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GAS–LIQUID CHROMATOGRAPHY OF PHENOTHIAZINE DERIVATIVES AND RELATED COMPOUNDS

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SUMMARY

Analytically packed columns prepared with SE-30, OV-1, OV-17, Lexan, STAP, QF-1, XE-60, FFAP, Versamid 900 and Carbowax 20M as liquid phases were compared for the gas–liquid chromatographic separation of phenothiazines and chemically related drugs. On the basis of better separation efficiency, higher plate number and shorter analysis time, two systems with different polarities were preferred for qualitative analysis: a 5% OV-1 and a 2% FFAP column coated on Chromosorb W (acid-washed and silane-treated, 100–120 mesh), Aeropak-30 (100–120 mesh) or Diatoport S (80–100 mesh). By using suitable internal standards, calibration factors, $\bar{k}$, of some representative compounds were determined and the linearities of response with respect to mass or concentration ratios were checked.

INTRODUCTION

Several workers have described gas–liquid chromatographic (GLC) separations of some phenothiazine drugs in their pure form[1–6] or in relatively simple mixtures[7–9]. The GLC determination of some chlorpromazine metabolites[10–12] and their thermal decomposition products[13] has been described. FONTAN *et al.*[14] examined the gas chromatographic properties of the pyrolysis products of phenothiazines. The analytical toxicological applications of the GLC of psychoactive phenothiazine drugs have been considered[15,16]. Chlorpromazine metabolites were determined in human urine[17] and plasma[18–21], and the metabolism of dibenzepine[22] and imipramine[23] was partly elucidated by GLC.

The present study involved a search for better liquid phases that would permit the separation of a wide range of structurally related compounds. By using two analytically packed columns with different polarities, we attempted to improve both the qualitative and quantitative analyses of phenothiazines and related compounds.

EXPERIMENTAL

Apparatus

An Aerograph Hy-Fi Model 600-D chromatograph equipped with a flame ionization detector, combined with an Aerograph Hy-Fi Model 650 hydrogen generator and a Sargent Model-SR recorder, 0–1 mV range, was used.

A Hewlett-Packard Research 5750 gas chromatograph equipped with dual flame ionization detectors and operated with an Elhygen hydrogen generator and a Hewlett-Packard 7128A recorder, 0–1 mV range, was also used.

Reference solutions

Free bases. A 100.0-mg sample of the salt form was dissolved in 20 ml of 0.005 *N* HCl; after saturation with nitrogen gas 1 ml of 10 *N* NaOH was added and the solution was extracted with two 50-ml volumes of peroxide-free diethyl ether (freshly distilled over reagent grade hydroquinone); the combined ethereal layers were dried over Na_2SO_4, filtered, evaporated under a stream of nitrogen to *ca.* 1 ml, and finally diluted with ethanol to 10.0 ml. The solutions were kept in a deep freezer at −20°. The concentration of the active substance was calculated by multiplying by the correction factor, f_c = molecular weight of free base/molecular weight of salt form.

Sulphoxides. A 100.0-mg amount of free base or salt was transferred to a 10-ml graduated cylinder, dissolved in 1 ml of 15% H_2O_2 and 0.2 ml of acetic acid was added; after a reaction time of 30 min in a water bath at 60°, the reaction mixture was diluted to 10.0 ml with water.

Acetylated derivatives. To a 10.0-mg sample of each substance in a conical siliconized tube of 15 ml capacity, 0.2 ml of pyridine (refluxed and distilled over KOH) and 0.2 ml of acetic anhydride (refluxed and distilled over calcium carbide) were added; after a reaction time of 1 h in a P_2O_5 desiccator, the reaction mixture was evaporated under a slow stream of nitrogen and the residue obtained was dissolved in 10.0 ml of ethyl acetate.

