authenticity of the acetates was established by alkaline hydrolysis to the starting oximes.

Conversion of VII to VIII. Five hundred milligrams of VII was dissolved in 50 ml. of 95% alcohol and refluxed. Samples were taken at one-hour intervals and after appropriate dilution measured in the spectrophotometer at 223 mμ. As can be seen from Table II the absorption peak disappears within 3 hr. After 6 hr. the solvent was evaporated yielding a pale yellow oil which could not be crystallized. The oil was insoluble in aqueous sodium carbonate and could not be extracted out of an ether solution with alkali.

TABLE II

REARRANGEMENT OF VII IN BOILING ALCOHOL

Time, hours	ϵ_{223}
0	9,400
1	1,410
3	427
4	427

Alkaline hydrolysis of VIII. To 500 mg. of the oil dissolved in 50 ml. of 95% alcohol 2.5 g. of potassium hydroxide was added and the solution refluxed for 20 hr. On cooling, the solution was evaporated *in vacuo* and the residue dissolved in 50 ml. of water. Ether extraction removed 60 mg. of yellow oil. Addition of concentrated hydrochloric acid to the clear aqueous phase precipitated a mixture of X and XI.

Isolation of 3β-hydroxy-16,17-seco-16,17-dioic acid 17-amide (XI). To the acidified aqueous suspension 60 ml. of chloroform was added and the whole shaken in a separatory funnel during which X passed into the chloroform phase. After separation, the aqueous suspension was filtered and the precipitate washed thoroughly with water and dried. This yielded 220 mg. of material, m.p. 214–218°. After 6 recrystallizations from methanol followed by drying *in vacuo* over phosphorous pentoxide at 120° needle crystals melting at 218.5–220.5° were obtained. The compound was completely insoluble in chloroform, benzene, ether, and ethyl acetate but was soluble in dilute aqueous sodium carbonate. Titration indicated that it was a monobasic acid. When dissolved in cold acetic acid and treated with a cold solution of sodium nitrite, no evolution of nitrogen was evident which confirmed the tertiary nature of the amide grouping.

Anal. Calcd. for $C_{19}H_{31}O_4N$: C, 67.65; H, 9.46; N, 4.16. Found: C, 67.67, 67.61; H, 9.36, 9.40; N, 4.25.

Isolation of 3β-hydroxy-16,17-seco-androstan-16,17-imide (X). The chloroform solution obtained in the isolation of XI was washed free of acid and dried over sodium sulfate. Evaporation of the chloroform solution *in vacuo* yielded 136 mg. of a yellowish flaky solid which melted at 100–110°. This was dissolved in 4 ml. methanol and a few drops of water added which removed the yellow impurity. The solution was then centrifuged and the supernatant removed. The concentration of the methanol solution was then adjusted by boiling and adding water so that on cooling the material crystallized out as fine colorless needles. After drying *in vacuo* at 120° over phosphorus pentoxide for 1.5 hr. 113 mg. of X melting at 180–182.5° was obtained. It was readily soluble in dilute aqueous sodium carbonate. On refluxing a sample in 10% aqueous potassium hydroxide for 24 hr. it was converted into XI.

Anal. Calcd. for $C_{19}H_{29}O_3N$: C, 71.48; H, 9.09; N, 4.38. Found: C, 71.55, 71.92; H, 8.97, 9.16; N, 4.72.

Conversion of XI to 3β-hydroxy-16,17-seco-androstan-16,17-dioic acid (VII). Forty milligrams of the pure semi-amide (XI) was added to 5 ml. of a solution of 20% potassium hydroxide in glycerol. The flask was placed in an oil bath and the temperature gradually raised to 200° whereupon a copious evolution of bubbles of a basic gas took place. After 2.5 hours heating was stopped. The material came out of solution on cooling but on dilution with water a clear, slightly yellow, solution was obtained. This was acidified and after standing for 2 hr. it was filtered and washed well with water. It was dried on suction and finally *in vacuo* over phosphorus pentoxide at 120°. This yielded 20 mg. of material which melted at 234–237° with previous softening at 225°. After one recrystallization from methanol the melting point was 237.5–238.5°. It titrated as a dibasic acid and sodium fusion indicated the absence of nitrogen. On admixture with an authentic sample of XII there was no depression in the melting point.

