

Separation of some selenides, sulfides and ethers by gas chromatography

Many organic compounds of selenium, like those of sulfur, are known to occur in trace amounts in living organisms¹⁻⁷. Since our attention has been focussed on these compounds, we have found that gas chromatography in view of their physico-chemical properties appears to be a suitable analytical tool for the reliable resolution of their mixtures. The method can be employed for the estimation of the compounds in living matter and in exhaled gases.

We have developed a separation procedure for these compounds and determined their behaviour in relation to analogous compounds of sulfur and oxygen.

Compounds of Se have so far received little attention in gas chromatography though EVANS AND SMITH⁸ mention the determination of dibutyl selenide and there is a somewhat more extensive study by EVANS AND JOHNSON⁹.

From the point of view of physiological research it was necessary first of all to obtain the purest possible preparation of dimethyl selenide, both non-radioactive and ¹⁴C-labelled. In this case, gas chromatography was used as a control method for checking the purity of the compound synthesized. From an analytical point of view it was important to examine the separation of some derivatives of hydrogen selenide, both from one another and from analogous derivatives of hydrogen sulfide. So that we could compare the chromatographic data of compounds of elements of the same group of the periodic system we have also examined analogous compounds of oxygen.

Experimental

Apparatus. The measurements were carried out in a Pye Argon Chromatograph (W. G. Pye and Co. Ltd., Cambridge, England) using a ⁹⁰Sr ionization detector. We used a 120 cm straight glass column of internal diameter equal to 4 mm. The liquid phases used were 20% (w/w) dinonyl phthalate (May and Baker Ltd., Dagenham, England) and 20% (w/w) squalane (B.D.H. Laboratory Chemicals Division, England). In both cases, 80-100 mesh Celite 545 (Johns-Manville, London, England) was used as carrier. The flow rate of argon was 50 ml/min. Measurements were carried out at 90° and at 110°. Dimethyl selenide was estimated at room temperature. For sample introduction we used a 10 μl syringe (Hamilton Co., Inc., Micromesure, The Hague, Holland). With radioactive dimethyl selenide we used a modified procedure wherein the gas flow was not interrupted. The sample was applied by piercing a disc made of silicone rubber which was located inside a tapered-cone stopper; the amounts applied were 0.2-0.6 μl of substance. The eluates from the column were condensed at -70°.

Substances examined. The substances investigated here can be divided into three groups: selenides, sulfides and ethers.

The first group contained: ¹⁴C-dimethyl selenide, dimethyl selenide, diethyl selenide, dipropyl selenide, di-*n*-butyl selenide, phenylselenol, phenyl methyl selenide and phenyl ethyl selenide. The second group contained: dimethyl sulfide, diethyl sulfide, dipropyl sulfide, di-*n*-butyl sulfide, thiophenol, phenyl methyl sulfide and phenyl ethyl sulfide. The ether group contained: dipropyl ether, di-*n*-butyl ether, phenol, anisole and phenetole.

Synthesis of ¹⁴C-dimethyl selenide (ref. 10). A mixture of 4.5 g Rongalite C and 1.25 g elementary selenium was placed in a 50 ml flask provided with a reflux condenser. While stirring with an electromagnetic stirrer and cooling with water, a

solution of 3.5 g NaOH in 15 ml water was added. After some 30 min we attached the apparatus (Fig. 1) through which 2 ml $^{14}\text{C}\text{H}_3\text{I}$ (specific activity 2.5 mC/ml) was distilled into the reaction mixture and the mixture was refluxed for 3 h. It was then distilled and the fraction boiling at 56–60° collected. This was redistilled in a collar flask and the 56–58° fraction collected. We obtained 1.4 g (81.4 % yield with respect to Se) of the product with a radiochemical yield of 4.64 mC (92%). The specific activity of the dimethyl selenide was 0.36 mC/mmol.

