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Liquid chromatographic retention of β-adrenoceptor antagonists: an index of lipid solubility

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The in-vitro lipophilicity of nine β-adrenoceptor antagonists was evaluated based on retention on a reverse-phase C-18 high-pressure liquid chromatographic (hplc) system at physiologic pH. Propranolol was by far the most lipophilic drug, while atenolol and sotolol were the least. Hplc retention was highly correlated (r = 0·92) with octanol : buffer partition coefficient. Thus hplc retention is a rapid and replicable approach to the determination of in-vitro lipophilicity that does not require radioactive drug.

Lipid solubility may be an important determinant of the pharmacokinetic and metabolic properties of β-adrenoceptor antagonists, as well as their uptake into central nervous system (Deacon et al 1981; Jack 1981; Taylor et al 1981; Arendt et al 1983a). The lipid solubility of these and other drugs has traditionally been evaluated using partition ratios, in particular the octanol : water partition coefficient (Leo et al 1971). The retention of a compound in a chromatographic system is another approach to evaluation of lipophilicity (Wells et al 1981; Hammers et al 1982). Reverse-phase high pressure liquid chromatography (hplc) partitions and separates drugs based in part on their polarity. The present study evaluated hplc retention as an approach to quantitating the lipid solubility of β-adrenergic blocking agents.

Methods

A Waters Associates (Milford, MA, USA) hplc system was used for analyses (Divoll et al 1982). The mobile phase was prepared by dissolving 1.65 g of K_2HPO_4 and 2·1 g of KH_2PO_4 in distilled water, final volume 0·6 litres. To this was added 0·4 litres or pure methanol and the pH was adjusted to 7·4 by dropwise addition of phosphoric acid. Aqueous and organic components of the mobile phase were separately filtered before mixing, and the final mixture was degassed before use. Flow rate was 1·5 ml min^{-1} along a 30 cm reverse-phase microBondapak C-18 stainless steel column, containing a stationary phase of octadecyl silane bonded silica, coated 12% with carbon and fully end-capped with trimethyl silane to remove free hydroxyl groups. All analyses were at room temperature (20 °C).

Pure reference standards of the nine drugs shown in Table 1, supplied from their manufacturers, were dissolved in methanol and sequentially injected onto the chromatograph. Column effluent was monitored using an ultraviolet detector operated at 254 nm.

Results

The retention time of the nine drugs ranged from 16·2 min for propranolol to 3·2 min for sotalol (Table 1). These retention times were compared with octanol : buffer partition ratios measured at 37 °C and at physiological pH (Fig. 1) (Woods & Robinson 1981). The two measures of lipophilicity were highly intercorrelated (r = 0·92).

Discussion

We have previously used hplc retention to quantitate lipophilicity of benzodiazepine derivatives (Arendt et al

Table 1. Liquid chromatographic retention of β-adrenoceptor antagonists.

	Drug	Retention time (min)
1.	Propranolol	16·20
2.	Oxprenolol	8·77
3.	Metoprolol	6·49
4.	Acebutolol	6·41
5.	Timolol	6·20
6.	Nadolol	4·32
7.	Pindolol	4·20
8.	Atenolol	3·35
9.	Sotalol	3·20

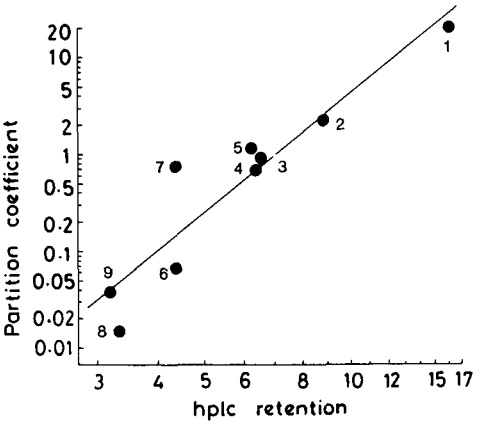

FIG. 1. Relation of hplc retention (min) to octanol : buffer partition ratio for each of the drugs. Both scales are logarithmic. The correlation (r = 0·92) is highly significant ($P < 0·01$). Figures relate to Table 1.