OTTAWA, CANADA

[CONTRIBUTION FROM U. S. DEPARTMENT OF AGRICULTURE]

Separation of Aliphatic Disulfides and Trisulfides by Gas-Liquid Partition Chromatography

JOHN F. CARSON AND FRANCIS F. WONG

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The polar stationary phases, Carbowax and Reoplex, and a nonpolar phase, Apiezon M, have been compared in the separation of aliphatic disulfides and trisulfides by gas-liquid partition chromatography. Mixtures of disulfides and trisulfides can be separated at 150° without decomposition. The polar phases are particularly useful for separating unsaturated disulfides from the corresponding saturated compounds.

This paper reports the application of gas-liquid partition chromatography to the separation and isolation of some simple aliphatic disulfides and trisulfides in connection with a study of the volatile components of onions.[1] A number of investigators have studied the separation of thiols and sulfides. Sunner, Karrman, and Sunden[2] reported quantitative separation of a number of thiols by gas-liquid partition chromatography, and Ryce and Bryce[3]

(1) Presented at the Joint Symposium of Analytical and Petroleum Chemistry, American Chemical Society meeting, New York, September 1957.

(2) S. Sunner, K. J. Karrman, and V. Sunden, *Mikrochim. Acta*, 1144 (1956).

(3) S. A. Ryce and W. A. Bryce, *Anal. Chem.*, **29**, 925 (1957).

separated mixtures of low-boiling mercaptans and sulfides and one disulfide, dimethyl disulfide. Amberg[4] has reported relative retention times for 8 thiols, 5 sulfides, and 11 thiophenes, and recently Coleman,[5] Thompson, Ward, and Rall have separated and identified a number of low-boiling mercaptans and sulfides in crude oil by gas-liquid partition chromatography. However, little has been reported on the separation of disulfides and trisulfides.

The separation of disulfides and trisulfides by distillation, especially in the presence of mercaptans, is difficult. Ionic displacement by mercaptide ion may produce new disulfides and mercaptans as artifacts as in

$$RS^- + R'SSR' \rightleftharpoons RSSR' + R'S^-$$
$$\Big\downarrow RS^-$$
$$RSSR + R'S^-$$

Fava, Iliceto, and Camera[6] have shown the importance of this displacement even under mild conditions. The production of artifacts by free radical decomposition of disulfides and trisulfides is particularly facile as demonstrated by Birch.[7] A third type of decomposition applicable to allylic disulfides is thermal decomposition in the presence of metals as zinc to yield the allylic monosulfides, metallic sulfide, and polymeric material.[8] Conditions have been found for the separation of a number of disulfides and trisulfides by gas-liquid partition chromatography without appreciable decomposition. In particular, unsaturated disulfides may be separated from the corresponding saturated compounds.

In this study, the aim has been to separate these compounds in quantities sufficient for identification by infrared methods. In some cases, by chromatographing several times, sufficient material could be isolated for identification by the formation of crystalline derivatives.[9]

EXPERIMENTAL

Apparatus. The gas-liquid partition chromatography apparatus was of the coiled tube type patterned after Dimick and Corse.[10] The columns were of stainless steel, 1/4 in. O.D. and 5 to 7 ft. in length coiled to fit into a 1 gal. stainless-steel Dewar vessel containing heating liquid, mechanical stirrer, and thermometer. The katharometer was a stainless-steel block, immersed in the heating bath, with two Gow-Mac filaments[11] operated at 175 ma. Electrical imbalance was measured on a recording potentiometer with a 2- or 4-mv. full scale deflection. The bath temperature was generally maintained with a maximum variation of ±0.5°. Helium temperature at the inlet injection chamber was maintained at approximately 20° higher than the column operating temperature. Samples were injected as liquid with a microsyringe. The stationary phases consisted of firebrick (C-22, Johns Manville) ground to 40–60 mesh, acid washed, and then heated to 400° and impregnated with the appropriate organic phase (dissolved in acetone or hexane) in the ratios of firebrick to liquid phase of 4:1 (by weight).

For larger scale separations a helical stainless-steel column, 1/2 in. O.D. and 5.5 ft. in length was used. The stationary phase consisted of 1 part of organic liquid to 2 parts of firebrick. This column could handle approximately 6 times the quantities used with the smaller columns. Although resolution was not so good as with the corresponding 1/4 in. column, partial purification could be obtained, and the fractions were then later purified on the smaller columns.