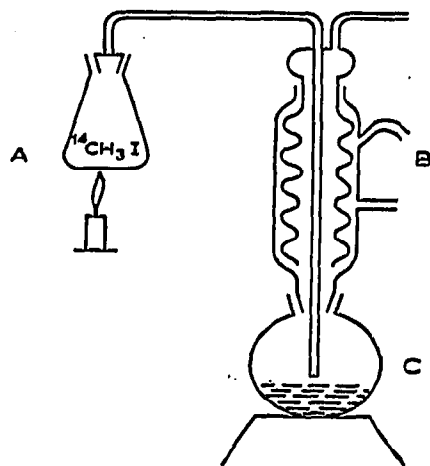


Fig. 1. Apparatus for producing ^{14}C -dimethyl selenide. A = Flask for distilling $^{14}\text{C}\text{H}_3\text{I}$ through the cooling system B into the reaction flask C, where dimethyl selenide is condensed.

Syntheses of all the other substances have been described in the papers quoted in Tables I, II and III.

Results and discussion

Characteristic chromatographic data were obtained for the series of selenides, sulfides and ethers under identical conditions in the two phases and at the two tem-

TABLE I

RELATIVE ELUTION VALUES REFERRING TO DIPROPYL SELENIDE

Selenides	B.p. (°C)	20% DNPHT		20% Squalane		Number of C atoms	Mol. wt.	Ref. to method of preparation
		90°	110°	90°	110°			
Dimethyl selenide	58	0.27	0.22	0.2	0.26	2	109.03	10
Diethyl selenide	108	0.28	0.25	0.23	0.26	4	137.08	10
Di-n-propyl selenide	159	1.00 ^a	1.00 ^b	1.00 ^c	1.00 ^d	6	165.13	10
Di-n-butyl selenide	81–83 (12)	4.5	3.84	4.42	3.74	8	193.18	10
Phenylselenol	183.6	1.3	1.3	0.87	0.89	6	157.07	11
Phenyl methyl selenide	79–80 (12)	6.2	5.32	3.53	3.19	7	171.09	12
Phenyl ethyl selenide	89–91 (12)	9.2	7.59	5.59	4.8	8	185.12	12

^a $V_0 = 600$ ml.

^b $V_0 = 303$ ml.

^c $V_0 = 545$ ml.

^d $V_0 = 292$ ml.

TABLE II

RELATIVE ELUTION VALUES REFERRING TO DIPROPYL SULFIDE

Sulfides	B.p. (°C)	20% DNPHT		20% Squalane		Number of C atoms	Mol. wt.	Ref. to method of preparation
		90°	110°	90°	110°			
Dimethyl sulfide	35	0.08	0.09	0.013	—	2	62.13	13
Diethyl sulfide	76-77	0.12	0.14	0.18	—	4	90.18	13
Di-n-propyl sulfide	140-146	1.00 ^a	1.00 ^b	1.00 ^c	—	6	118.23	13
Di-n-butyl sulfide	182	4.59	3.88	4.55	—	8	146.28	13
Thiophenol	168-169	3.2	2.88	1.7	—	6	110.17	14
Phenyl methyl sulfide	69 (12)	7.15	5.97	4.01	—	7	124.20	12
Phenyl ethyl sulfide	96-98 (12)	10.3	8.44	6.22	—	8	138.22	12

^a $V_0 = 382.5$ ml.^b $V_0 = 204$ ml.^c $V_0 = 320$ ml.

TABLE III

ELUTION VALUES REFERRING TO DIPROPYL ETHER

Ethers	B.p. (°C)	20% DNPHT		20% Squalane		Number of C atoms	Mol. wt.
		90°	110°	90°	110°		
Dimethyl ether	—	—	—	—	—	2	46.07
Diethyl ether	34	—	—	—	—	4	74.12
Di-n-propyl ether	91	1.00 ^a	1.00 ^b	1.00 ^c	1.00 ^d	6	102.17
Di-n-butyl ether	142.4	4.87	6.16	5.82	4.55	8	130.22
Phenol	181.4-2	16.2	20.1	9.42	—	6	94.11
Anisole	154.5- 155.5	12.1	15.0	7.00	5.44	7	108.13
Phenetole	172	19.8	23.7	12.5	9.0	8	122.16

