acetates. A slight change in the composition will be effected by converting these values to the original acids and alcohols.

Conclusions

Gas-liquid chromatography has been shown to be applicable to the analysis of fatty alcohols. Through the use of polyester columns these alcohols have been separated according to chain length and degree of unsaturation. A study has been made of the relationship between peak areas of the chromatograms and the actual weight percentages of the four C_{18} alcohols found in the fatty alcohols derived from linseed oil. Fatty alcohols, prepared from soybean, linseed, and sperm oil have been prepared and analyzed by the proposed procedure.

Craig and Murty (1) have recently reported that polyesters based on succinic acid are preferable for the liquid phase of the chromatographic column to those made from adipic in that they afford a better separation of methyl stearate from methyl oleate. Conversely adipic columns gave a more effective separation of the esters of linolenic and arachidic acids.

The application of these polyesters to the analysis of fatty alcohol acetates is expected to improve their separation in a similar fashion, but further work is indicated in the search for a liquid phase that will permit both separations in the minimum time.

REFERENCES

1. Craig, B. M., and Murty, N. L., Can. J. Chem., *36*, 1297 (1958).
2. Keulemans, A. I. M., Gas Chromatography, Reinhold Publishing Corporation, New York, 1957.
3. Link, W. E., Hickman, H. M., and Morrissette, R. A., J. Am. Oil Chemists' Soc., *36*, 20 (1959).
4. Lipsky, S. R., Chem. Eng. News, *36*, (No. 5), 48 (1958).
5. Nystrom, R. F., J. Am. Chem. Soc., *69*, 1197, 2548 (1947).
6. Orr, C. H., and Callen, J. E., J. Am. Chem. Soc., *80*, 249 (1958).

[Received November 12, 1958]

The Occurrence of Higher Fatty Acids in Corn Pollen[1, 2]

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IN RECENT YEARS the lipide constituents of pollen have been the subject of several investigations. From the ether extracts of corn pollen Anderson (1) isolated a mixture of two phytosterol palmitates, a saturated hydrocarbon, a C_{30}-saturated alcohol, and a phosphatide. Previously Miyake (2) reported the presence of phytosterol. A summary by Lunden (3) reports the occurrence of other acids, hydrocarbons, and sterols in the pollen of other plants. Since little information is available on the fatty acid content of corn pollen, a study was made to identify these lipides in the saponifiable fraction of corn pollen.

Two kg. of freshly collected corn pollen, *Zea mays* (variety Ohio M-15 hybrid field corn), were placed in a 5-liter flask. The pollen was covered with ethyl ether and permitted to stand for a day at room temperature. Periodically during extraction the ether was kneaded into the dough-like mass of pollen. The extract was then decanted and replaced with fresh ether, and this procedure of extraction and decantation was subsequently repeated at daily intervals until 10 fresh portions of ether had been used for each flask of pollen. The combined ether extracts were concentrated by distilling off the ether. The last traces of solvent were removed by evacuating the flask containing the extract. From 7.89 kg. of pollen were obtained 304 g. of ether-soluble oil.

The saponifiable fraction was obtained by refluxing a solution of the lipides (214.6 g.) in 400 ml. of 1.5 *N* ethanolic potassium hydroxide for 8 hrs. The hydrolyzate was diluted with 1,600 ml. of water, and the unsaponifiable fraction was removed by extraction with Skellysolve "B." Acidification of the aqueous-

[1] This investigation was supported by a grant from the National Science Foundation.
[2] Journal Article No. 2139 of the Michigan Agricultural Experiment Station.

TABLE I
Fractionation of the Methyl Esters of the Fatty Acids in Corn Pollen

Fraction number	Boiling point in degrees C. at 2–3 mm. of pressure[a, b]	Weight of fraction in grams	Iodine number	Refractive index at 25°C.	Saponification equivalents of methyl esters		Melting point of derivative, degrees C.		Acid identified
					Found	Calcd.	Observed[b]	Reported	
Original methyl ester	.....	100.2	142	1.468	293	.....	.....	.....	.....
1	155–162 (155)	10.1	20	1.444	262	270.5	86[c]	86	Palmitic
2	162–168 (165)	17.1	80	1.450	280	270.5 294.5	86[c] 114[e]	86 114.7–115.2	Palmitic Linoleic
3	168–176 (174)	16.9	128	1.455	281	270.5 296.5 294.5	86[c] 132[d] 114[e]	86 132 114.7–115.2	Palmitic Oleic Linoleic
4	176–180 (177)	23.5	217	1.468	291	298.5 296.5 292.5	90[c] 132[d] 181[f]	90 132 181.5–181.6	Stearic Oleic Linolenic
5	180–184 (184)	9.1	210	1.469	293	298.5 294.5 292.5	90[c] 114[e] 181[f]	90 114.7–115.2 181.5–181.6	Stearic Linoleic Linolenic
Residue	.....	23.6	.....	.....	.....	.....	.....	.....	.....