Preparation of compounds. All of the disulfides with the exception of methyl-*n*-propyl disulfide, di-*iso*propyl and *n*-propyl-*iso*propyl disulfides and allyl-*n*-propyl and diallyl disulfides were Eastman Chemicals. The trisulfides were synthesized as described.

Allyl propyl disulfide. The preparation of the compound is described in detail, since it has not been heretofore described in the literature. Allyl mercaptan was prepared by alkaline hydrolysis of allyl isothiourea hydrobromide and the oily product resulting on acidification was used immediately without purification. A mixture of crude allyl mercaptan, 16 g., 0.22 mole, and *n*-propyl mercaptan, 12 g., 0.16 mole, was dissolved in a cold solution of 20 g. of sodium hydroxide in 200 cc. of water. A solution of 122 g., 0.37 equiv., of potassium ferricyanide in 400 cc. of water was added in 10-cc. portions over a 2-hr. period with mechanical stirring and cooling in an ice bath, and stirring was continued for 2 hr. longer. The resulting yellow milky emulsion was extracted with ether, the ether extract was dried with calcium chloride, and after removal of ether *in vacuo*, the resulting liquid was distilled at 5 mm. Hg from a water bath at 62°–70° to yield 19.8 g. (*ca.* 70%) of pale yellow distillate. The disulfide mixture was separated into dipropyl, allyl propyl, and diallyl disulfides by passage through a gas-liquid partition column of coiled stainless steel, 1/2 in. O.D. and 5.5 ft. in length, packed with a stationary phase consisting of 1 part of Carbowax 1540 on 2 parts of 40–60 mesh firebrick. Quantities of 160–180 μl. of crude disulfide mixture were injected with a helium flow rate of 180 cc./min. and a column temperature of 140°. The collected fractions were further purified by chromatography on a 1/4 in. O.D. column with the same packing. Measurement of peak areas indicated the composition of the disulfide mixture to be approximately diallyl, 32%, allyl propyl disulfide, 41%, and dipropyl disulfide, 23%, with approximately 4% of low-boiling material (partly diallyl sulfide). For the preparation of allylic disulfides, the ferricyanide oxidation procedure was found to be superior to the usual iodine oxidation which often led to excessive tar formation.

Methyl-n-propyl disulfide was prepared by alkaline ferricyanide oxidation of a mixture of 0.375 mole of methyl mercaptan and 0.21 mole of *n*-propyl mercaptan. The disulfide was isolated from the crude distillate by gas-liquid partition chromatography under conditions similar to those used with the mixed allyl propyl disulfides. Peak areas indicated the composition of the distilled disulfide mixture to be dimethyl, 9.2%; methylpropyldisulfide, 38.5%, and dipropyldisulfide, 52.3%.

(4) C. H. Amberg, *Can. J. Chem.*, **36**, 590 (1958).

(5) H. J. Coleman, C. J. Thompson, C. C. Ward, and H. J. Rall, *Anal. Chem.* **30**, 1592 (1958).

(6) A. Fava, A. Iliceto, and E. Camera, *J. Am. Chem. Soc.*, **79**, 833 (1957).

(7) S. F. Birch, T. V. Cullum, and R. A. Dean, *J. Inst. Petroleum*, **39**, 206 (1953).

(8) F. Challenger and D. Greenwood, *J. Chem. Soc.*, 26 (1950).

(9) J. F. Carson and F. F. Wong, *J. Org. Chem.*, **22**, 1725 (1957).

(10) K. P. Dimick and J. Corse, *Food Technol.*, **10**, 360 (1956).

(11) Gow-Mac Co., 100 Kings Road, Madison, N. J. Mention of commercial names does not imply endorsement by the Department of Agriculture.

Di-isopropyl disulfide and isopropyl-n-propyl disulfide were similarly prepared from an equimolar mixture of isopropyl and *n*-propyl mercaptans. Yields of the three disulfides were di-isopropyl disulfide, 18.3%; isopropyl-*n*-propyl disulfide, 45.8% and di-*n*-propyl disulfide, 35.8%.

