• *Letter to the Editor*

# The Gossypol Content and Oil Composition of "Gossypol-Free" Cottonseeds

As a result of selective breeding, a strain of cottonseed has been developed which is said to be free of gossypol. V. L. Frampton of the Southern Utilization Research and Development Division in New Orleans supplied us with a sample of such seeds, grown in Shafter, Calif., during 1958 by Scott McMichal of the United States Department of Agriculture, Crops Research Division. The widespread interest in this program leads us to report briefly the results of our analyses for the presence of gossypol in a sample of this seed. In addition, the oil from this seed has been examined to determine whether any changes in this component occurred.

The seeds were rolled, flaked, extracted with hexane, and air-dried. The desolventized flakes and a corresponding sample of flakes from a sample of cottonseed that contained pigment glands were analyzed for gossypol content. "Free" gossypol was determined by the chromatographic method of Schramm and Benedict (1). We have recently found that some of the gossypol in cottonseed flakes is associated with other components, which give it physical properties that have led us to refer to it as "soluble-bound" gossypol. This "soluble-bound" gossypol was fractionated into that associated with phospholipid and a second fraction of a more hydrophilic nature. The basis for this separation has been reported (2) and will be described in detail in a subsequent publication.

The values obtained for these various gossypol components are given in Table I. Only trace amounts of any of the forms of gossypol were found in the gland-free cottonseeds.

TABLE I

Gossypol Components in Hexane-Extracted Flakes from Gland-Free and Gland-Containing Cottonseeds

| | Gland-free cottonseed | Gland-containing cottonseed |
|---|---|---|
| | %[a] | %[a] |
| "Free" gossypol | 0.002 | 0.40 |
| Phospholipid bound gossypol | 0.002 | 0.16 |
| Hydrophilic soluble-bound gossypol | 0.007 | 0.07 |

[a] The values are reported as percentage of gossypol equivalent.

The crude oil obtained by hexane extraction of the gland-free cottonseeds was analyzed by the standard procedures of the American Oil Chemists' Society (3). The values obtained are given in Table II and are similar to those for oil obtained from cottonseeds that contain pigment glands.

The triglycerides were isolated from the oil by the chromatographic procedure of Quinlin and Weiser (4). The location of the fatty acids in the triglyceride molecules was determined by hydrolysis with pancreatic lipase (5). The fatty acid composition of the original triglycerides and the monoglycerides isolated at the end of the enzymatic splitting was determined by vapor-phase chromatography. The results of these analyses are given in Table III. For purposes of comparison the values obtained on oil from cottonseeds which contained glands are shown also. Trace amounts of myristic, palmitoleic, and linolenic acid were detected in both samples but are not reported because of the large relative error at these low concentrations.

TABLE II

Analytical Values Obtained on Crude Oil Extracted from Gland-Free Cottonseeds

| | |
|---|---|
| Acid value | 2.5 |
| Hydroxyl value | 4.0 |
| Unsaponifiable, % | 1.2 |
| Total fatty acids, % | 95.0 |
| Iodine value | 108.1 |
| Fatty acid composition:[a] | |
| Saturated, % | 27.6 |
| Oleic, % | 19.3 |
| Linoleic, % | 53.1 |

[a] Alkali isomerization-spectrophotometric method.

TABLE III

The Percentage Distribution of Fatty Acids in the Triglycerides of Gland-Free and Gland-Containing Cottonseeds[a]

| | Palmitic | Stearic | Oleic | Linoleic |
|---|---|---|---|---|
| Fatty acid composition of whole triglycerides | | | | |
| Gland-free | 25 | 3 | 18 | 53 |
| Gland-containing | 22 | 3 | 17 | 56 |
| Composition of fatty acids esterified with the 2-position | | | | |
| Gland-free | 2 | 0 | 22 | 75 |
| Gland-containing | 3 | 1 | 20 | 76 |
| Composition of fatty acids esterified with the 1- and 3-positions | | | | |
| Gland-free | 36 | 4 | 16 | 42 |
| Gland-containing | 32 | 4 | 16 | 46 |

[a] Gas chromatographic separation by the method of Orr and Callen (6).

The values obtained for the two fats are quite similar. The almost complete absence of saturated fatty acids from the 2-position of the triglyceride molecule is apparent and is in agreement with previous observations from this laboratory.

These results demonstrate that this selectively bred cottonseed is essentially free of gossypol. Moreover the oil from this seed is similar to that in seeds containing gossypol.

F. H. Mattson, J. B. Martin, R. A. Volpenhein,
Procter and Gamble Company, Miami Valley Laboratories, Cincinnati, Ohio

REFERENCES

1. Schramm, Guenter, Benedict, J. H., J. Am. Oil Chemists' Soc., *35*, 371–373 (1958).
2. Martin, J. B., "Gossypol-Chemistry, Cottonseed Meal Components and Physiological Activity," presented at the Conference on Chemical Structure and Reactions of Gossypol and Nongossypol Pigments of Cottonseed, March 19–20, 1959, New Orleans, La.
3. American Oil Chemists' Society Official and Tentative Methods, V. C. Mehlenbacher and T. H. Hopper, editors, 2nd ed., rev. 1958, Chicago, Ill.
4. Quinlin, P., Weiser, H. J., J. Am. Oil Chemists' Soc., *35*, 325–327 (1958).
5. Mattson, F. H., Lutton, E. S., J. Biol. Chem., *233*, 868–871 (1958).
6. Orr, C. H., Callen, J. E., Ann. N.Y. Acad. Sci., *72*, 649–665 (1959).

[Received September 16, 1959]