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Rapid and complete chiral chromatographic separation of racemic quaternary tropane alkaloids

Complementary use of a cellulose-based chiral stationary phase in reversed-phase and normal-phase modes -a mechanistic study

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ABSTRACT

Complete chromatographic separations of a wide range of different tropane alkaloid stereoisomers on a commercially available cellulose-based chiral stationary phase (CSP) are described. The separations were achieved by using cellulose tris(3,5-dimethylphenyl)carbamate as CSP with both an aqueous and an organic mobile phase with different ionic modifiers. The effects of totally different modifiers in both mobile phases on the chromatographic resolution are described.

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INTRODUCTION

Tropane alkaloids are anticholinergic drugs. Well known are the esters of tropic acid with scopine (scopolamine) and tropine (atropine). The racemate atropine (3-tropoyloxytropane) is a mixture of the (*S*)- and (R)-hyoscyamine enantiomers, whereas the mixture of (*R*)- and (Q-scopolamine enantiomers is called atroscine (3-tropoyloxy-6β,7β-epoxytropane). The chromatographic enantioseparation of the tertiary compounds atropine and atroscine have been described previously [1,2]. By quaternization of these. tropane alkaloids the central effects of these drugs are avoided. These compounds are in use as neurotropic spasmolytics in modern therapy. Until now no methods existed (except for the imprecise measurement of optical rotation) for the determination of the enantiomeric purity of these compounds [3]. We describe here conditions for the determination of the optical purity of these drugs by chromatographic methods.

STEREOCHEMISTRY

Anticholinergics of the tropane alkaloids are esters of the N-alkylated nortropine (1) and norscopine (2) and racemic tropic acid (3). The quaternary N,N-dialkylated noratropine (4) and N,N-dialkylated noratroscine (5) molecules have one chiral centre in the 2′-position and two pseudoasymmetric centres in the C-3 and N-8 positions (see Fig. 1).

The direct chromatographic separation of the racemates refers to the chiral centre in the 2′-position in all instances. Therefore, tropine is characterized as 1(*R*),3(*r*),5(*S*),8(*r*)-azabicyclo[3.2.1.]octan-3-ol. For quaternary N,N-dialkylated nortropine the pseudoasymmetric nitrogen in the 8-position is described as "*r*" for $R_1 > R_2$ and "*s*" for $R_1 <$ R,.

The introduction of the epoxy group into the piperidine system leads to atroscine with a completely converted configuration of the saturated cyclic system. Therefore, the absolute configuration of scopine is described as 1(*S*),3(*s*), 5(*R*),6(*R*),7(*S*),8(*s*)-aza-6,7-epoxybicyclo[3.2.1.]-octan-3-01.

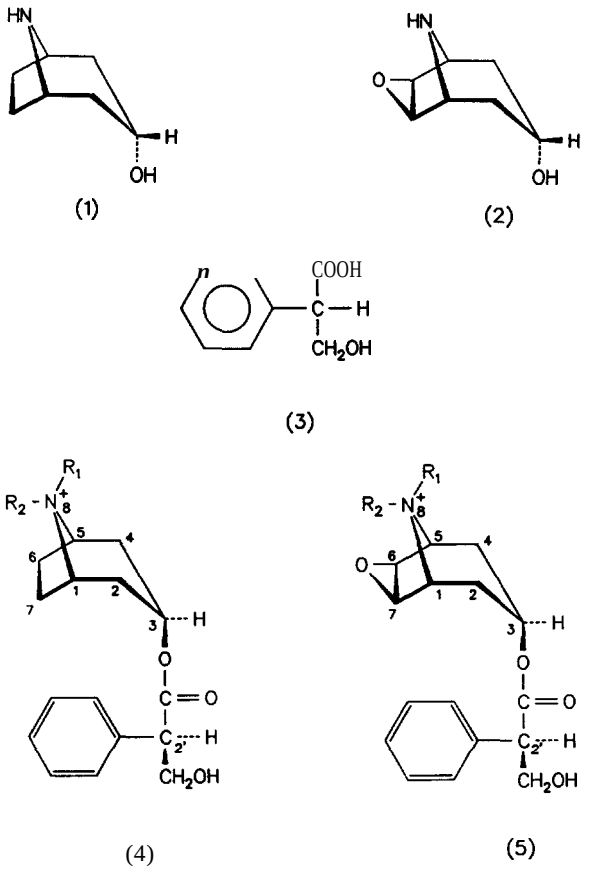

Fig. 1. Structures of nortropine **(1),** norscopine **(2),** tropic acid **(3),** quateinary N,N-dialkylated noratropine (4) and quaternary N,N-dialkylated noratroscine (5).

Similarly to N,N-dialkylnortropine, the quaternary N,N-dialkylnorscopine with different alkyl chains fixes the pseudoasymmetric centre in the 8-position, characterized as "*s*" for $R_1 > R_2$ and "r" for $R_1 < R_2$ [4–7].