Mixtures. From the "free base" solutions, four mixtures, M3, M4, M5 and M6, were prepared: M3, 20 µl of 0.89% isothipendyl + 20 µl of 0.84% prothipendyl + 20 µl of 0.88% imipramine; M4, 20 µl of 0.88% desmethylimipramine + 20 µl of 0.88% imipramine + 20 µl of 0.88% amitriptyline + 20 µl of 0.88% nortriptyline; M5, 20 µl of 0.89% promethazine + 20 µl of 0.41% alimemazine + 20 µl of 0.91% triflupromazine + 20 µl of 0.89% isothipendyl + 20 µl of 0.88% imipramine; M6, 20 µl of 0.89% diethazine + 20 µl of 0.89% profenamine + 20 µl of 0.85% aminopromazine + 20 µl of 0.89% promazine + 20 µl of 0.88% imipramine + 20 µl of 0.72% trimeprimine.

Calibration solutions. Fixed volumes of the "free base" solutions of the compound to be determined and of internal standard were transferred with a 50-µl Hamilton 705N syringe into capillary tubes and thoroughly mixed; for each calibration, four different solutions of equal mass, $\Delta(m_x/m_s)$, or concentration, $\Delta(c_x/c_s)$, ratio increments were prepared.

Preparation of column packings

In a 250-ml Rotavapor flask, x g of liquid phase were dissolved in an excess of suitable (according to manufacturers' recommendations) solvent and were mixed with

TABLE I

RETENTION TIMES, t_R (min) OF PHENOTHIAZINES AND RELATED DRUGS DETERMINED ON A 2% SE-30 ON CHROMOSORB W (ACID-WASHED AND SILANE-TREATED), 100–120 MESH, COLUMN (1.50 m LENGTH AND 3 mm I.D.) OPERATED UNDER ISOTHERMAL CONDITIONS AT 20° INTERVALS

Substance	*Mol. wt.*	*Oven temperature (°C)*					
		180	*200*	*220*	*240*	*260*	*280*
I.1 Aminoalkyl phenothiazines							
(a) Dialkylaminoethyl derivatives							
Diethazine	298.46	11.8	6.0	1.5			
Dimethoxanate	358.47			—	—	—	—
Dimetiotazine	391.56			12.3	5.2	2.2	1.0
Profenamine	312.48	19.1	5.7	1.4			
Promethazine	284.43		4.1	1.0			
Propiomazine	340.49			4.4	1.9		
Thiazinamium	299.46			—	—	—	—
(b) Dialkylaminopropyl derivatives							
Acepromazine	326.47			4.4	1.9		
Alimemazine	298.46		4.5	1.3			
Aminopromazine	327.50		8.2	1.8			
Chlorproethazine	346.93			3.1	1.5		
Chlorpromazine	318.88			2.0			
Levomepromazine	328.48			2.3			
Methiomeprazine	344.55			3.8	1.8	0.8	
Oxomemazine	330.46			4.3	2.0		
Promazine	284.43		5.0	1.4			
Propiopromazine	340.49			5.3	2.4		
Triflupromazine	352.43	11.9	3.7	1.0			
I.2 Piperidylalkyl phenothiazines							
Pecazine	310.47			2.3			
Propericiazine	365.50				12.2	4.0	1.6
Thioridazine	370.59			12.2	5.5	2.5	1.0
I.3 Piperazinylalkyl phenothiazines							
Acetophenazine	411.57			—	—	—	—
Dixyrazine	427.62			—	—	—	—
Fluphenazine	437.54			—	—	—	—
Perazine	339.51			4.7	2.3		
Perphenazine	403.99			—	—	—	—
Prochlorperazine	373.96			7.7	3.8	1.6	
Thiopropazate	446.03			—			2.8
Thioproperazine	446.65			—			3.7
Trifluoperazine	407.51			3.6	1.7		
II.1 Azaphenothiazines							
Isothipendyl	285.42		4.3	1.1			
Prothipendyl	285.42			1.3			
II.2 Thioxanthenes							
Chlorprothixene	315.88			2.0			
Methixene	309.48			1.9			