Methyl-n-propyl trisulfide and di-n-propyl trisulfide. These compounds were prepared by an adaptation of the procedure of Westlake, Laquer, and Smyth.[12] A mixture of dimethyl disulfide, 12 g. (0.13 mole), di-*n*-propyl disulfide, 10 g. (0.067 mole), sulfur, 7 g. (0.22 g. atom), and 0.5 ml. of di-*n*-butylamine were heated in an oil bath at 130–135° for 5 hr. An ethereal solution of the brown reaction solution was washed with dilute hydrochloric acid and water, dried over calcium sulfate and concentrated *in vacuo* to yield 25 cc. of a brown oil. Distillation *in vacuo* (1 mm.) (bath temp. 50–125°) yielded 20 ml. of yellow liquid distillate. Gas-liquid chromatography on a 1/2 in. O.D. column of Carbowax 1540 at 140° and 180 cc./min. of helium yielded 5 separate peaks corresponding to dimethyl disulfide, methyl-*n*-propyl disulfide, dimethyl trisulfide and di-*n*-propyl disulfide (not separated), methyl-*n*-propyl trisulfide, and di-*n*-propyl trisulfide. The unresolved peak containing dimethyl trisulfide and dipropyl disulfide was separated into the two components with an Apiezon column. Methyl-*n*-propyl trisulfide and di-*n*-propyl trisulfide were collected and purified by rechromatography on a 1/4-in. Carbowax column.

Dimethyl trisulfide was isolated as a by-product from the previous preparation and was also prepared by the procedure of Gorin and Dougherty.[13]

Diethyl trisulfide was prepared from diethyl disulfide, sulfur, and di-*n*-butylamine by the procedure of Westlake, Laquer, and Smyth.[12]

DISCUSSION

Mixtures of aliphatic disulfides and trisulfides up to and including di-*n*-butyl disulfide and di-*n*-propyl trisulfide can be separated by gas-liquid partition chromatography at 150° without serious decomposition. No exchange reactions occurred between mercaptans, disulfides, and trisulfides as demonstrated by the fact that mixtures of unsymmetrical disulfides and trisulfides in the presence of mercaptans could be chromatographed without the formation of any symmetrical disulfides or trisulfides and, similarly, chromatography of mixtures of the symmetrical compounds yields no corresponding unsymmetrical disulfide or trisulfide. Allylic disulfides could be separated without the formation of monosulfides. Absence of decomposition during chromatographic separation was confirmed by infrared analysis of collected fractions. Decomposition was observed, however, with one stationary phase, U.S.P. solid white petrolatum. Diallyldisulfide decomposed when injected into a petrolatum-firebrick column at temperatures of 130–150° as evidenced by a peak for diallyl sulfide and a long plateau between this peak and the disulfide peak. Collected fractions were yellow although the original disulfide was colorless. This behavior has not been observed with any of the other stationary phases tested.

(12) H. E. Westlake, Jr., H. L. Laquer, and C. P. Smyth, *J. Am. Chem. Soc.* **72**, 436 (1950).

(13) G. Gorin and G. Dougherty, *J. Org. Chem.*, **21**, 241 (1956).

TABLE I

RETENTION TIMES OF DISULFIDES AND TRISULFIDES

Com ound	Retention Times (Corr. for Dead Space)[a] Carbowax 1540,[b] 1/4" O.D., 6' × 5" length	Reoplex 400,[c] 1/4" O.D., 5' × 10" length	Apiezon M,[d] 1/4" O.D., 5' × 10" length
$(CH_3)_2S_2$	3.85 min.	3.20 min.	2.90 min.
$(C_2H_5)_2S_2$	7.30	5.90	8.00
CH_3S_2-*n*-C_3H_7	7.65	6.40	8.30
(*i*-$C_3H_7)_2S_2$	8.50	6.95	14.2
(*n*-C_3H_7)S_2-*i*-C_3H_7	11.2	9.05	18.6
(*t*-$C_4H_9)_2S_2$	11.8	9.30	23.6
(*n*-$C_3H_7)_2S_2$	14.3	11.8	22.3
CH_2=CH—$CH_2S_2CH_2CH_2CH_3$	17.7	14.9	20.3
$(CH_2$=CH—$CH_2)_2S_2$	21.7	18.3	18.4
(*i*-$C_4H_9)_2S_2$	18.1	15.1	38.0
(*n*-$C_4H_9)_2S_2$	31.8	25.6	65.4
(*i*-$C_5H_{11})_2S_2$	44.3	36.6	>110
$(CH_3)_2S_3$	15.9	12.8	12.3
$(C_2H_5)_2S_3$	25.1	20.8	30.4
CH_3S_3-*n*-C_3H_7	28.0	23.3	32.9
(*n*-$C_3H_7)_2S_3$	48.8	41.6	82.4
Air	0.75	0.70	0.60
Column efficiency[e] for (*n*-$C_3H_7)_2S_2$	806	905	680