EXPERIMENTAL

The quaternary tropane alkaloids tested were obtained from Boehringer Ingelheim (Ingelheim, Germany). The solvents used for chromatography were of HPLC grade and were used as received. The different ionic modifiers were of analytical-reagent grade.

Chromatography

Separation of the enantiomers was achieved by a column based on the chiral stationary phase

(CSP) **tris(3,5-dimethylphenyl)carbamate** cellulose **[8],** commercially available as Chiralcel OD (Daicel). All mobile phases were premixed and degassed by ultrasonic treatment for 3 min. The enantiomeric separations were performed using two totally different mobile phases, one organic and the other aqueous.

The organic mobile phase always consisted of n-hexane-ethanol-methanol **(600:150:250)** with different modifiers when not specified otherwise. The modifier was dissolved in the alcoholic part of the mobile phase and diluted with n-hexane.

For the aqueous mobile phase **[9],** different modifiers were dissolved in water and mixed with methanol or acetonitrile.

All chromatographic studies were performed with a flow-rate of 0.5 **ml/min**. Detection was carried out at a wavelength of 230 nm unless stated otherwise. The volume injected was 10 **μl** at a concentration of 1 mg/ml.

RESULTS AND DISCUSSION

Stereochemistry of the tested racemates

The investigated racemates are quaternary N,N-dialkylated derivatives of noratropine and noratroscine. Different chain lengths of the axial bonded R_1 and the equatorial R_2 substituent lead to **(*r*)-** and (s)-configurations.

When the equatorial bonded substituent **(R_2)** is larger than the axial bonded substituent (R,), the quaternary nitrogen of atropine has the **(*s*)-**configuration, and when R_1 is larger than R_2 the molecule has the **(*r*)-configuration**. Analogous substitution in N-alkylated atroscine converts the configuration of the saturated ring system completely, including the quaternary nitrogen. In contrast to all other racemates, the racemates 4b and **4p** have **(*s*)-configuration** in the 3-position (pseudotropine = **β-tropine**).

The compounds can be divided structurally as follows.

(1) Compounds **4a** to 4g are of **(*s*)-configuration** (except 4b) with increasing R_2 (equatorial) alkyl chain length and a methyl group in R_1 (axial);

(2) Compounds **4a, 4h** to 41 are of **(*r*)-configuration** with a methyl group in R_2 (equatorial) and increasing alkyl chain length in R_1 (axial);

(3) Compounds **4a,** 4m and **4n:** the substances have in each instance the same alkyl substituents in equatorial (R,) and axial (R,) positions increasing in size from **4a** to **4n;**

(4) Compounds **4o** to **4q** are three of the four possible stereoisomers of the racemic tropic ester of **N-isopropylmethyl-3_/β-tropanol;**

(5) Compounds 4b and. **4p** are **3β-derivatives** of **4a** and 40 (so-called pseudotropanols);

(6) Compounds **5a** to **5f** are of **(*s*)-configuration** with a methyl group in R_2 (equatorial) and increasing alkyl chain length in R_1 (axial);

(7) Compounds **5d** and **5q** are the **(*r*)-** and @)-configuration racemic tropic esters of **N-butylmethyl-3α-tropanol;**

(8) Compounds **5a** and 5h: the nitrogen of **5a** is substituted with two methyl groups whereas the nitrogen of **5h** is substituted with two ethyl groups.

TABLE I

QUATERNARY N,N-DIALKYL DERIVATIVES OF NORATROPINE (4) AND NORATROSCINE (5)

Compound	R, (axial)	R_2 (equatorial)	Configuration
4a	CH_3	CH_3	–
4b	CH_3	CH_3	– (β-tropine)
4c	CH_3	C_2H_5	*s*
4d	CH_3	C_4H_9	*s*
4e	CH_3	C_5H_{11}	*s*
4f	CH_3	C_6H_{13}	*s*
4g	CH_3	C_7H_{13}	*s*
4b	C_2H_5	CH_3	
4i	C_3H_7	CH_3	
4k	C_4H_9	CH_3	r
41	C_5H_{11}	CH_3	
4m	C_2H_5	C_2H_5	
4n	C_4H_9	C_4H_9	–
40	iso-C_3H_7	CH_3	*r*
4p	iso-C_3H_7	CH_3	*r* (β-tropine)
4q	CH_3	iso-C_3H_7	*s*
5a	CH_3	CH_3	–
5b	C_2H_5	CH_3	*s*
5c	C_3H_7	CH_3	*s*
5d	C_4H_9	CH_3	*s*
se	C_5H_{11}	CH_3	
5f	C_6H_{13}	CH_3	*s*
5g	CH_3	C_4H_9	r
5h	C_2H_5	C_2H_5	–

Chromatographic results

The two different mobile phases were able to resolve all the investigated quatemary tropane alkaloids (see Table I). In all instances complete baseline separations with separation factors of up to 7.0 were achieved.

