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Note

High-speed liquid chromatographic separation of some Strychnos alkaloids

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Bisset and his co-workers have studied the separation of some *Strychnos* alkaloids by means of thin-layer chromatography $(TLC)^1$ and gas-liquid chromatography $(GLC)^2$. Since we had obtained a fairly good separation of a series of alkaloids by high-performance liquid chromatography $(HPLC)^{3,4}$, we found it of interest to try this technique for the separation of a number of alkaloids related to strychnine.

EXPERIMENTAL

The analyses were carried out on a Packard Model 8200 liquid chromatograph equipped with a UV detector (the wavelength 254 nm was used) and a stainless-steel column (30 cm \times 2 mm I.D.) filled with Merckosorb Si 60 (5 μ m); the balanced-density slurry technique was used for filling the column. The column temperature was maintained at 20°. The solvents used (diethyl ether, methanol and diethylamine) were *pro analysi* grade (Baker). The analyses were carried out at a flow-rate of 2.00

TABLE I

RETENTION TIMES OF SOME STRYCHNOS ALKALOIDS Column conditions as specified in Experimental.

Alkaloid	<i>Retention time</i> (<i>min</i>) <i>in solvent</i> <i>system</i>	
	1	11
Icajine	4.2	2.6
Vomicine *	4.6	1.6
Pseudostrychnine	6.8	
Strychnine	7.2	12.4
4-Hydroxystrychnine	7.6	
a-Colubrine	8.8	14.3
Spermostrychnine	9.8	
β-Colubrine	10.3	10,2
Diaboline	16.0	10.9
Brucine	18.4	17.6
Serpentine	>20	
Alstonine	>20	

NOTES



Fig. 1. Chromatogram of some *Strychnos* alkaloids in solvent system I (see text). I = Icajine; V = vomicine; S = strychnine; P = pseudostrychnine; $\alpha = \alpha$ -colubrine; $\beta = \beta$ -colubrine; D = diaboline; B = brucine.

ml/min at a pressure of 205 kg/cm² for solvent system I (diethyl ether containing 1% of diethylamine) and at a flow-rate of 1.15 ml/min at 200 kg/cm² for solvent system II [diethyl ether-methanol (1:1)].

DISCUSSION

The separation of 12 Strychnos alkaloids related to strychnine by means of HPLC is shown in Table I and Fig. 1. When the results are compared with the separations obtained by Bisset and co-workers with TLC¹ and GLC², the following differences are observed: diaboline is retained more in HPLC than in TLC, perhaps because of the higher amount of diethylamine used in TLC; and α -colubrine and β -colubrine, which could not be separated completely with GLC or TLC, are well separated by HPLC. Although fairly good separation of α - and β -colubrines could also be obtained in a neutral solvent system (system II), tailing made this system less useful because of the acidic properties of the silica gel⁴.

REFERENCES

- 1 J. D. Phillipson and N. G. Bisset, J. Chromatogr., 48 (1970) 493.
- 2 N. G. Bisset and P. Fouché, J. Chromatogr., 37 (1968) 172.
- 3 R. Verpoorte and A. B. Svendsen, J. Chromatogr., 100 (1974) 227.
- 4 R. Verpoorte and A. B. Svendsen, J. Chromatogr., 100 (1974) 231.