[a] Retention times in minutes = time from air peak to peak maximum, column temp. = 150 ± 0.5°, helium flow rate 45 cc./min., recorder sensitivity 2 mv. or 4 mv. full scale, filament current = 175 ma. [b] One part of Carbowax 1540 to 4 parts firebrick (by weight), weight of stationary phase 28.0 g. [c] One part Reoplex 400 to 4 parts firebrick, weight of stationary phase = 22.2 g. [d] One part Apiezon M to 4 parts firebrick, weight of stationary phase = 21 g. [e] Given in number of theoretical plates for 3 μl. sample calculated by the formula:

$$n = \frac{16\,D^2}{W^2}$$ where D = distance from air peak to peak maximum and W = width of peak at the base measured between tangents to the peak inflection points [H. W. Johnson and F. H. Stross; *Anal. Chem.*, **30**, 1586 (1958)].

Table I lists retention times for a number of disulfides and trisulfides at 150° for three different substrates, two highly polar phases, Carbowax 1540[14] and Reoplex 400[15] and one extreme nonpolar phase Apiezon M.[16] Peaks were sufficiently sharp that a difference of 5–10% in retention times of two components gave two observable peaks for a mixture of the two components in equal proportions but without complete separation.[17] Thus, mixtures of diethyl disulfide and

(14) Union Carbide Chemicals Co., 30 East 42nd St., New York 17, N. Y.

(15) Geigy Pharmaceutical Division, Geigy Chemical Corp., Ardsley, N. Y.

(16) Metropolitan-Vickers Electrical Co., Ltd., London, England.

(17) Although complete separation cannot be attained in these cases, relative retention times are sufficiently reliable to constitute presumptive evidence for the identity of the compounds, particularly if determined with several substrates.

methyl-*n*-propyl disulfide could not be separated on Carbowax or Reoplex columns although two peaks were observable, and similarly mixtures of di-*n*-propyl, allyl propyl, and diallyl disulfide on Apiezon showed three distinct peaks without separation. A difference of 15% or more in retention times between components allowed complete separation (return of recorder pen to base line between peaks). Percentage variation in retention time for a given compound was less than 2% except for the slowest disulfides and trisulfides with retention times greater than 40 min. where reproducibility was within 4%. Table II records retention times relative to cyclohexanone as a standard (retention time/retention time of cyclohexanone) for the disulfides and trisulfides shown in Table I. These ratios are independent of small variations in temperature and flow rate but are quite variable if the column is overloaded. Relative retention times were substantially constant for quantities varying from 0.1 μl. to 3 μl. For quantities greater than 5 μl., the higher disulfides and trisulfides have delayed retention times, due to peak asymmetry, resulting in increased ratios.

TABLE II

RELATIVE RETENTION TIMES OF DISULFIDES AND TRISULFIDES (RELATIVE TO CYCLOHEXANONE)

Compound	Retention Time/Ret. Time of Cyclohexanone (Corr. for Dead Space)		
	Carbowax 1540, 1/4" O.D., 6' × 5" length	Reoplex 400, 1/4" O.D., 5' × 10" length	Apiezon M, 1/4" O.D., 5' × 10" length
Cyclohexanone	1.00	1.00	1.00
$(CH_3)_2S_2$	0.333	0.308	0.349
$(C_2H_5)_2S_2$	0.629	0.567	0.964
CH_3S_2-*n*-C_3H_7	0.660	0.615	1.00
(*i*-$C_3H_7)_2S_2$	0.733	0.668	1.71
i-$C_3H_7S_2$-*n*-C_3H_7	0.961	0.870	2.24
(*t*-$C_4H_9)_2S_2$	1.02	0.894	2.84
(*n*-$C_3H_7)_2S_2$	1.23	1.13	2.69
CH_2=CH—CH_2S_2-*n*-C_3H_7	1.53	1.43	2.45
(CH_2=CH—$CH_2)_2S_2$	1.87	1.76	2.22
(*i*-$C_4H_9)_2S_2$	1.56	1.45	4.58
(*n*-$C_4H_9)_2S_2$	2.74	2.46	7.88
(*i*-$C_5H_{11})_2S_2$	3.82	3.52	>13.00
$(CH_3)_2S_3$	1.37	1.23	1.48
$(C_2H_5)_2S_3$	2.16	2.00	3.66
CH_3S_3-*n*-C_3H_7	2.42	2.34	3.96
(*n*-$C_3H_7)_2S_3$	4.20	4.00	9.93

