

Table III. Aroma-Bearing Fractions from Separation of Aroma Concentrate from Blue Cheese (Conditions as in Figure 1 and Table II)

| Fraction | Odor description | Strength |
|----------|---|----------------------|
| 10 | Sweet, ether-like | Faint, fleeting |
| 18 | Sweet, ether-like, mucilage | Faint to no response |
| 21 | Ether-like, medicinal | Faint |
| 28 | Pleasant, Blue Cheese, paint, oily | Strong |
| 31 | Unpleasant, rotten cheese, musty, dirty socks | Strong |

These results indicate the capability and versatility of the descending silica gel method for simplifying complex mixtures of volatile constituents. But equally important, the method as described provides an easy means for locating those components which contribute to a particular aroma. For example, in separations of the test mixtures, the ester-containing fractions were located by simply "sniffing" the fractions after evaporation of the Freon. Since Freon 11 is virtually odorless, the peak ester fractions could often be located even before evaporation. Sniffing of Freon 11 presents no toxicological problems since it is "less toxic than CO₂" (Merck Index, 1960).

Application to Aroma Concentrates from Foods. *Bananas.* Table I summarizes a separation of a banana aroma concentrate. The characteristic aroma notes of the banana fruit were detected in relatively few fractions. Identification of a representative group of compounds in each fraction indicated the expected separation by functional group class as well as some separation of saturated from unsaturated components within the classes.

Quast (1970) has scaled up this separation considerably and reports further details on the separation and identification of volatile constituents present in ripe bananas, particularly those components occurring in fractions with typical banana aroma.

Coffee. Table II summarizes the results of separation of a coffee aroma concentrate. The oil appeared to have deteriorated somewhat during transport, as a black residue had settled out. A 1:20 dilution of the oil in the water was generally characterized as a strong or stale coffee aroma.

Identification of the components in aroma-bearing fractions of Table II was not attempted, but it is clear that separation of some typical flavor "notes" of coffee was achieved.

Blue Cheese. When Blue Cheese aroma concentrate was separated, the panel members were again able to locate several aroma-bearing fractions (Table III).

DISCUSSION

The procedure described in this report has proven exceptionally useful in our laboratories, both for routine separations of the constituents of aroma concentrates by functional class and for seeking sensory correlations. Our general approach now is to first separate crude aroma concentrates on silica gel and then to concentrate identification efforts (*via* combined glc-mass spectrometry) on those fractions with typical aroma notes. Since the selected fractions normally contain relatively few components of the same functional class, identification generally presents few problems. Further studies of the aroma impact of individual components can then be carried out with pure samples. Where particular fractions are too complex, the silica gel separation is easily repeated with appropriate adjustment of elution conditions.

The procedure has been used primarily with fruit flavors (Table I; Quast, 1970), but the results with coffee and Blue Cheese (Tables II and III) suggest general applicability.

ACKNOWLEDGMENT

I thank Jack Fisher, Walter Brandes, and Sonia Valenzuela Quast for their technical assistance.

LITERATURE CITED

- Day, E. A., in "The Chemistry and Physiology of Flavors," Schultz, H. W., Day, E. A., Libbey, L. M., Ed., Avi, Westport, Conn., 1967, p 353.
 Merck Index of Chemicals and Drugs, 7th ed., Merck & Co., 1960, p 1060.
 Murray, K. E., Stanley, G., *J. Chromatogr.* 34, 174 (1968).
 Murray, K. E., Palmer, J. K., Whitfield, F. B., Kennett, B. H., Stanley, G., *J. Food Sci.* 33, 632 (1968a).
 Murray, K. E., Shipton, J., Whitfield, F. B., *Aust. J. Chem.* 25, 1921 (1972).
 Murray, K. E., Shipton, J., Whitfield, F. B., Kennett, B. H., Stanley, G., *J. Food Sci.* 33, 290 (1968b).
 Quast, S. V., "Volatile Constituents of Ripening Banana Fruit," Ph.D. Thesis, Massachusetts Institute of Technology, 1970.

James K. Palmer

Department of Nutrition and Food Science
 Massachusetts Institute of Technology
 Cambridge, Massachusetts 02139

Received for review November 16, 1972. Accepted June 15, 1973.