The chromatographic data presented in Tables II and III show similar results. In both systems racemic quatemary tropane alkaloids with larger axial alkyl groups were resolved with significantly higher separation factors than the respective N-epimers in the 8-position.

With mobile phase system A (Table II) the α -values are constant for larger equatorially bonded groups whereas more voluminious axially bonded groups show increasing separation factors leading to a maximum with the propyl-substituted molecule. Longer alkyl chains result in poorer separations. These effects are shown in

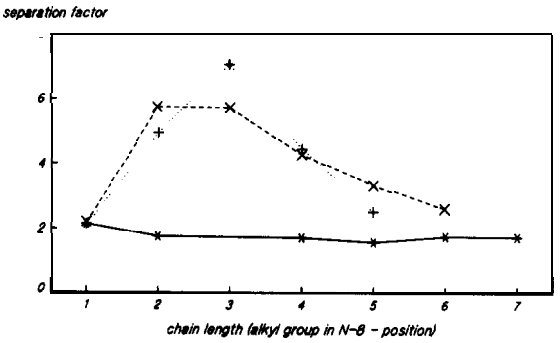

Fig. 2. Separation factor versus chain length (mobile phase A). Solid line = quatemary N-alkyl-N-methylnoratropine (s); dashed line = N-alkyl-N-methylnoratropine (r); dotted line = quatemary N-alkyl-N-methylnoratroscine (s).

Fig. 2. The same tendency is shown by the (*s*)- and (r) -configuration quatemary N-alkyl-N-methylatroscine derivatives.

With system B (see Table III) the separation factors do not depend on the chain length of the

TABLE II

MOBILE PHASE A: CAPACITY AND SEPARATION FACTORS OF QUATERNARY TROPANE ALKALOIDS USING *n*-HEXANE–ETHANOL–METHANOL (60:15:25) CONTAINING 2 m*M* OF TETRAPROPYLAMMONIUMBROMIDE

Compound	t_1	t_2	k_1'	k_2'	α
4a	8.67	13.02	0.84	1.77	2.11
4b	8.30	8.76	0.77	0.86	1.12
4c	8.14	10.69	0.73	1.27	1.74
4d	7.20	8.91	0.53	0.90	1.70
4e	6.84	8.09	0.46	0.72	1.57
4f	7.07	8.78	0.50	0.87	1.74
4g	7.05	8.76	0.50	0.86	1.72
4b	8.53	23.48	0.81	4.00	4.94
4i	7.86	26.88	0.67	4.72	7.04
4k	7.26	15.96	0.54	2.40	4.44
4l	7.21	10.89	0.53	1.32	2.49
4m	8.10	17.54	0.72	2.73	3.79
4n	6.39	11.38	0.36	1.42	3.94
4o	9.34	36.84	0.99	6.84	6.91
4Q	8.25	9.74	0.76	1.07	1.41
4q	7.67	9.57	0.63	1.04	1.65
5a	9.29	14.69	0.98	2.13	2.17
5b	9.83	33.99	1.09	6.23	5.72
5c	8.81	27.99	0.87	4.96	5.70
5d	8.27	19.95	0.76	3.24	4.26
5e	7.83	15.15	0.67	2.22	3.31
5f	8.64	14.90	0.84	2.17	2.58
5g	8.25	10.92	0.76	1.32	1.74
5b	9.70	26.78	1.06	4.70	4.43

TABLE III

MOBILE PHASE B: CAPACITY AND SEPARATION FACTORS OF QUATERNARY TROPANE ALKALOIDS USING 0.05 *M* SODIUM PERCHLORATE (pH 2)–ACETONITRILE (75:25) AS MOBILE PHASE

Compound	t_1	t_2	k_1'	k_2'	α
4a	9.41	10.27	0.74	**0.90**	1.21
4b	9.75	–	0.81	–	1.00
4c	11.53	15.17	1.14	1.81	1.59
4d	24.51	43.91	3.54	7.13	2.02
4e	37.14	45.67	5.88	7.46	1.27
4f	92.64	120.63	16.16	21.34	1.32
4g	198.99	265.67	35.85	48.20	1.34
4h	11.43	12.55	1.12	1.32	1.19
4i	15.67	25.84	1.90	3.79	1.99
4k	24.66	29.79	3.57	4.52	1.27
4l	36.81	45.23	5.82	7.38	1.27
4m	14.18	18.94	1.63	2.51	1.54
4n	64.60	143.00	**10.96**	25.48	2.32
4o	16.92	34.49	2.13	5.39	2.53
4p	15.43	–	1.86	–	1.00
4q	15.50	17.41	1.87	2.22	1.19
5a	8.90	10.07	0.65	0.86	1.33
5b	9.35	12.88	0.73	1.39	1.89
5c	14.25	20.54	1.64	2.80	1.71
5d	22.85	34.89	3.23	5.46	1.69
Se	39.89	64.83	6.39	11.01	1.72
5f	77.97	140.12	13.44	24.95	1.86
5g	22.31	28.08	3.13	4.20	1.34
5i	14.25	18.42	1.64	2.41	1.47