Disulfide and trisulfide peaks were generally symmetrical for quantities less than 5 μl. Larger quantities, particularly of the 6 carbon (or higher) disulfides or of the trisulfides showed highly skewed peaks with long fronts and sharp tails. This was more pronounced with the polar substrates than with Apiezon. A possible cause of this asymmetry is incomplete volatilization in the injection chamber with large samples, particularly since the temperature of the helium flowing through the injection chamber is lower than the boiling point of many of the compounds. Asymmetry of this type, long front and sharp tail, has been attributed to a type of nonlinear behavior where the partition coefficient (concentration of solute in stationary phase/concentration in gas phase) increases with increasing concentration of solute.[18]

The polar substrates, Carbowax and Reoplex, are particularly useful in that allylic disulfides can be separated from the corresponding saturated compounds. As expected from the known behavior of these materials, with other unsaturated compounds, allyl propyl disulfide and diallyl disulfide are retarded by these substrates and can be separated from each other and from di-*n*-propyl disulfide. Fig. 1 shows a separation of these disulfides with a

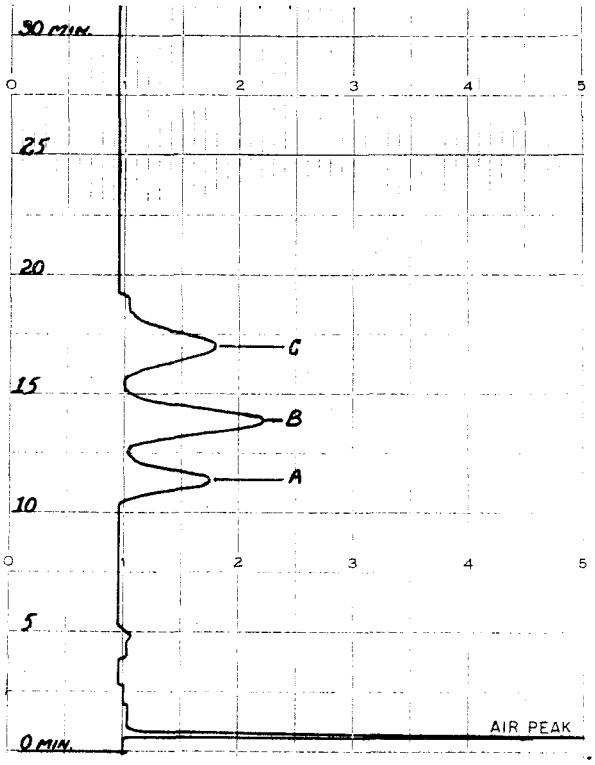

Fig. 1. Separation of mixed allyl and *n*-propyldisulfides. Reoplex 400 column at 150°; 45 ml. He/min.; filament current, 175 ma.; 4-mv. full scale deflection. Sample volume = 3/μl. A = Di-*n*-propyldisulfide; B = allyl-*n*-propyldisulfide; C = diallyldisulfide

1/4-in. Reoplex column. These particular substrates are also faster than the paraffinic types and the higher boiling disulfides and trisulfides can be eluted in a reasonable time. Similar behavior was experienced with other polyglycols. Octyl phenoxypolyethylene glycol (O.P.E. 30)[19] was

(18) P. E. Porter, C. H. Deal, and F. H. Stross, *J. Am Chem. Soc.*, **78**, 2999 (1956).

(19) Rohm and Haas Co., Washington Square, Philadelphia 5, Pa.

found to be very close to the polyethylene glycols in separating ability and retention time, but a sample of polypropylene glycol[14] was somewhat inferior in separating unsaturates. A silicone column (General Electric SF96-40) was found to be intermediate in retention time and separating ability between the polyglycol type and the paraffin type.

