Sesamin, Sesamolin, and Sesamol Content of the Oil of Sesame Seed as Affected by Strain, Location Grown, Ageing, and Frost Damage

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THE DISCOVERY that the insecticidal potency of pyrethrins may be markedly synergized by sesame oil (6) led Haller and coworkers (8, 9) to investigate the principles responsible for this activity. One of these principles was found to be sesamin. By investigating a series of compounds similar to sesamin, these workers established the importance of the methylenedioxyphenyl group in this insecticidal activity. This finding has made possible the commercial development of such excellent synthetic synergists as sulfoxide (15), piperonyl butoxide (16), piperonyl cyclonene (16), and *n*-propyl isome (14).

Recently sesamolin, another constituent of sesame oil, was reported to be a potent synergist, being about five times as active as sesamin (2). Systematic entomological examination of the chromatographic fractions of sesame oil indicated that sesamin and sesamolin were the only two synergists present in appreciable quantity. Studies on the chemistry of sesamolin indicated a close similarity between it and sesamin, and a tentative formula for its chemical structure was advanced. A more recent investigation has given further confirmation of this structure (4).

Sesamol has been found to be an antioxidant that gives excellent protection to vegetable oils against rancidity (5). This compound occurs in sesame oil in free form and is also liberated from sesamolin by dilute mineral acids or by hydrogenation (2). The stabilization of sesame oil by hydrogenation is undoubtedly due to the liberation of sesamol from sesamolin (5). Sesamol is not active as a pyrethrum synergist (7).

The investigation described herein was undertaken to obtain information on the contents of sesamin, sesamolin, and sesamol in the oil expressed from different strains of sesame seed and to determine whether they are affected by the location grown. The amounts of the three constituents in the oil may contribute toward the value of sesame as an oil-seed crop. Kinman and Stark (12) have recently reported the yield and chemical composition of sesame as affected by variety and location grown. They also make reference to the history of sesame cultivation and point out that this crop may have commercial possibilities in this country owing to the breeding of indehiscent (non-shattering seed capsules) strains which make possible machine instead of hand-harvesting. The breeding program being carried out by Kinman and Martin (11) has made excellent progress.

The effect of accelerated ageing and frost damage on the content of sesamin, sesamolin, and sesamol was also investigated.

Materials and Methods

Thirty-three dehiscent strains of sesame seed, all grown at College Station, Tex., in 1952, were used to study the influence of genotype. The oil from these strains, which, incidentally, are not commercial varieties but sources of germ plasm in the breeding program, should yield representative data on the range of the three constituents normally present in sesame oils since they were chosen to represent a wide variety of genetic material. The study on the effect of location grown was made on four strains grown at six locations in 1953. Two of the strains, K-10 and Venezuela 51, are dehiscent, and the others were experimental indehiscent strains.

TABLE 1
Percentage of Sesamin, Sesamolin, and Sesamol in the Oil of 33 Strains of Sesame Seed

Strain	Sesamin	Sesamolin	Sesamol
K 6-3-1-B	0.492	0.421	0
K 8-1-1-B	0.462	0.342	0
K 9-1-1-B	0.340	0.387	0
N 1253-9-1-1-B	0.536	0.424	0
N 17-1-1-1-B-B	0.727	0.439	0
N 57-2	0.635	0.320	0
N 68-1-5-1-B	0.615	0.363	0
N 406-61-3-19-1-B-B	0.588	0.377	0
N 413-1-BB	0.674	0.396	0
N 1032-3-1-1-1-B	0.679	0.384	0
N 1039-1-1-B-1-B	0.691	0.399	0
N 1040-2-1-B	0.604	0.374	0
N 1119-3	0.720	0.394	0
SC 895-1-B-2-B	0.744	0.523	0
SC 896-1-1-B	0.901	0.511	0.0001
SC 936-1-1-B	0.623	0.476	0
SC 939-3-1-B	1.12	0.426	0
SC 967-1-1-B	1.13	0.316	0
SC 4516-1-B	0.862	0.398	0
SC 4520-B	0.641	0.363	0
Early Russian	0.868	0.400	0
PI 153517-1-1-B	1.02	0.373	0
PI 156618-1-1-B	0.735	0.404	0
PI 158040-B-1-B	0.748	0.449	0
PI 158049-B-2-B	0.485	0.337	0
PI 158056-B-1-B	0.725	0.589	0.0003
PI 158058-B-1-B	0.604	0.406	0.0001
PI 158062-1-1-B	0.868	0.501	0
PI 158073	0.714	0.514	0.0002
PI 158901-2-6-B	0.582	0.434	0.0004
PI 158935-1-1-B	0.615	0.460	0
PI 162563-2-1-B	0.892	0.445	0
PI 198991-B	0.425	0.131	0
Mean	0.699	0.408	0
Standard deviation	0.184	0.079	....
Coefficient of variation	26.3	19.4	....