analytes. There is a linear dependence of log k_1' on chain length for both atropine and atroscine derivatives (Fig. 3). This is opposite of the chromatographic results with system A, where the capacity factors of the (*s*)-enantiomers decreased with increasing chain length of the alkyl chain in the 8-position (except for 4a).

In both systems A and B the quaternary tropane alkaloids with both R_1 and R_2 alkyl groups of increasing size (**4a, 4m, 4n; 5a, 5i**) are separated with higher separation factors. Esters of α-tropanol were obviously resolved better than the respective esters of β-tropanol (**4b, 4p**). The separation of these compounds is similar to that of compounds with larger equatorial alkyl chains [(*s*)-configuration for N-alkyl-N-methylnoratropine derivatives and (r) -configuration for N-alkyl-N-methylnoratroscine derivatives]. With mobile phase system B no separations of esters of β-tropanol were observed.

Figs. 4 and 5 show examples of the complete direct enantioseparation of selected racemates.

Influence of different modifiers

The above-mentioned correlations between retention behaviour and stereochemistry are also valid for the subsequently investigated modifiers.

Alkylsulphonic acids. In addition to the already described effects, the following dependences of the separation on the alkyl chain of the alkylsulphonic acids can be observed (methanesulphonic acid = MSA; butanesulphonic acid = BSA; heptanesulphonic acid = HSA; camphorsulphonic acid = CSA). With system A (Table IV) the capacity factors decrease with increasing size of the alkyl groups of the sulphonic acids, whereas no significant effect towards enantioselectivity is observed. In contrast, a minimum of retention with BSA as modifier in mobile phase system B (Table V) is found. The highest a-values (except for the N,N-dimethyl derivatives **4a** and **5a**) are observed with HSA as modifier.

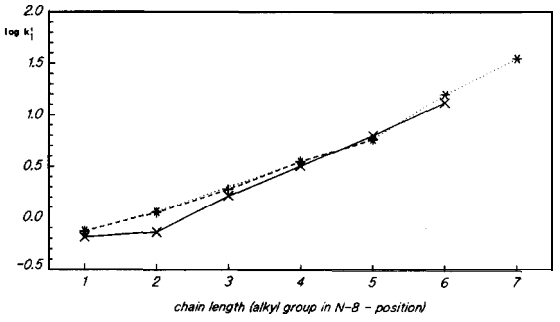

Fig. 3. Log (capacity factor, k_1') versus chain length (mobile phase B). Dotted line = quatemary N-alkyl-N-methylnoratropine (*s*); dashed line = quatemary N-alkyl-N-methylnoratropine (r); solid line = N-alkyl-N-methylnoratroscine (*s*).

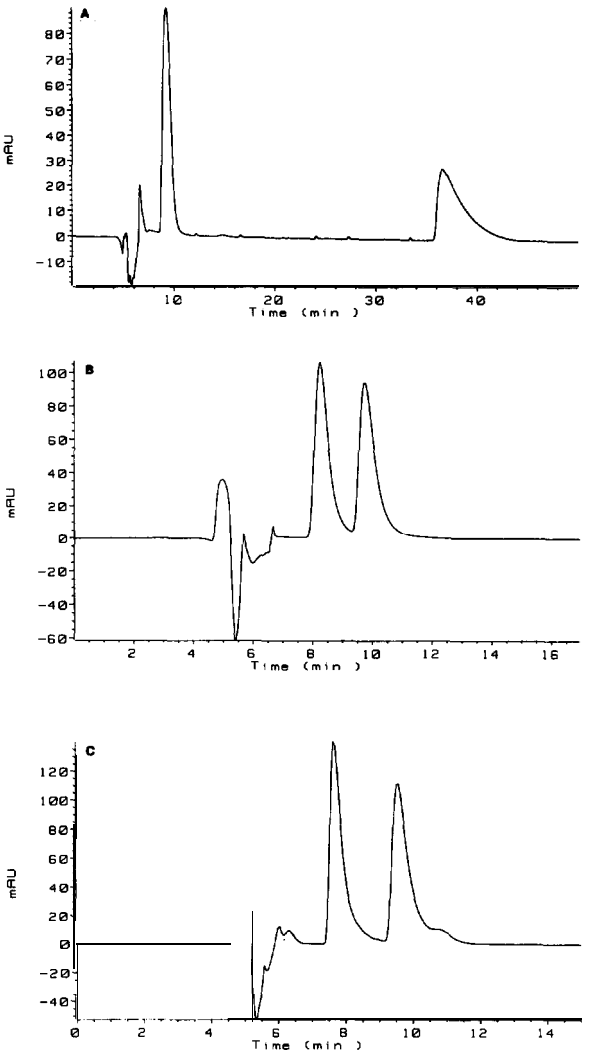

Fig. 4. Separation of (A) 4o, (B) the respective β-tropine form 4p and (C) the N-epimer 4q with mobile phase system A (for eluent, see Table II).