With Apiezon M, retention times generally followed boiling points, and the order of emergence of the allylic disulfides was reversed from that of the polar phases and separation was not complete. This stationary phase also had the disadvantage that higher disulfides and trisulfides had unusually long retention times. However, certain combinations not completely resolvable with the polar phases can be completely separated with Apiezon. Inspection of Table I or Table II shows that of the 16 disulfides and trisulfides listed, 9 pairs would probably not be completely separated on the polar substrates because the retention times are too close (< 15%). With the exception of the straight chain isomers, methyl-*n*-propyl disulfide and diethyl disulfide and the pair of corresponding trisulfides each of these pairs should be completely separable on Apiezon. This has been confirmed experimentally for several cases. In particular, the pairs dimethyl trisulfide and di-*n*-propyl disulfide, di-*i*-butyl disulfide and allyl-*n*-propyl disulfide, methyl-*n*-propyl trisulfide and di-*n*-butyl disulfide, and di-*i*-propyl disulfide and diethyl disulfide (or methyl-*n*-propyl disulfide) were incompletely resolved on Carbowax but were easily separated with Apiezon. This is merely one more example of the advantage of using two or more different stationary phases and in this case with a wide variation in polar character.

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[CONTRIBUTION FROM THE RESEARCH LABORATORIES, NATIONAL DRUG CO.]

Synthesis in the 5-Hydroxyindole Series. *N*-Acetyl-5-hydroxytryptophan and Related Compounds

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A number of pharmacologically interesting 5-hydroxyindole derivatives, including *N*-acetyl-5-hydroxytryptophan (X), 5-hydroxyindole-3-acetamide (XVI) and 5-hydroxytryptophol (XIX) were synthesized. An improved process for the large scale preparation of 5-hydroxytryptophan was reported.

The importance of the physiological properties of indole derivatives has been emphasized again by the isolation of the powerful vasoconstrictor principle, serotonin.[2] The confirmation of its structure as 5-hydroxytryptamine[3,4] prompted us to initiate a study of 5-hydroxyindole derivatives. While our work was in progress, a few communications[3-5] on this subject appeared in the literature. This paper deals with the syntheses of *N*-acetyl-5-hydroxytryptophan, 5-hydroxytryptophol and related substances of potential pharmacological importance, and reports on improved methods for the large scale preparation of the important compound, 5-hydroxytryptophan.

The synthesis of 5-hydroxytryptophan by condensation of 5-benzyloxygramine with diethyl formaminomalonate, followed by saponification, decarboxylation, and hydrogenolysis has been reported.[4] Since large quantities of 5-hydroxytryptophan and related compounds were required by us, the commercially available diethyl acetamidomalonate, rather than the formamido analog, was employed for the condensation with the benzyloxygramine[6] (I). The reaction proceeded successfully to give the indole malonicester II in 78% yield. Catalytic debenzylation of II provided the 5-hydroxy-compound III. A combined decarboxylation and deacetylation of the acetaminomalonic acid IV, which was obtained by mild saponification of the corresponding ester II,

(1) Present address: Research Division, Ethicon, Inc., Somerville, N. J.

(2) M. M. Rapport, A. A. Green, and I. H. Page, *Science*, **108**, 329 (1948); *J. Biol. Chem.*, **176**, 1243 (1948); M. M. Rapport, *J. Biol. Chem.*, **180**, 961 (1949).

(3) K. E. Hamlin and F. E. Fischer, *J. Am. Chem. Soc.*, **73**, 5007 (1951).

(4) M. E. Speeter, R. V. Heinzelmann, and D. I. Weisblat, *J. Am. Chem. Soc.*, **73**, 5514 (1951).

(5) A. Ek and B. Witkop, *J. Am. Chem. Soc.*, **75**, 500 (1953).

(6) During our early experiments this compound was prepared by modification of the method of H. Kühn and O. Stein [*Ber.*, **70**, 567 (1937)], and later by the procedure of Ek and Witkop[7] with improved yields. We are indebted to Dr. B. Witkop for making available to us their procedure far in advance of publication.

(7) A. Ek and B. Witkop, *J. Am. Chem. Soc.*, **76**, 5579 (1954).