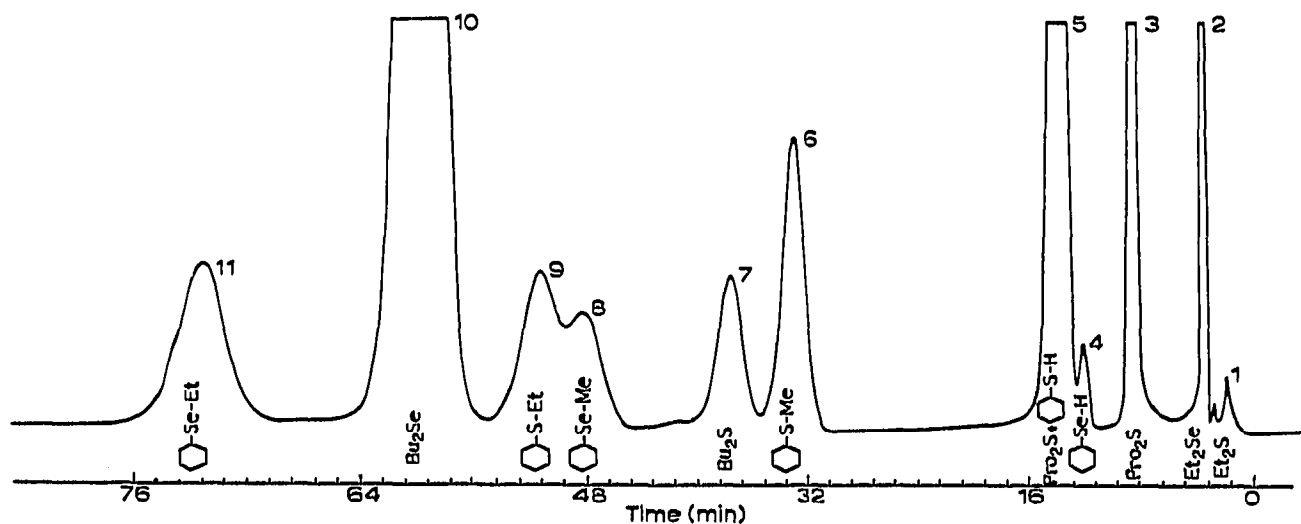
^a $V_0 = 64$ ml.^b $V_0 = 24$ ml.^c $V_0 = 48$ ml.^d $V_0 = 36$ ml.

Fig. 2. Separation of the mixture of selenides and sulfides. Stationary phase, 20% squalane; column temperature, 90°; flow rate of argon, 60 ml/min; argon ionization detector.

peratures and the results are summarised in Tables I, II and III. We found 20% squalane suitable at 90° and 20% dinonyl phthalate at both 90° and 110°. At temperatures above 90°, separation on squalane is less favorable. Under these conditions, the individual representatives of the various groups are readily separated. The only exception is the separation of dimethyl selenide (sulfide) from diethyl selenide (sulfide) in view of the relatively high column temperature. For the same reason in the ether series, separation only above di-*n*-propyl ether was observed.

The effect of the character of the liquid phase on the separation of the individual compounds may be seen in the various elution times. Very close elution data are displayed by the pairs $(C_3H_7)_2X$, C_6H_5XH and $(C_4H_9)_2X$, $C_6H_5XCH_3$ (where X =