Potential Juvenile Hormone Activity: Preparation of Fatty β -Methylcrotonyl and 3,4-Methylenedioxyphenyl Derivatives

Certain compounds with β -methylcrotonyl and 3,4-methylenedioxyphenyl groups are known to possess juvenile hormone activity because they prevent insect maturation and reproduction. A series of compounds containing these groups has been prepared from fatty acids and their deriva-

tives. These chemicals showed little or no juvenile hormone activity at 10-100 μ g/insect in the yellow mealworm, *Tenebrio molitor* L. Of the compounds tested an isomeric mixture of β -methylacrylates from linseed oil-derived C₁₈ aromatic cyclic acids showed the greatest activity.

A number of insect juvenile hormones have been isolated, identified, and their chemical analogs synthesized (Bowers *et al.*, 1965, 1966; Dahm *et al.*, 1967; Myer *et al.*, 1968; Röllner and Bjerke, 1965; Williams, 1967). The juvenile hormone activity of these chemicals seems related to

the trans double bond at the 6,7 position, β -methylcrotonyl, epoxy, and branch-chain groups. Certain other compounds used as insecticide synergists having a high order of juvenile hormone activity contain the 3,4-methylenedioxyphenyl group (Bowers, 1968, 1969).

Table I. Juvenile Hormone Activity

| Compound | Test compounds (predominant isomers) | Activity at indicated μg level ^a | | | Gas-liquid chromatography analysis, % | C, % | | H, % | |
|----------|---|--|-----|-----|---------------------------------------|-------|-------|-------|-------|
| | | 10 | 50 | 100 | | Calcd | Found | Calcd | Found |
| 1 | Methyl 11-(2- <i>n</i> -propylphenyl)-3-methyl-2-undecenoate | 1 | 1 | | 99+ | 79.94 | 80.61 | 10.37 | 10.63 |
| 2 | Methyl 3-methyl-2-nonadecenoate | 1 | 1 | | 99+ | 77.72 | 77.09 | 12.42 | 12.45 |
| 3 | Piperonyl 9-(2- <i>n</i> -propyl-3,5-cyclohexadienyl)nonanoate ^b | 1 | | 1 | 92.7 | 75.34 | 75.26 | 8.60 | 9.12 |
| 4 | Piperonyl 8-(<i>p</i> - <i>n</i> -hexylphenyl)octanoate | 1 | | 1 | 99+ | 82.61 | 79.95 | 9.20 | 9.62 |
| 5 | Methyl 3-methyl-2-eicosenoate ^c | 0 | 0-1 | | 93 | 78.05 | 69.42 | 12.50 | 11.53 |
| 6 | Methyl 3-methyl-2,11-eicosadienoate | 0 | 0-1 | | 99 | 78.51 | 78.79 | 11.98 | 12.32 |
| 7 | Methyl 9-(4-hexyl-2-cyclohexenyl)-3-methyl-2-nonenoate | 0 | | 0-1 | 98 | 79.49 | 82.38 | 11.68 | 12.01 |
| 8 | Piperonyl 9-(<i>o</i> - <i>n</i> -propylphenyl)nonanoate | 0 | | 0-1 | 99+ | 75.73 | 75.02 | 8.13 | 8.43 |
| 9 | Piperonyl 11-(2- <i>n</i> -propylphenyl)-3-methyl-2-undecenoate ^d | 0 | 0 | 0-1 | 99+ | | | | |
| 10 | Methyl 3-methyl-11(12)-epoxy-2-eicosenoate ^e | | 0 | 0 | | 74.95 | 71.31 | 11.44 | 11.05 |
| 11 | Methyl (4- <i>n</i> -hexyl-2,3-epoxycyclohexyl)-3-methyl-2-nonenoate ^f | 0 | 0 | 0 | | | | | |
| 12 | Methyl 9-(3,4-methylenedioxyphenyl)-8-formyl-8-nonenoate ^g | 0 | 0 | 0 | 99+ | | | | |

^a Juvenile hormone activity of compounds in "tenebrio genitalia" test, degrees of modification are represented numerically: 0, no effect; 0-1, retention of immature genital characters in 10-50% of the insects treated; 1, retention of one or more partially immature genital characters in 80% of insects tested. ^b Contained 7.3% methyl ester of C₁₈ unsaturated cyclic acids derived from linseed oil. ^c Contained 7% methyl heptadecyl ketone. ^d Compound No. 1 transesterified with piperonyl alcohol. ^e Compound No. 6; 90% epoxidized. ^f Compound No. 7; 93% epoxidized; contained 5% 2,3-dihydroxycyclohexyl derivative. ^g Condensation product of 99.8% pure methyl azelaaldehyde and piperonal.