(Continued on p. 342)

TABLE I *(continued)*

Substance	*Mol. wt.*	*Oven temperature (°C)*					
		180	*200*	*220*	*240*	*260*	*280*
III.1 Dibenzazepines							
(a) Iminodibenzyl derivatives							
Desmethylimipramine	266.39		3.7	1.0			
Imipramine	280.42		3.1	0.9			
Trimeprimine	294.44	10.0	3.0	0.9			
(b) Iminostilbene derivatives							
Opipramol	363.51			—	—	—	—
III.2 Dibenzodiazepines							
Dibenzepine	295.39			2.0	1.0		
III.3 Dibenzocycloheptadienes							
Amitriptyline	277.41	9.6	2.7	0.8			
Nortriptyline	263.38		3.2	0.9			

a g of Chromosorb W (acid-washed and silane-treated, 100–120 mesh), Aeropak-30 (Aerograph, 100–120 mesh) or Diatoport S (Hewlett-Packard, 80–100 mesh) support material. The flask was connected to a rotatory evaporator and the solvent removed under reduced pressure. Rotation was stopped when particles moved freely without sticking. The complete packing was dried for 1 h in a porcelain dish in a drying oven at 105°. After pouring it into a 2.5 × 30 cm glass column, the finest particles were removed by blowing them off with a stream of nitrogen.

The mixed packing involved the application of KOH in methanol and subsequent coating with a polar liquid phase.

The percentage of liquid phase, P, was calculated from the equation $P = 100x/(a+x)$.

GLC operating conditions

Spiral silanized glass columns of 3 or 4 mm I.D. and 1.50 or 1.80 m length were filled with column packings by applying a vacuum unidirectionally from the detector site. After connecting the column with the injector port, conditioning was carried out as follows: first for 1 h at 120° with a 10 ml/min nitrogen stream, then for 12 h at 10° below the maximum permissible temperature of the liquid phase with no nitrogen flow, and finally for 3 h under the appropriate operating conditions.

All of the analyses were carried out under isothermal conditions in the temperature range 180–280° with temperatures that were 20° higher for the injector and detector blocks. Nitrogen was used as the carrier gas at a flow-rate (bubble flow meter measurement) of 25–30 and 60–80 ml/min for 3 and 4 mm I.D. columns, respectively (inlet pressure *ca.* 4 kg/cm^2). The air and hydrogen flow-rates were adjusted so as to give good stability and optimum sensitivity: air, 360–370 or 750 ml/min; hydrogen: 23–24 or 70 ml/min. The electrometer settings used were: range × 10 and attenuation × 4 or × 2 for the Aerograph 600-D instrument; and range × 10 and attenuation × 16 or × 8, range × 1 and attenuation × 32 for the Hewlett-Packard Model 5750 instru-

TABLE II

RELATIVE RETENTIONS, r_{21}, OF PHENOTHIAZINE SULPHOXIDES WITH RESPECT TO FREE BASES MEASURED ON A 2% SE-30 ON CHROMOSORB W (ACID WASHED AND SILANE TREATED), 100–120 MESH, COLUMN (1.50 m LENGTH AND 3 mm I.D.) OPERATED UNDER ISOTHERMAL CONDITIONS AT 20° INTERVALS