Halogen-containing modifiers. The influence of halogen-containing modifiers was tested. The following dependences of the separation on the

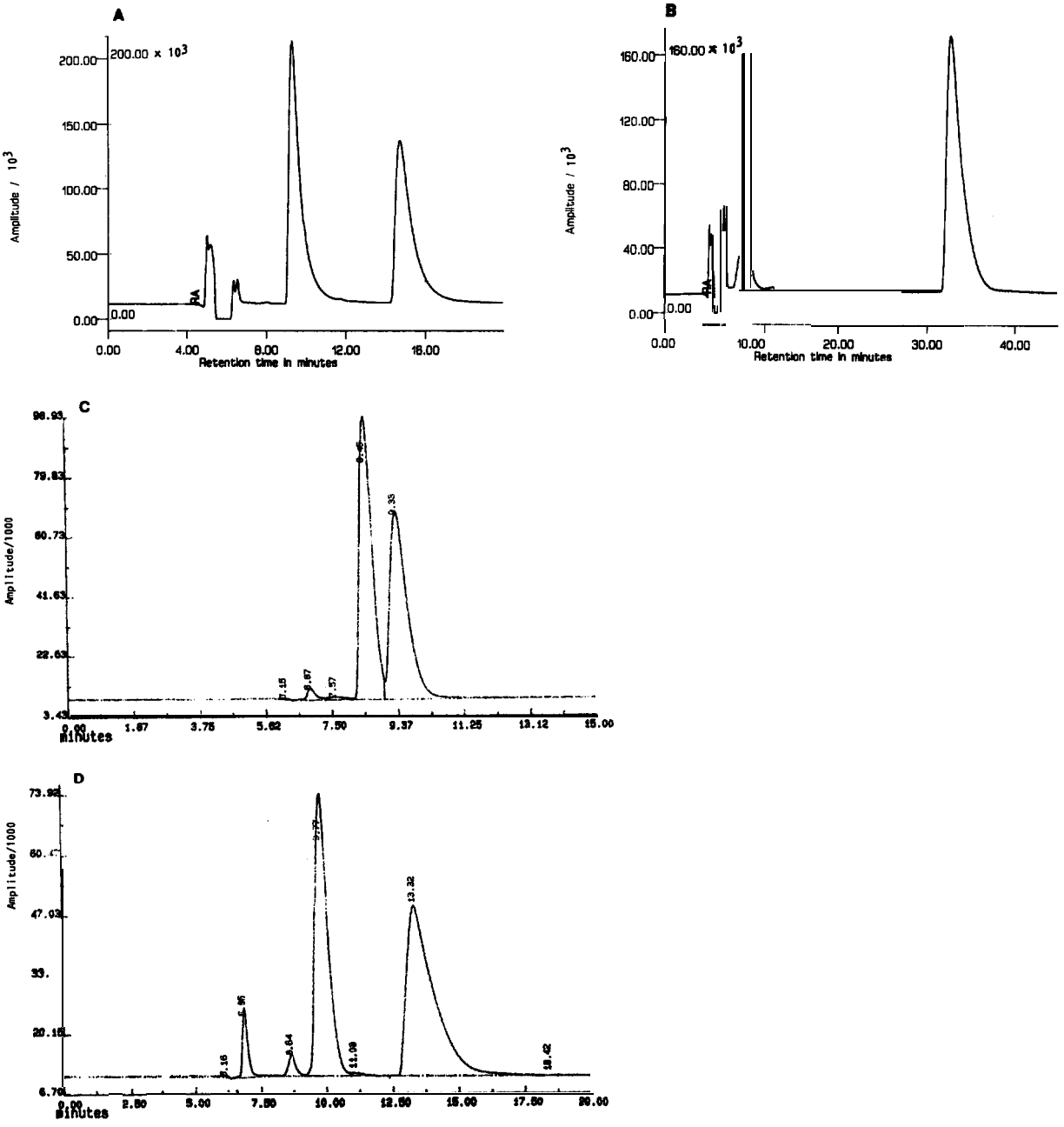

Fig. 5. Separation of the racemates 5a and 5b with both mobile phase systems (for eluents, see Tables II and III). (A) Racemate 5a in mobile phase A; (B) racemate 5b in mobile phase A; (C) racemate 5a in mobile phase B; (D) racemate 5b in mobile phase B.