This communication reports the preparation of new compounds derived from linseed and soybean oils containing β -methylcrotonyl and 3,4-methylenedioxyphenyl groups. The compounds are isomeric mixtures prepared in small amounts and screened for juvenile hormone activity. Purity of the compounds (>93%) was determined by gas-liquid chromatography and elemental analyses.

The fatty β -methylcrotonyl derivatives were synthesized in three steps: (a) condensation of fatty methyl esters with methylsulfinyl carbanion; (b) aluminum amalgam reduction of resultant β -keto sulfoxides to corresponding methyl ketones (Corey and Chaykovsky, 1965); and (c) reaction of phosphonate carbanion and fatty ketones to yield methylcrotonyl esters (Wadsworth and Emmons, 1961). Fatty 3,4-methylenedioxyphenyl derivatives were prepared by the condensation of piperonal with fatty aldehydes and by esterification of piperonyl alcohol with fatty acid derivatives.

The cyclohexenyl and isolated bonds of certain β -methylcrotonyl esters were selectively epoxidized with equimolar amounts of *m*-chloroperbenzoic acid.

A number of these chemicals showed juvenile activity at 10-100 μg /insect when applied topically to pupae of the yellow mealworm, *Tenebrio molitor* L. (Bowers, 1968, 1969). Of the compounds tested the greatest activity was shown by an isomeric mixture of β -methylacrylates prepared from C₁₈ aromatic cyclic acids. The compounds are listed in Table I in decreasing order of the activity. Because of the low activity, the compounds were not purified further, and no attempt was made to separate possible active isomers. The activity of the compounds is not sufficient for commercial consideration.

Structures given for the compounds were confirmed by elemental analysis, infrared, and nuclear magnetic resonance spectra.

ACKNOWLEDGMENT

George McManis performed the infrared analyses, Curtis Glass performed the nuclear magnetic resonance anal-

yses, and Clara McGrew, Bonita Heaton, and Karen Jones did the microanalyses.

LITERATURE CITED

- Bowers, W. S., *Science* 161, 895 (1968).
 Bowers, W. S., *Science* 164, 323 (1969).
 Bowers, W. S., Fales, H. M., Thompson M. J., Eubel, E. C., *Science* 154, 1020 (1966).
 Bowers, W. S., Thompson, M. J., Uebel, E. C., *Life Sci.* 4, 2323 (1965).
 Corey, E. J., Chaykovsky, M., *J. Amer. Chem. Soc.* 87, 1345 (1965).
 Dahm, K. H., Trost, B. M., Röller, H., *J. Amer. Chem. Soc.* 89, 5292 (1967).
 Myer, A. C., Schneiderman, H. A., Hanzmann, E., Ko, J. H., *Proc. Nat. Acad. Sci. U. S.* 60, 853 (1968).
 Röller, H., Bjerke, J. S., *Life Sci.* 4, 1617 (1965).
 Wadsworth, W. S., Jr., Emmons, W. D., *J. Amer. Chem. Soc.* 83, 1733 (1961).
 Williams, C. M., *Sci. Amer.* 217, 13 (1967).

Edward W. Bell
 Lyle E. Gast
 John C. Cowan*
 John P. Friedrich
 William S. Bowers¹

Northern Regional Research Laboratory
 Agricultural Research Service
 U. S. Department of Agriculture
 Peoria, Illinois 61604

¹ Entomology Research Division
 Beltsville, Maryland 20705

Received for review October 20, 1972. Accepted April 9, 1973. Mention of firm names or trade products does not imply endorsement or recommendation by the Department of Agriculture over other firms or similar products not mentioned. Presented at the AOCS Meeting, Minneapolis, Minn., October 1969.