Substance	*Temperature (°C)*		
	240	*260*	*280*
I.1 Aminoalkyl phenothiazines			
(a) Dialkylaminoethyl derivatives			
Diethazine sulphoxide	4.0		
Dimetiotazine sulphoxide		2.9	
Profenamine sulphoxide	3.4		
Promethazine sulphoxide	3.3		
Propiomazine sulphoxide	3.0	2.5	
(b) Dialkylaminopropyl derivatives			
Acepromazine sulphoxide		2.5	
Alimemazine sulphoxide	3.1		
Aminopromazine sulphoxide	2.6		
Chlorproethazine sulphoxide	3.3	2.7	
Chlorpromazine sulphoxide	3.1		
Levomepromazine sulphoxide	3.8		
Methiomeprazine sulphoxide		5.8	4.8
Promazine sulphoxide	3.7		
Propiopromazine sulphoxide		2.7	
Triflupromazine sulphoxide	3.1		
I.2 Piperidylalkyl phenothiazines			
Pecazine sulphoxide	3.2		
Propericiazine sulphoxide		—	—
Thioridazine sulphoxide		—	—
I.3 Piperazinylalkyl phenothiazines			
Perazine sulphoxide		3.6	2.7
Prochlorperazine sulphoxide		2.8	2.8
Trifluoperazine sulphoxide		2.3	
II.1 Azaphenothiazines			
Isothipendyl sulphoxide	2.7		
Prothipendyl sulphoxide	3.1		
II.2 Thioxanthenes			
Methixene sulphoxide	3.0		

ment. The recorder speeds were 0.3 in./min for the Sargent and 0.25 in./min for the Hewlett-Packard Model 7128A.

Sampling

Volumes varying from 0.5 to 2.0 μl of solutions were injected on to the top of the columns with 1-μl (7001N) or 10-μl (701N) Hamilton syringes.

Evaluation of chromatograms

Retention times, t_R, were evaluated by measuring the time interval between the solvent front and the peak maximum. Relative retentions, r_{21}, were calculated from the equation $r_{21} = t_R$ of the compound examined divided by t_R of the reference

TABLE III

RELATIVE RETENTIONS, r_{21}, AND PLATE NUMBERS, N, OF SOME REPRESENTATIVE PHENOTHIAZINES AND RELATED DRUGS WITH RESPECT TO IMIPRAMINE, DETERMINED ON NINE DIFFERENT COLUMNS (1.80 m LENGTH AND 3 mm I.D.)

All liquid phases packed on Aeropak-30, 100–120 mesh.

Substance	*5% OV-1*		*5% OV-17*		*5% Lexan*		*5% STAP*	
	r_{21}	N	r_{21}	N	r_{21}	N	r_{21}	N
I.1 Aminoalkyl phenothiazines								
(a) Dialkylaminoethyl derivatives								
Diethazine	1.62	2,070	1.70	2,773			1.99	3,077
Profenamine	1.60	2,948	1.59	2,909			1.55	3,270
Promethazine	1.17	3,410	1.32	3,280	1.61	242	1.55	3,156
(b) Dialkylaminopropyl derivatives								
Alimemazine	1.27	3,003	1.33	3,553			1.45	3,159
Aminopromazine	1.96	2,949	2.09	3,037			2.20	3,278
Levomepromazine					—	—		
Promazine	1.37	3,443	1.60	2,934	2.07	365	2.02	3,331
Triflupromazine	1.03	3,387	0.86	3,034			0.93	3,015
I.2 Piperidylalkyl phenothiazines								
Propericiazine							—	—
I.3 Piperazinylalkyl phenothiazines								
Dixyrazine							—	—
Thioproperazine							—	—
Trifluoperazine							4.63	2,873
II.1 Azaphenothiazines								
Isothipendyl	1.18	3,441	1.36	3,301			1.57	3,248
Prothipendyl	1.45	3,494	1.71	3,078			2.10	3,191
III.1 Dibenzazepines								
(a) Iminodibenzyl derivatives								
Desmethylimipramine	1.07	5,717	—	—	—	—	—	—
Imipramine	1.00	3,700	1.00	3,006	1.00	187	1.00	3,085
Trimeprimine	1.00	3,202	0.92	2,855			0.82	2,723
III.2 Dibenzodiazepines								
Dibenzepine								
III.3 Dibenzocycloheptadienes								
Amitriptyline	0.94	4,431	0.91	3,710	0.81	160	0.82	3,142
Nortriptyline	0.99	4,182	1.08	3,653	—	—	—	—
Retention time, t_R (min), of imipramine	4.8		11.4		12.6		12.7	