[a] Figures in parentheses show temperature in which major portion of the fraction was distilled.
[b] Uncorrected.
[c] *p*-Bromophenacyl ester (5)
[d] Hydroxy acid (6).
[e] Tetrabromo-addition compound (7).
[f] Hexabromo-addition compound (7).

ethanolic phase with concentrated hydrochloric acid to pH 3, subsequent extraction with ethyl ether, and removal of the ether by distillation under reduced pressure in an atmosphere of carbon dioxide yielded 112.4 g. (52%) of crude fatty acids.

The methyl esters were prepared by refluxing a solution of 112.4 g. of the fatty acids in 250 ml. of absolute methanol, containing 5% sulfuric acid (by weight) for 6 hrs. Removal of the methanol by distillation *in vacuo* gave a residual oil, which was diluted with 300 ml. of water and neutralized with a 10% sodium bicarbonate solution. The neutral solution was extracted with ethyl ether, the ether extracts were washed with water and dried over anhydrous sodium sulfate, and the solvent was removed under reduced pressure, giving 105 g. of crude methylated fatty acids.

The crude methyl esters were fractionated in an electrically heated, 24-in. Stedman column equipped with a D. M. Smith still head. Five fractions were collected at temperatures between 155–184°C. at 2–3 mm. of pressure with a reflux ratio of 10:1. The iodine absorption number (Wijs method) (4), saponification equivalent (5), and refractive index ($n^{25/D}$) of each fraction were determined.

Each fraction of the distilled esters was saponified, and the fatty acids were separated by the lead salt-ether procedure (4). The saturated fatty acids were identified by their *p*-bromophenacyl-derivatives (5) and the unsaturated fatty acids by their hydroxy- (6) or bromo- (7) derivatives. The *p*-bromophenacyl esters were recrystallized from ethanol and the dihydroxy acids from ethyl acetate. Fractionation of the bromo-derivatives was accomplished from their different solubilities in petroleum ether, ethyl ether, and benzene. The tetrabromo acids were recrystallized from ethylene dichloride and the hexabromo acids from dioxane.

In corn pollen the saponifiable material was found to represent 52% of the ether extracts. The lipides of corn pollen thus contain large amounts (48%) of unsaponifiable materials; this value is much greater than has been reported for corn oil (8). Results of the fractionation of the methyl esters from the saponifiable material in corn pollen and the physical and chemical characteristics used in identifying the fatty acids are shown in Table I. Palmitic acid was identified in fractions 1, 2, and 3; linoleic (9:10-, 12:13-octadecadienoic) acid in fractions 2, 3, and 5; oleic (9:10-octadecenoic) acid in fractions 3 and 4; stearic acid in fractions 4 and 5; and linolenic (9:10-, 12:13-, and 15:16-octadecatrienoic) acid in fractions 4 and 5.

Summary

The methyl esters of the fatty acids of corn pollen were prepared and fractionated through a Stedman column. Palmitic, stearic, oleic, linoleic, and linolenic acids were identified by the melting points of the *p*-bromophenacyl esters of the saturated acids and the hydroxy and bromine addition compounds of the unsaturated acids.

REFERENCES

1. Anderson, R. J., J. Biol. Chem., *55*, 611 (1923).
2. Miyake, S., J. Biochem. (Japan), *2*, 27 (1922).
3. Lunden, R., Swensk Ken. Tidskr., *66*, 201 (1954).
4. "Official Methods of Analysis," 8th Ed., Association of Official Agricultural Chemists, Washington, D. C., 1955, pp. 460, 465, 469.
5. Shriner, R. L., and Fuson, R. C., "The Systematic Identification of Organic Compounds," New York, John Wiley and Sons Inc., 1948, pp. 134, 157, 223.
6. Lapworth, A., and Mottram, E. N., J. Chem. Soc., *127*, 1628 (1925).
7. McCutcheon, J. W., "Organic Syntheses," Coll. Vol. 3, New York, John Wiley and Sons Inc., 1955, pp. 527, 532.
8. Jamieson, G., "Vegetable Fats and Oils," New York, Reinhold Publishing Corporation, 1943, pp. 178, 345.

[Received October 22, 1957]

Search for New Industrial Oils. I. Selected Oils from 24 Plant Families

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AN EXTENSIVE PROGRAM has been initiated in the U. S. Department of Agriculture (12) to search for new industrial raw materials among the many plants that have had little or no study of their chemical composition. Ideally such raw materials would fill a present or anticipated need and would not be in competition with presently grown crops, especially those now in surplus supply. Examples of preferred products from major plant constituents are cellulosic fibers for the paper industry, proteins for feed and industrial use, vegetable oils of special composition, and useful polysaccharides other than starch. Insecticides, alkaloids, waxes, essential oils, and many other constituents of potential value may also be found as the program develops.

In one phase of the screening research, seeds from many species have been analyzed for moisture, ash, protein, and oil and have been tested qualitatively for starch, tannin, and alkaloids. The protein, oil and starch analyses indicate major components; the remaining tests give supplementary information without any great increase in the time required for analysis. Limiting the variety of analyses performed initially permitted examination of an increased number of samples even though such limitation might result in incomplete identification of, or sometimes failure to find, components of special interest.

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