The accelerated-ageing study was conducted on the dehiscent check K-10 and three experimental indehiscent strains, all grown in 1953 at College Station. Seed samples, in open 600-ml. beakers, were placed in an oven at 100°F., and the oil from a portion of each sample was expressed periodically and analyzed. At the same time samples of oil expressed from each strain at the start of the experiment were placed in cork-stoppered bottles and put into the same oven. These oils were likewise periodically analyzed.

The oil samples were prepared by cold-pressing the seed and filtering the oil exudate. They were analyzed for the three constituents by the method of Suarez *et al.* (13) except when interference in the ultraviolet method for sesamin became appreciable, in which case the chromatographic method of Beroza (3) was used. The sesamolin data were based on a known sesamolin solution run at the same time (3).

Fat acidity was determined by the A.O.A.C. method (1) and expressed as the percentage of free fatty acids as oleic. The peroxide values were obtained by the method of King *et al.* (10) and expressed as milliequivalents per kilogram of oil.

The seed used to study the effect of frost damage was of the K-10 strain, grown at Lubbock, Tex., in 1952.

Effect of Strain

The data on the effect of strain are assembled in Table I. The sesamin content of the oils ranged from 0.340 to 1.13% and averaged 0.699% with a standard deviation of 0.184%. The coefficient of variation was 26.3%. The sesamolin content ranged from 0.131 to 0.589% and averaged 0.408% with a standard deviation of 0.079%. The sesamolin content was apparently somewhat less subject to variation since its coefficient of variation was 19.4%. In view of the low values obtained for sesamol no statistical treatment of the data on this compound was made.

Effect of Location

Table II presents the data on the sesamin, sesamolin, and sesamol content of the oil from seed of four strains grown at six locations. Analyses of variance of the untransformed data for sesamin and sesamolin indicated that only differences in sesamin content due to strains were appreciable (significant at the 1% level). These data indicated no consistent differences in sesamolin content due to strains, nor was either sesamolin or sesamin consistently influenced by location. The data indicate that strain x location interactions may influence both sesamin and sesamolin content; but since samples from the replications at the various locations were not subjected to chemical analysis, no general error term was available to test the statistical significance of these interactions.

TABLE II

Percentage of Sesamin, Sesamolin, and Sesamol in Oil from Four Strains of Sesame Seed Grown at Six Locations in 1953

Location	K-10	Venezuela 51	R 44-B-1-B	R 329	Location Mean
		Sesamin			
Mesa, Ariz.	0.075	0.925	0.764	0.431	0.549
Clemson, S. C.	0.375	0.779	0.541	0.518	0.553
Florence, S. C.	0.165	0.770	0.384	0.407	0.432
College Station, Tex.	0.531	0.767	0.588	0.507	0.598
Lubbock, Tex.	0.318	0.559	0.402	0.355	0.408
Temple, Tex.	0.233	0.583	0.686	0.590	0.523
Strain mean	0.283	0.730	0.561	0.468	0.510
		Sesamolin			
Mesa, Ariz.	0.176	0.350	0.388	0.251	0.291
Clemson, S. C.	0.376	0.352	0.249	0.355	0.333
Florence, S. C.	0.254	0.315	0.082	0.252	0.226
College Station, Tex.	0.389	0.291	0.205	0.273	0.290
Lubbock, Tex.	0.331	0.277	0.197	0.238	0.261
Temple, Tex.	0.330	0.281	0.346	0.358	0.329
Strain mean	0.309	0.311	0.244	0.288	0.288
		Sesamol			
Mesa, Ariz.	0.0001	0.0003	0.0002	0.0002	0.0002
Clemson, S. C.	0.0002	0.0003	0.0002	0.0005	0.0003
Florence, S. C.	0.0002	0.0005	0.0004	0.0005	0.0004
College Station, Tex.	0.0000	0.0003	0.0002	0.0005	0.00025
Lubbock, Tex.	0.0004	0.0004	0.0003	0.0003	0.00035
Temple, Tex.	0.0004	0.0005	0.0004	0.0003	0.0004
Strain mean	0.0002	0.0004	0.0003	0.0004	0.0003

The coefficients of variation for the sesamin and sesamolin data were 26.1 and 25.5%, respectively.

Because of their limited scope, results of these experiments do not completely eliminate the possibility of increasing sesamolin (which is much more potent than sesamin as a synergist with pyrethrins) either by breeding or by environmental control. However it appears that increases in sesamin content would be much more readily attained by appropriate breeding procedures.