5% QF-1		5% XE-60		5% KOH+2.5% FFAP		5% KOH+2.5% Versamid 900		5% KOH+2.5% Carbowax 20M	
r_{21}	N	r_{21}	N	r_{21}	N	r_{21}	N	r_{21}	N
		1.93	1,832	1.97	2,711	2.01	1,509		
		1.65	1,520	1.58	2,773	1.66	1,158		
1.34	3,091	1.46	2,180	1.48	2,725	1.44	1,880	1.54	3,766
1.37	2,527	1.39	2,315	1.40	2,791	1.38	1,925	1.44	3,661
		2.11	1,986	2.17	2,780	2.12	1,176		
2.86	3,611			3.65	2,718	3.21	1,881	3.91	3,835
1.66	3,578	1.81	2,277	1.92	2,741	1.81	1,407		
1.32	2,693	1.13	1,859	0.97	3,006	0.90	1,678	0.97	3,463
				—	—	—	—		
				—	—	—	—		
				—	—	—	—		
4.51	2,877			5.02	2,310	4.52	1,851	4.80	3,593
1.15	2,771	1.25	2,230	1.46	2,204	1.38	1,834	1.51	3,525
1.51	3,419	1.62	2,347	1.99	2,483	1.87	1,951	1.99	3,623
1.23	2,676	1.40	2,862	1.45	2,804	1.37	1,843	1.51	3,705
1.00	2,094	1.00	2,212	1.00	2,875	1.00	1,603	1.00	3,310
		0.85	2,043	0.82	2,791	0.84	827		
4.47	2,964			4.77	2,954	4.65	2,389	4.92	3,978
0.84	1,569	0.85	1,598	0.84	2,851	0.87	1,882	0.85	3,441
1.11	1,907	1.15	2,750	1.19	2,780	1.12	1,807	1.21	3,456
3.0		2.3		5.1		10.3		13.4	

substance. The column efficiency was estimated in terms of the number of theoretical plates, $N = 5.53\,(d/w_{0.5h})^2$, where d is the distance in millimetres on the chromatogram from the point of injection to the peak maximum and $w_{0.5h}$ is the peak width at half-height.

Mean calibration factors, $\bar{k}$, were determined by chromatographing each calibration solution of a given series three times and using the following equations:

$$\bar{k} = \sum_{t=1}^{t=3} k_t/3$$

$$k_t = A_x\, w_s / A_s\, w_x$$

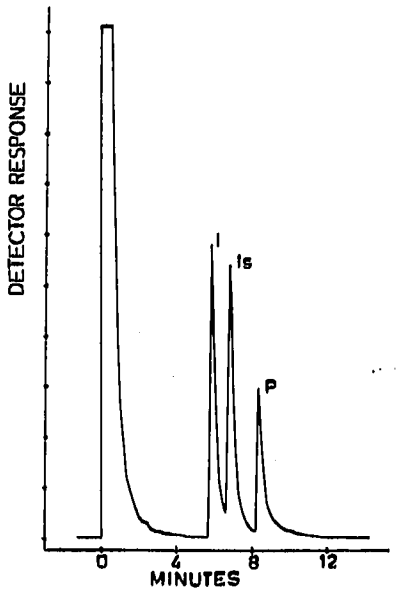
where A_x, A_s and w_x, w_s are the peak areas and weights of the compound to be calibrated and the internal standard, respectively. Peak areas were measured by the method of peak height multiplied by peak width at half-height*, $A = h_{max}\, w_{0.5h}$.