alkyl chain length of the halides as anionic modifiers were observed. With mobile phase system A (Table VI), the capacity factors increase with increasing atomic number of the halide. The retention behaviour of the quaternary tropane alkaloids is independent of the cation of the modifier. Sodium bromide and tetrapropylammonium bromide as modifiers result in compar-

TABLE IV

CAPACITY AND SEPARATION FACTORS OF SELECTED QUATERNARY COMPOUNDS USING DIFFERENT ALKYLSULPHONIC ACIDS (2 m*M*) AS MODIFIERS IN MOBILE PHASE A

Compound	MSA			BSA			HSA		
	k_1'	k_2'	a	k_1'	k_2'	a	k_1'	k_2'	a
4e	0.45	0.51	1.13	0.34	0.49	1.44	0.32	0.42	1.31
4c	0.72	1.11	1.54	0.51	0.91	1.78	0.49	0.83	1.69
4h	0.79	3.17	4.01	0.64	2.87	4.48	0.51	2.72	5.33
41	0.49	1.02	2.08	0.47	0.87	1.85	0.45	0.79	1.76
5b	0.94	4.79	5.10	0.74	4.28	5.78	0.68	4.17	6.13
5e	0.51	1.51	2.96	0.49	1.36	2.78	0.45	1.28	2.84

TABLE V

CAPACITY AND SEPARATION FACTORS OF SELECTED QUATERNARY COMPOUNDS USING DIFFERENT ALKYLSULFONIC ACIDS (0.05 m*M*) AS MODIFIERS IN MOBILE PHASE B

Compound	MSA			BSA			HSA			CSA		
	k_1'	k_2'	a	k_1'	k_2'	a	k_1'	k_2'	a	k_1'	k_2'	a
4a	*0.22*	*0.56*	*2.50*	–	–		*0.65*	*0.72*	*1.12*	0.30	0.67	2.35
4h	*0.81*	–	**1.00**	0.34	–	**1.00**	*0.91*	**1.02**	1.12	2.91	2.63	1.20
4c	**0.81**	1.15	1.41	0.33	0.44	1.35	0.94	1.37	1.46	0.41	0.56	1.36
4e	7.92	8.57	1.08	2.42	4.11	1.70	7.89	14.34	1.82	2.94	4.85	1.65
41	1.00	1.15	1.15	1.32	1.58	1.20	4.18	5.26	1.26	1.59	1.93	1.21
5a	0.37	0.48	1.30	0.21	0.24	1.14	0.57	0.70	1.22	0.22	0.28	1.25
5b	0.59	0.93	1.56	0.27	0.41	1.53	0.78	1.32	1.68	0.33	0.52	1.56
5e	4.00	7.33	1.83	1.46	2.39	1.64	5.01	8.18	1.63	1.74	2.85	1.64

TABLE VI

CAPACITY AND SEPARATION FACTORS OF SELECTED QUATERNARY COMPOUNDS USING DIFFERENT HALOGEN-CONTAINING SALTS (2 m*M*) AS MODIFIERS IN THE MOBILE PHASE A

Compound	NaF			TMeACl			TPrABr			KI			NaBr		
	k_1'	k_2'	a	k_1'	k_2'	a	k_1'	k_2'	a	k_1'	k_2'	a	k_1'	k_2'	a
4c	0.45	1.06	2.36	*0.64*	**1.11**	**1.73**	**0.79**	**1.38**	**1.75**	**1.06**	**1.77**	**1.67**	**0.74**	**1.30**	**1.76**
4e	0.28	0.45	1.61	**0.40**	**0.51**	**1.28**	**0.47**	**0.77**	**1.64**	**0.60**	**0.98**	**1.63**	**0.47**	**0.74**	**1.57**
4h	0.70	1.30	1.86	**0.70**	**3.60**	**5.14**	**0.87**	**4.53**	**5.21**	**1.15**	**4.89**	**4.25**	**0.81**	**4.26**	**5.26**
4l	0.47	0.70	1.49	**0.45**	**1.17**	**2.60**	**0.57**	**1.43**	**2.51**	**0.72**	**1.91**	**2.65**	**0.53**	**1.38**	**2.60**
5b	0.40	0.64	**1.60**	**0.87**	**5.55**	**6.38**	**1.09**	**6.23**	**5.72**	**1.43**	**7.66**	**5.36**	**1.02**	**6.70**	**6.57**
5e	0.43	0.74	1.72	**0.45**	**1.89**	**4.20**	**0.64**	**2.21**	**3.45**	**0.89**	**2.53**	**2.84**	**0.62**	**2.17**	**3.50**

able capacity and separation factors. It can be concluded that the chromatographic behaviour is strictly independent of the different cations, but is clearly determined by the anion. The use of fluoride leads to lower a-values for the analytes with larger axial substituents, whereas no significant differences were observed with the other halides.