No statistical treatment of sesamol content was made because of the small variation in these determinations.

Effect of Aging

The first part of Table III shows the data obtained by exposing sesame oil, as the oil, in cork-stoppered bottles to 100°F. It should be pointed out that, after the corks came in contact with the oil, the stoppers fitted loosely and may have permitted the entry of air, which could have brought on rancidity. However the authors were primarily interested in the effects of ageing and rancidity on the sesamin, sesamolin, and sesamol contents rather than in the resistance of the oil itself to ageing and rancidity. At the 2-month analysis it was found that T50290-B-1-1 was rancid. A measure of rancidity—that is, peroxide value—was therefore determined. No appreciable changes in the sesamin, sesamolin, and sesamol content were observed until the oils became rancid. Rancidity was accompanied by a lower sesamin and sesamolin content and a higher sesamol content. The free fatty acids content tended to rise with rancidity, but not appreciably.

TABLE III

Ageing Tests on Sesame Oil

Strain and time of analysis	Sesamin %	Sesamolin %	Sesamol %	Free fatty acids (as oleic) %	Peroxides, m.e./kg.	Germination %
		Exposed to 100°F. as the Oil in Cork-Stoppered Bottles				
R 44-B-1-B-3						
Start	0.642	0.221	0.0000	1.54	....	36.7
20 days	0.65	0.224	0.0000	1.54	....	
2 months	0.652	0.23	0.0015	1.58	3.9	
3 months	0.522 [a]	0.199	0.0017	1.64	188 [b]	
R 329-2-7						
Start	0.469	0.252	0.0000	2.42	....	27.0
20 days	0.472	0.255	0.0001	2.41	....	
2 months	0.47	0.26	0.0000	2.40	2.0	
3 months	0.46	0.259	0.0002	2.30	14.7	
T-50290-B-1-1						
Start	0.555	0.263	0.0000	1.36	....	42.7
20 days	0.545	0.270	0.0000	1.34	....	
2 months	0.44 [a]	0.227	0.0020	1.36	242 [b]	
3 months	0.31 [a]	0.191	0.0049	1.50	364 [b]	
K-10-1-1-1						
Start	0.648	0.374	0.0000	0.22	....	97.3
20 days	0.627	0.391	0.0001	0.20	....	
2 months	0.56 [a]	0.36	0.0009	0.20	121 [b]	
3 months	0.53 [a]	0.326	0.0023	0.25	257 [b]	
		Exposed to 100°F. as the Seed in Open Beakers [c] (For Analyses at start see above)				
R 44-B-1-B-3						
6 months	0.661	0.231	0.0008	1.87	1.8	
R 329-2-7						
6 months	0.467	0.264	0.0011	2.49	1.0	
T-50290-B-1-1						
6 months	0.535	0.278	0.0010	1.64	1.9	
K-10-1-1-1						
6 months	0.637	0.399	0.0003	0.22	0.5	

[a] Analyzed by chromatography (2).
[b] Rancid.
[c] Analyses, conducted at approximately monthly intervals over the 6-month period were sufficiently similar, so that only the original and final data are given.

The ultraviolet spectra of the rancid oils were very much altered. The 288 mμ maximum was shifted toward 280 mμ; the minimum, usually about 257 mμ, was shifted in some cases to 263 mμ. The amount of absorbance at the minimum was so great that the ratio of the absorbance at the foregoing minimum over that at the maximum, normally below 20%, was as high as 85% in rancid oils. Because of this large background absorbance the spectrophotometric determination for sesamin could not be applied to the rancid oils. Judging by odor, oil K-10-1-1-1 after 2 months seemed to have just turned rancid.

The second part of Table III shows that sesame oil exposed to 100°F. as the seed in open 600-ml. beakers was much more resistant to rancidity than when the oil itself was exposed. The sesamin and sesamolin content were unaffected over a 6-month period although the sesamol content rose toward the end of this period. The free fatty acids content also tended to be slightly higher. None of the oils became rancid for the peroxide values remained low and even showed a tendency toward becoming lower.

It should be pointed out that threshing methods required for the three experimental indehiscent strains were much more severe than for the dehiscent check, K-10. Damage in threshing of the indehiscent strains resulted in the lower germination percentages shown in Table III. K-10 had little or no original seed damage, a fact that is reflected in its high germination percentage. Any influence on experimental findings must be considered in the light of differences in damage in the original seed. The materials were chosen with this point in mind. It is heartening to note that the seed damage in the three indehiscent strains did not impair the ability of these seeds to resist rancidity upon exposure to 100°F. for six months.