RESULTS AND DISCUSSION

Preliminary results on the chromatographic behaviour of phenothiazines and related drugs were obtained on a 2% SE-30 on Chromosorb W (acid-washed and silane-treated), 100–120 mesh, column (1.50 m length and 3 mm I.D.) and are summarized in Table I. In these experiments, a temperature range of 180–280° at 20° intervals was covered. Most compounds chromatograph satisfactorily at 220° and show retention times corresponding to their boiling points or molecular weights. Exceptions are substances with branched aliphatic side-chains (profenamine, alimemazine, isothipendyl and trimeprimine) or a CF_3 substituent on the tricyclic system (trifluopromazine and trifluoperazine), which show decreased retention times. Most compounds that contain a free primary alcohol group (acetophenazine, dixyrazine, fluphenazine and perphenazine) cannot be eluted even at 280°, but chromatograph easily as the acetates. Thiazinamium, with a quaternary ammonium structure, and dimethoxanate, which possesses a combined ester–ether function, are irreversibly adsorbed because of their high polarities. Phenothiazine sulphoxides, for which relative retentions, r_{21}, are given in Table II, are readily separated at 240° and show chromatographic characteristics similar to those of corresponding free phenothiazines.

It is known from previous work that it is not possible to separate complex mixtures by using only one apolar SE-30 column. For this reason, we performed a systematic search for two-column systems with different polarities but which complement each other in terms of their separation capabilities. In order of increasing polarity, the following columns (1.80 m length and 3 mm I.D.) were tested: 5% OV-1 (methylsilicone polymer), 5% OV-17 (methylphenylsilicone polymer), 5% Lexan (polycarbonate resin), 5% STAP ("steroid analysis phase", Aerograph), 5% QF-1 (fluoroalkylsilicone polymer), 5% XE-60 (nitrilesilicone gum), 5% KOH + 2.5% FFAP (reaction product of Carbowax 20M and *m*-nitroterephthalic acid), 5% KOH + 2.5% Versamid 900 (dimer of linoleic acid copolymerised with ethylenediamine), 5% KOH + 2.5% Carbowax 20M (polyethylene glycol polymer), all coated on Aeropak-

* Measured by use of a 7× magnifying lens, Bausch and Lomb, 81-34-38.



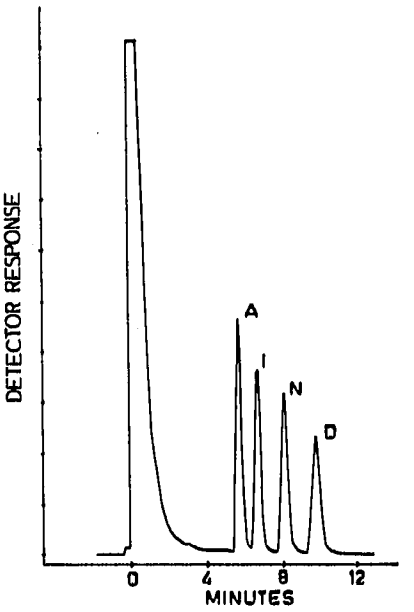


Fig. 1. Gas–liquid chromatogram of synthetic mixture M3 determined on a 5% OV-1 on Aeropak-30, 100–120 mesh, column (1.80 m length and 3 mm I.D.). I = imipramine; Is = isothipendyl; P = prothipendyl.

Fig. 2. Gas–liquid chromatogram of synthetic mixture M4 determined on a 5% KOH + 2.5% FFAP on Aeropak-30, 100–120 mesh, column (1.80 m length and 3 mm I.D.). A = amitriptyline; I = imipramine; N = nortriptyline; D = desmethylimipramine.

30, 100–120 mesh. The relative retentions, r_{21}, with imipramine as the reference substance, and plate numbers, N, obtained for representative compounds are given in Table III. Furthermore, in order to obtain an insight into separation efficiency we examined under identical conditions on several columns the synthetic mixtures M3, M4, M5 and M6. The results are shown in Figs. 1 and 2 and in Table IV. OV-1 appears to be the best apolar liquid phase, whereas FFAP and Carbowax 20M seem to be

TABLE IV

NUMBER OF DISCERNABLE PEAKS OBTAINED WITH SYNTHETIC MIXTURES M3, M4, M5 AND M6 ON EIGHT DIFFERENT COLUMNS (1.80 m LENGTH AND 3 mm I.D.)

