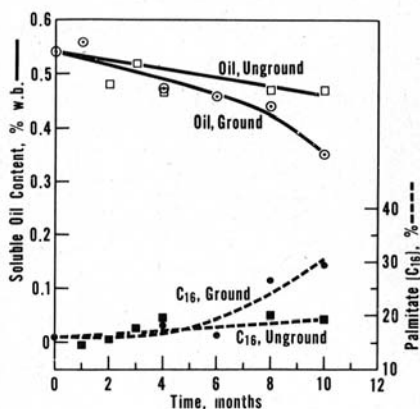Fig. 3. (left). Soluble oil and palmitate contents of −25+pan fines (ground to −60-mesh) stored in air 16 hr.

Fig. 4.(right). Influence of storage time and particle size on the soluble oil content of −4+6 grits stored at 25°C.

Storage of Ground and Unground Corn Fractions

Data in Fig. 4 illustrate that whole grits withstood 10-month storage at 25 °C. with less loss of oil than did the sample ground to −60-mesh. Particle size showed a greater effect when ground and unground samples of these grits were stored at 160°C. for 16 hr. (Fig. 5). At this higher temperature, oil content of the unground grits decreased by only 20%, whereas 75% of the oil was lost in the ground fraction.

Storage of Finely Ground Fractions in Different Atmospheres

Figure 1 depicts the effect of storage time and temperature on oil content of degerminator fines ground to −60-mesh and stored in an air atmosphere. When samples of −4+6 grits ground to −60-mesh were stored at 25° C. for a 7-month period under atmospheres of nitrogen, air, or oxygen, the loss in oil varied with the amount of oxygen present (Fig. 6). Best protection was provided by nitrogen; with it the oil value decreased by a comparatively small 16% during the 7-month interval. With either an air or oxygen atmosphere, the decrease was considerably more—45 to 50%. The relatively small difference between the air and oxygen values indicated that sufficient oxygen was present in the air to promote large losses of oil, and that the higher oxygen concentration in the pure oxygen atmosphere had only a minor effect. Storage under any inert atmosphere that excluded oxygen greatly diminished oil loss.

The effect of a protective atmosphere on oil content of ground grits is vividly illustrated by the actual methyl ester peaks produced in a gas-chromatographic oil analysis. Curves in Fig. 7 show that areas of the C_{16} peaks remained essentially constant, whereas areas of the unsaturated C_{18} peaks diminished. The sample stored under nitrogen was heated at 140°C. for 16 hr. with only a 7% loss of oil, whereas the sample stored in the air atmosphere lost 54% oil after being heated for 16 hr. at only 100°C.

For samples in which a loss of soluble oil has occurred, the original oil content can be calculated from the C_{16} value, as demonstrated by use of the data from Fig. 7. For samples b and c the original oil content can be calculated as follows: b(16.2/15.2) (0.63) = 0.67%, c (33.1/15.2)(0.31) = 0.67%. Both of these values are in good agreement with sample a. These calculated estimates of oil content are
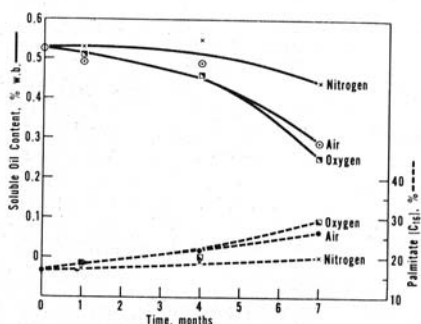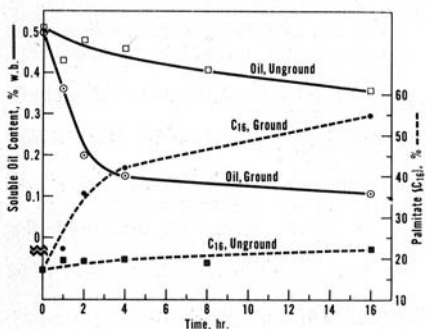

Fig. 5.(left). Influence of storage time and particle size on soluble oil content of −4+6 grits stored at 160 C.

Fig. 6. (right). Relationship between storage time, atmosphere, and soluble oil content of −4+6 grits (ground to −60-mesh) stored at 25 C.
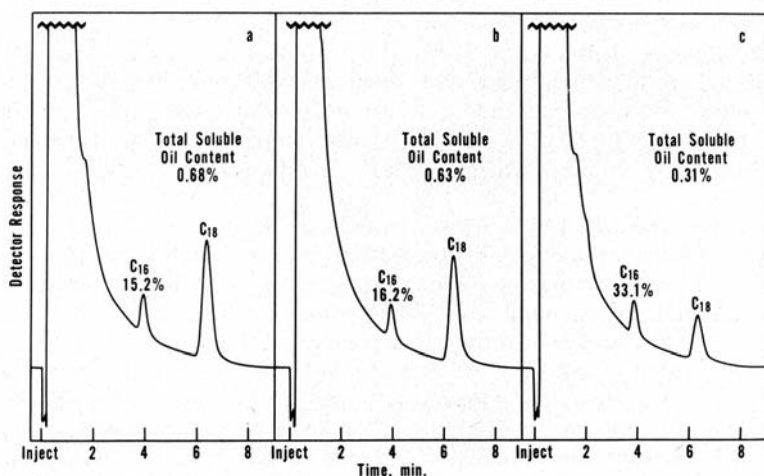
Fig. 7. Gas-chromatographic oil analysis of $-3\frac{1}{2}+4$ grits (ground to -60-mesh), illustrating the relation between atmosphere and peak area of the methyl ester: a) air atmosphere (control); b) nitrogen atmosphere (16 hr. at $140°C.$); c) air atmosphere (16 hr. at $100°C.$).

based on the principle that the same amount of palmitate (C_{16}) is recovered from samples a, b, and c in spite of the variation in total amount of soluble oil.

A change in the physical nature of the samples was also observed. The sample stored under nitrogen at $140°C.$ darkened considerably and was somewhat gummy and partially charred. The one held in the air atmosphere at $100°C.$ showed only slight darkening and remained a free-flowing powder. Apparently browning of the sample due to roasting had no correlation with the oil loss.

These findings indicate that the observed losses of soluble oil during storage of milled corn fractions are due to 1) loss of the unsaturated fatty acid components of the glyceride molecule, undoubtedly through oxidative change, and 2) some degree of polar association between the oil and the meal.

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