The data in the first part of Table III show that the oils turned rancid before any appreciable amount of sesamol was formed. Furthermore there is no relationship between the amount of sesamolin and ability to resist rancidity. Under the conditions of the test it appears that the sesamolin content did not affect the stability of the oil. As pointed out by Budowski *et al.* (5), hydrogenation—or more correctly hydrogenolysis because of our knowledge of the formula of sesamolin (4)—of the sesamolin in the oil liberates sesamol, which markedly stabilizes the oil. The presence of sesamolin therefore contributes toward the stability of sesame oil when the oil is hydrogenated. Budowski and coworkers further showed that alkali-refining or deodorization will remove free sesamol and thereby reduce the resistance of the oil to oxidative rancidity.

Effect of Frost

The previous finding that the sesamin and sesamolin contents of the oil are lowered after frost damage of the seed (3) was confirmed by the data in Table IV. Again the sesamin content was more markedly lowered by frost damage than the sesamolin content. Since the purpose that sesamin and sesamolin serve in the plant is unknown, it is possible that these compounds aid in the biological defense of the plant.

TABLE IV
Effect of Frost on Percentages of the Three Constituents of Sesame Oil

Time of Harvest	Sesamin	Sesamolin	Sesamol
Before frost	0.404	0.349	0.0003
After first light frost	0.247	0.292	0.0001
After second light frost	0.159	0.220	0.0000

Summary

The effect of strain and location grown on the sesmin, sesamolin, and sesamol content of oils from sesame seed chosen to represent a wide variety of genetic material is reported. Only differences in sesamin content due to strain were significant.

Three of four oils exposed as the oil to 100°F. became rancid in two to three months. Rancidity of the oil was accompanied by lesser sesamin and sesamolin contents, and the ultraviolet spectrum of the oil was much altered. Oil from seed exposed as the seed to the same conditions for six months did not become rancid even though most of the seeds were damaged in threshing. The sesamol content of all the oils subjected to the accelerated ageing procedure increased, but the increase was greatest in the rancid oils.

Frost damage of sesame seed markedly diminished the sesamin and sesamolin content of the oil.

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REFERENCES

1. Association of Official Agricultural Chemists, Official Methods of Analysis, 7th ed., p. 197 (1950).
2. Feroza, M., J. Am. Oil Chemists' Soc., *31*, 302-305 (1954).
3. Beroza, M., Anal. Chem., *26*, 1173-1176 (1954).
4. Beroza, M., J. Am. Chem. Soc., (in press).
5. Budowski, P., Menezes, F. G. T., and Dollear, F. G., J. Am. Oil Chemists' Soc., *27*, 377-380 (1950).
6. Eagleson, C., U. S. Patent 2,202,145 (May 28, 1940); Soap and Sanit. Chem., *18*(12), 125 (1942).
7. Gersdorff, W. A., Mitlin, N., and Beroza, M., J. Econ. Entomol., *47*, 839-842 (1954).
8. Haller, H. L., McGovran, E. R., Goodhue, L. D., and Sullivan, W. N., J. Org. Chem., *7*, 183-185 (1942).
9. Haller, H. L., LaForge, F. B., and Sullivan, W. N., J. Org. Chem., *7*, 185-188 (1942).
10. King, A. E., Roschen, H. L., and Irwin, W. H., Oil and Soap, *10*, 105-109 (1933).
11. Kinman, M. L., and Martin, J. A., Agr. J., *46*(1), 24-27 (1954).
12. Kinman, M. L., and Stark, S. M. Jr., J. Am. Oil Chemists' Soc., *31*, 104-108 (1954).
13. Suarez, C. C., O'Connor, R. T., Field, E. T., and Bickford, W. G., Anal. Chem., *24*, 668-671 (1952).
14. Synerholm, M. E., and Hartzell, A., Contrib. Boyce Thompson Inst., *14*, 79-89 (1945).
15. Synerholm, M. E., Hartzell, A., and Cullmann, V., Contrib. Boyce Thompson Inst., *15*, 35-45 (1947).
16. Wachs, H., Science, *105*, 530-531 (1947).

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ERRATUM

E. W. Eckey writes that an error has been made in the paper entitled "Production of Polyvinyl Esters by Ester Interchange," by himself, R. O. Alderson, and R. J. Woestmann in the April issue of the Journal of the American Oil Chemists' Society. The error was apparently made in make-up at the printer's since it did not appear in the galley proof. Three lines were omitted from one paragraph, and three lines from the succeeding paragraph were inserted. As a result, these three lines appear twice.

On page 187 the last three lines of the paragraph beginning "Hydrogenated Sardine Oil" (near the bottom of the first column) should read thus: "used. It had the following characteristics: Acid V., 0.3; Sap. V., 192; I.V., 123; Capillary Melting Point, 60° C.; Cloud Point, 40°C."