With mobile phase system B (Table VII) no significant difference between the different halides and the perchlorate anion was observed.

Influence of modifier concentration. All tropane alkaloids displayed similar behaviour in each of the mobile phase systems (see Tables VIII and IX).

Discussion of the separation mechanism

For the counter ion bromide the capacity factor k_1' increases with increasing concentration of the counter ion up to a point where ion-pair formation reaches a maximum and then remains constant. Knox and Laird [10] suggested that a combination of adsorption (partition) and cluster formation may be the dominant factor for retention. At lower concentrations, partitioning of the ion pair into the stationary phase may be the major factor controlling the retention. As proof of this theory, the chromatographic data without the use of an ionic modifier can be considered: no separation occurs with mobile phase systems B and distorted peak shapes were observed; with mobile phase A also very poor peak shape were obtained, thus giving only very slight separations.

When cluster formation occurred at higher concentrations, the solubility of the ion pair increased in the mobile phase, resulting in a decrease in retention [11]. The curves show a concentration maximum of about 2 mmol for mobile phase A (Fig. 6) and about 10 mmol for mobile phase B (Fig. 7). These are the optimum concentrations of bromide for maximum retention for the tested racemates. The optimum concentrations are dependent on the strength of the ion pair formed, the extent of adsorption on the stationary phase and the extent of the formation of clusters of the counter ion bromide.

The chromatographic data show a dependence of capacity factors on the concentration of the ionic modifier and so participation of an ion-pair mechanism can be concluded.

The ion-pair mechanism for the relationship between retention times and modifier concentration is not useful for explaining the enantiomeric separation. A possible interpretation may be the inclusion of ion pairs in cellulose cavities, similarly to the mechanism discussed for a CSP in the normal-phase mode (hexane–propanol). Hence an interplay between an ion-pair mechanism and inclusion leads to the direct enantiomeric separations of the quaternary tropane alkaloids achieved here.

TABLE VII

DEPENDENCE OF CAPACITY AND SEPARATION FACTORS OF QUATERNARY COMPOUNDS ON HALIDE AND PERCHLORATE ANIONS IN MOBILE PHASE B (0.05 *M*; pH 2)

Compound	NaBr			NaCl			NaF			$NaClO_4$		
	k_1'	$k;$	α	$k;$	k_2'	a	k_1'	$k;$	a	k_1'	k_2'	a
4a			1.31			1.44			2.26			1.21
4c	0.24	0.32	1.31	0.26	0.34	1.32	0.27	0.36	1.33	1.14	1.81	1.59
4h	0.25	–	1.00	0.86	–	Shoulder	0.96	–	Shoulder	1.12	1.32	1.19
4e	1.76	2.86	1.62	1.88	3.01	1.60	1.89	3.09	1.63	5.88	7.46	1.27
41	0.98	1.17	1.19	1.06	1.23	1.16	1.09	1.29	1.18	5.82	7.38	1.27
5a	0.17	–	Shoulder	0.44	–	Shoulder	0.51	0.61	Shoulder	0.65	0.86	1.33
5b	0.20	0.30	1.48	0.22	0.33	1.48	0.24	0.34	1.42	0.73	1.39	1.89
se	1.02	1.61	1.59	1.04	1.67	1.61	1.13	1.73	1.54	6.39	11.01	1.72

TABLE VIII

DEPENDENCE OF CAPACITY AND SEPARATION FACTORS OF SELECTED QUATERNARY COMPOUNDS ON TETRAPROPYLAMMONIUM BROMIDE CONCENTRATION IN SYSTEM A

Compound	0 mM			0.02 mM			0.56 mM			2 mM			18 mM			34 mM			50 mM		
	k_1'	k_2'	α	k_1'	k_2'	a	k_1'	k_2'	α	k_1'	k_2'	a	k_1'	k_2'	α	k_1'	k_2'	α	k_1'	k_2'	α
4c	0.47	0.87	1.85	0.40	0.70	1.75	0.74	1.30	1.76	0.79	1.38	1.75	0.64	1.17	1.83	0.53	1.02	1.92	0.64	1.00	1.56
4e	0.31	0.48	1.55	0.13	0.40	3.08	0.45	0.79	1.76	0.47	0.77	1.64	0.38	0.66	1.74	0.36	0.57	1.58	0.34	0.60	1.77
4h	0.59	1.78	3.02	0.43	2.00	4.65	0.83	4.00	4.82	0.87	4.53	5.21	0.70	3.89	5.56	0.62	3.43	5.53	0.68	2.89	4.25
4l	0.39	0.83	2.13	0.36	0.72	2.00	0.47	1.34	2.85	0.57	1.43	2.51	0.45	1.23	2.73	0.38	1.11	2.92	0.38	1.00	2.43
5b	0.70	2.74	3.91	0.45	3.19	2.74	0.79	5.94	7.52	1.09	6.23	5.72	0.89	6.00	6.74	0.79	5.34	6.76	0.79	4.34	5.49
Se	0.46	1.24	2.70	0.45	1.26	2.80	0.45	1.98	4.40	0.64	2.21	3.45	0.47	1.98	4.21	0.40	1.74	4.35	0.60	1.49	2.48