All liquid phases coated on Aeropak-30, 100–120 mesh.

Liquid phase	*M3*	*M4*	*M5*	*M6*
5% OV-1	3	2	3	4
5% OV-17	3	3	3	5
5% STAP	3	2	3	5
5% QF-1	3	2	3	4
5% XE-60	3	4	4	5
5% KOH + 2.5% FFAP	3	4	2	5
5% KOH + 2.5% Versamid 900	3	4	3	5
5% KOH + 2.5% Carbowax 20M	3	4	3	

TABLE V

MEAN CALIBRATION FACTORS, $\bar{k}$, AND LINEARITY CHECKS OF SOME REPRESENTATIVE PHENOTHIAZINES AND RELATED COMPOUNDS USING THE METHOD OF INTERNAL STANDARDIZATION

Substance	*Internal standard*	*Column system*	*Apparatus*				*Mean calibration factor, $\bar{k}$*	*Linearity over mass ratio range, A (m_x/m_s)*
			Type[a]	*Oven temperature (°C)*	*Electrometer*			
					Range	*Attenuation*		
1.1 Aminoalkyl phenothiazines								
(a) Dialkylaminopropyl derivatives								
Chlorpromazine	Promazine	2% SE-30[b]	A.	220	× 10	× 2	0.80	0.00/1 to 3.03/1
Levomepromazine	Trifluoperazine	5% OV-1[c]	H.P.	230	× 10	× 16	0.97	0.25/1 to 1.00/1
	Perazine	2% FFAP[c]	H.P.	240	× 10	× 8	1.65	0.25/1 to 0.43/1
	Trifluoperazine	2% FFAP	H.P.	240	× 10	× 8	1.17	0.25/1 to 1.00/1
	Trifluoperazine	2% FFAP	H.P.	230	× 10	× 8	1.04	0.25/1 to 1.00/1
Levomepromazine sulphoxide	Dimetiotazine	5% OV-1	H.P.	240	× 10	× 8	1.32	1.00/1 to 4.00/1
	Trifluoperazine	5% OV-1	H.P.	230	× 10	× 16	0.80	1.50/1 to 4.00/1
Desmethyllevomepromazine acetamide	Trifluoperazine	5% OV-1	H.P.	230	× 10	× 16	0.46	0.43/1 to 1.00/1
Promazine	Levomepromazine	2% SE-30	A.	220	× 10	× 2	1.28	0.00/1 to 4.80/1
	Levomepromazine	2% SE-30	A.	220	× 10	× 4	1.40	0.00/1 to 4.80/1
1.2 Piperidylalkyl phenothiazines								
Propericiazine acetyl ester	Dixyrazine acetyl ester	5% OV-1	H.P.	260	× 1	× 32	1.94	0.25/1 to 1.00/1
1.3 Piperazinylalkyl phenothiazines								
Dixyrazine acetyl ester	Thioproperazine	5% OV-1	H.P.	260	× 1	× 32	1.06	0.25/1 to 0.67/1
Perazine	Trifluoperazine	5% OV-1	H.P.	240	× 10	× 8	0.69	1.00/1 to 4.00/1
		5% OV-1	H.P.	240	× 10	× 8	0.67	1.00/1 to 4.00/1
		2% FFAP	H.P.	240	× 1	× 32	0.55	1.00/1 to 4.00/1
		2% FFAP	H.P.	240	× 10	× 8	0.53	1.00/1 to 4.00/1