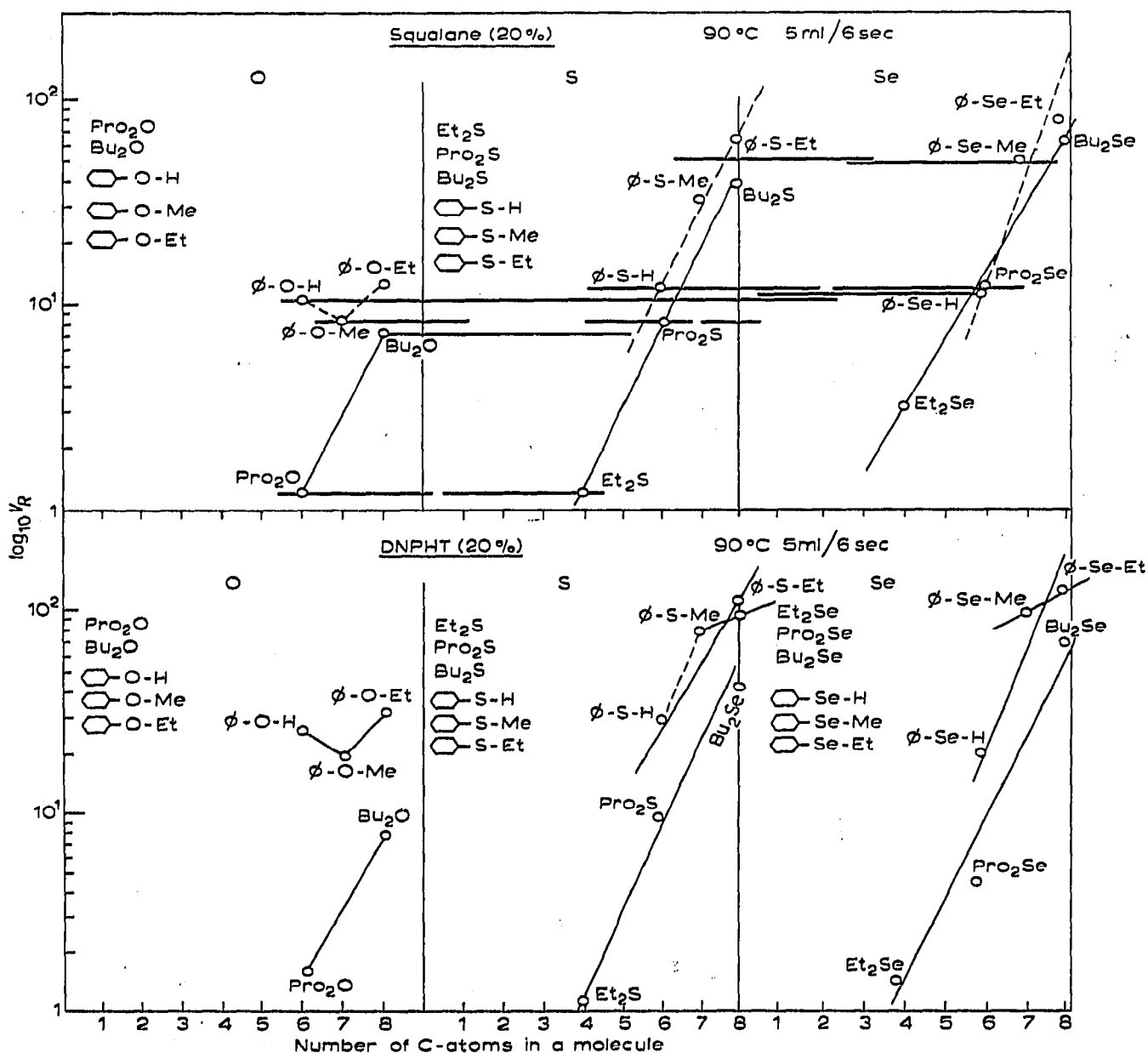


Fig. 3. The relationship between $\log_{10} V_R$ and number of carbon atoms in a molecule. Temperature, 90°; flow rate, 5 ml/6 sec. Stationary phase: (a) 20% squalane, (b) 20% dinonyl phthalate.

Se,S). The similarity of elution times is affected by the boiling points and by the molecular weights.

An example of different separation of substances on two different phases is provided by di-*n*-butyl sulfide which, on squalane at 90°, has a higher elution time than phenyl methyl sulfide, whereas on dinonyl phthalate under the same conditions the reverse is true. Similarly, phenylselenol has a shorter elution time than dipropyl selenide, and dibutyl selenide greater than phenyl methyl selenide, whereas on dinonyl phthalate it is reversed.

Optimum separation of a mixture of sulfides and selenides is achieved at 90° on 20 % squalane at a flow rate of 60 ml/min. Thiophenol is not separated from dipropyl selenide (Fig. 2).

Of the various graphical relationships, we plotted the relationship between $\log V_R$ and the number of carbon atoms in the individual derivatives on both liquid phases. Fig. 3 shows its linear course in dialkyl and alkyl aryl derivatives.

When the elution data for the various ethers, sulfides and selenides are compared it may be seen that the elution values rise from oxygen to selenium, *i.e.* with the increasing atomic weight of the central element.

Results have been summarized in tables and graphs.

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