TABLE IX

DEPENDENCE OF CAPACITY AND SEPARATION FACTORS OF SELECTED QUATERNARY COMPOUNDS ON THE CONCENTRATION OF SODIUM BROMIDE IN SYSTEM B

The peak shape in the absence of modifier is distorted and the results cannot be evaluated.

Compound	0.001 *M*			0.002 *M*			*0.005 M*			0.01 *M*			*0.05 M*		
	k_1'	k_2'	*a*	k_1'	k_2'	*a*	k_1'	k_2'	*a*	k_1'	k_2'	α	k_1'	k_2'	α
4a	0.20	–	1.00	0.12	0.24	1.98	0.12	0.23	1.91	0.11	–	1.00	0.19	0.25	1.31
4c	0.27	0.39	1.41	0.31	0.43	1.39	0.28	0.39	1.39	0.32	0.45	1.39	0.24	0.32	1.31
4h	0.28	–	1.00	0.31	–	1.00	0.30	–	1.00	0.33	–	1.00	0.25	–	1.00
4e	2.13	3.83	1.80	2.17	4.66	I.87	2.16	3.86	1.79	3.60	5.88	1.63	1.76	2.86	1.62
4l	1.14	1.42	1.24	1.26	1.54	1.23	1.16	1.42	1.22	1.31	1.61	1.23	0.98	1.17	1.19
5a	0.19	–	Shoulder	0.22	–	Shoulder	0.21	–	Shoulder	0.23	0.27	1.17	0.17	–	Shoulder
5b	0.23	0.36	1.55	0.27	0.42	1.56	0.25	0.37	1.48	0.27	0.42	1.55	0.20	0.30	1.48
5e	1.34	1.91	1.42	0.72	1.16	1.61	0.51	0.85	1.44	0.77	1.29	1.45	1.02	1.61	1.59

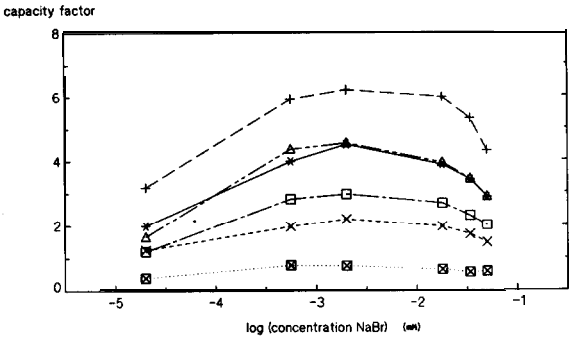

Fig. 6. Dependence of capacity factor (k_2') on log (modifier concentration) with a maximum at a concentration at 2 m*M* (mobile phase A). Δ = 4c; ❑ =de; * = 4h; ❑ =4I; + = 5b; x = 5e.

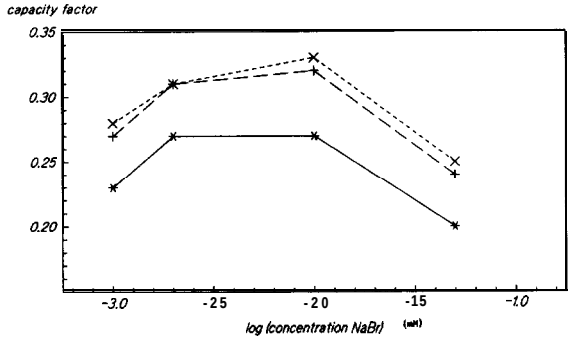

Fig. 7. Dependence of capacity factor (k_1') on the log (modifier concentration) with a maximum at a concentration at 10 m*M* (mobile phase B). + = 4c; x = 4h; * = 5b.

CONCLUSIONS

Because of the relationship between the retention behaviour of the racemates and the the ionic modifiers used, the involvement of an ion-pair mechanism in chiral recognition can be concluded. For the resolution of the tertiary derivatives of atropine and atroscine inclusion in the chiral cavities of the cellulose carbamate is sufficient whereas for quaternary derivatives the formation of ion pairs is a basic requirement. Systematic investigations on the separation mechanism permit the separation of quaternary tropane alkaloids to be predicted.

In addition to the known synthetic methods for the preparation of quaternary tropane alkaloids, there is now an alternative possibility of obtaining enantiomerically pure drugs by methods of preparative liquid chromatography.

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