Thioproperazine	Propericiazine acetyl ester	5% OV-1	H.P.	260	×1	×32	0.46	0.43/1 to 2.33/1
Trifluoperazine	Perazine	5% OV-1	H.P.	230	×10	×8	1.28	0.11/1 to 0.67/1
		2% FFAP	H.P.	230	×1	×32	1.60	0.11/1 to 0.67/1
Trifluoperazine sulphoxide	Perazine	5% OV-1	H.P.	230	×10	×8	0.91	0.11/1 to 0.67/1
II.1 Azaphenothiazines								
Prothipendyl	Levomepromazine	5% OV-1	H.P.	230	×10	×8	0.86	0.43/1 to 1.50/1
		2% FFAP	H.P.	230	×1	×32	0.75	0.43/1 to 1.50/1
III.1 Dibenzazepines								
(a) Iminodibenzyl derivatives								
Desmethylimipramine	Profenamine	2% SE-30	A.	210	×10	×2	0.57	0.73/1 to 1.47/1
Desmethylimipramineacetamide	Perazine	2% FFAP	H.P.	240	×10	×8	3.28	0.43/1 to 1.00/1
Imipramine	Profenamine	2% SE-30	A.	210	×10	×2	0.93	0.00/1 to 1.00/1
	Promazine	5% OV-1	H.P.	185	×10	×8	1.32	0.25/1 to 1.00/1
		5% OV-1	H.P.	210	×10	×8	1.33	0.25/1 to 1.00/1
		2% FFAP	H.P.	185	×10	×8	1.42	0.25/1 to 1.00/1
		2% FFAP	H.P.	210	×10	×8	1.45	0.25/1 to 1.00/1
III.2 Dibenzodiazepines								
Dibenzepine	Prochlorperazine	1% FFAP[c]	H.P.	245	×10	×16	2.04	0.40/1 to 0.50/1
III.3 Dibenzocycloheptadienes								
Nortriptylineacetamide	Levomepromazine	5% OV-1	H.P.	240	×10	×8	1.43	1.00/1 to 4.00/1
		2% FFAP	H.P.	240	×10	×8	1.59	1.00/1 to 4.00/1

[a] A. = Aerograph Model 600-D; H.P. = Hewlett-Packard 5750.
[b] Coated on Chromosorb W (acid-washed and silane-treated), 100–120 mesh, column (1.50 m length and 3 mm I.D.).
[c] Coated on Diatoport S, 80–100 mesh, column (1.80 m length and 4 mm I.D.).

equivalent as polar systems. The latter show a comparable separation efficiency but FFAP requires a shorter analysis time and was therefore preferred for subsequent assays.

Calibration factors, $\bar{k}$, were determined on a 2% SE-30 on Aeropak-30, 100–120 mesh, column (1.80 m length and 3 mm I.D.) and a 5% OV-1 or 2% FFAP on Diatoport S, 80–100 mesh, column (1.80 m length and 4 mm I.D.). The linearities over the mass, $\Delta(m_x/m_s)$, or concentration, $\Delta(c_x/c_s)$, ratio ranges obtained were checked by graphical analysis. Complete results are presented in Table V. Even with structurally related compounds, some of the calibration factors differ markedly from 1.00, undoubtedly due to their different ionization efficiencies. Pronounced differences occur especially for acetamide derivatives, *e.g.*, desmethyllevomepromazineacetamide (0.46), desmethyllimipramineacetamide (3.28) and nortriptylineacetamide (1.43 and 1.59).

Detection limits are estimated at a total amount of active substance of 0.5–3.0 μg with the Aerograph Model 600-D instrument equipped with a 2% SE-30 on Aeropak-30, 100–120 mesh, column (1.50 m length and 3 mm I.D.), operated at an electrometer range of $\times$ 10 and attenuation $\times$2 for injections of 0.1–0.5 μl of 1% solutions, and at 0.05–1.5 μg with the Hewlett-Packard 5750 instrument equipped with a 5% OV-1 or 2% FFAP on Diatoport S, 80–100 mesh, column (1.80 m length and 4 mm I.D.), operated at an electrometer range of $\times$1 and attenuation $\times$16 for injections of 0.1–3.0 μl of 0.05% solutions.

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