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1983b; Greenblatt et al 1983). Comparison of in-vitro hplc retention, based on the method described above, with in-vivo drug distribution both in animals and man indicates that hplc retention is at least as reliable an index of in-vivo distribution as is the octanol : buffer partition coefficient. The present report indicates that hplc retention can be used to quantitate lipophilicity of β-adrenoceptor antagonists. Hplc retention was highly correlated with the octanol : buffer partition coefficient and confirmed propranolol to be by far the most lipophilic, with sotalol and atenolol the least lipophilic.

Hplc retention has a further merit in that it does not require the use of radioactive material, and it is easily standardized for comparisons of relative lipophilicity among many drugs.

REFERENCES

Arendt, R. M., Greenblatt, D. J., deJong, R. H. (1983a) Clin. Research. 31: 626A

Arendt, R. M., Greenblatt, D. J., deJong, R. H., et al (1983b) J. Pharmacol. Exp. Ther. 227: 98–106

Deacon, C. S., Lennard, M. S., Bax, N. D. S., Woods, H. F., Tucker, G. T. (1981) Br. J. Clin. Pharmacol. 12: 429–431

Divoll, M., Greenblatt, D. J., Shader, R. I. (1982) Pharmacology 24: 261–266

Greenblatt, D. J., Arendt, R. M., Abernethy, D. R., et al. (1983) Br. J. Anaesth. 55: 985–989

Hammers, W. E., Meurs, G. J., De Ligny, C. L. (1982) J. Chromatogr. 247: 1–13

Jack, D. B. (1981) Br. J. Clin. Pharmacol. 11: 402–406

Leo, A., Hansch, C., Elkins, D. (1971) Chem. Rev. 71: 525–616

Taylor, E. A., Jefferson, D., Carroll, J. D., Turner, P. (1981) Br. J. Clin. Pharmacol. 12: 549–559

Wells, M. J. M., Clark, C. R., Patterson, R. M. (1981) J. Chromatogr. Sci. 19: 573–582

Woods, P. E., Robinson, M. L. (1980) J. Pharm. Pharmacol. 33: 172–173

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The effect of week-long infusion of propranolol, via an osmotic minipump, on blood pressure, heart rate and vascular responses in spontaneously hypertensive rats

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Propranolol was infused in SHR subcutaneously for 7 days at two concentrations (either 3·75 or 7·5 mg kg^{-1} day) via a minipump. Mean blood pressure and heart rate measured under pentobarbitone anaesthesia on day 7 after implantation showed a significant dose-dependent decrease in both propranolol-treated groups. In the low-dose propranolol-treated rats, there was no change in contractile responses to phenylephrine over controls. In rats receiving the higher dose of propranolol there was a significant increase in the response to phenylephrine. There was no change in the relaxation response of any of the groups to isoprenaline. The results indicate that propranolol, while lowering blood pressure and heart rate, is also modifying the α-receptor response of the vascular wall in the spontaneously hypertensive rat.

Some studies have suggested that β-adrenergic blocking drugs may lower arterial pressure by a central mechanism (Day & Roach 1974). Smits et al (1980a) concluded, however, that the antihypertensive effects of propranolol in spontaneously hypertensive rats (SHR) were not caused by an action of the drug in the central nervous system. Pritchard & Gillam (1969) postulated that the hypotensive effect resulted from a resetting of the baroreceptors. Other studies (Frohlich et al 1968; Buhler et al 1973) have suggested that reductions in cardiac output or renin release are the primary causes. Some in-vitro studies have indicated that a prejunctional block of sympathetic nerve activity may be produced by propranolol (Day et al 1968). Others (Eliash & Weinstock 1971) also reported that propranolol in low doses reduced contractions of the nictitating membrane in the cat.

Smits et al (1980b), using the Alzet minipump for chronic, controlled delivery of propranolol in the SHR, were able to show a decrease in both blood pressure and heart rate. We have recently reported (Sun & Hanig 1983a) that the contractile response of aortic rings to phenylephrine in untreated SHR is markedly lower than that of controls of the Wistar-Kyoto (WKY) strain. We have also shown that week-long exposure of the normal Holtzman rat to propranolol, via the minipump, is without effect on blood pressure, heart rate or vascular reactivity (Sun & Hanig 1983b). We therefore considered it relevant to investigate the effect of prolonged infusion of propranolol on in-vitro vascular responses to phenylephrine and isoprenaline in the SHR and to relate these to observed changes in blood pressure and heart rate. The objective of this approach was to determine whether changes in reactivity of the arterial wall play a significant role in the cardiovascular actions of propranolol.

Materials and methods

Male SHR (Tac(N)SHR, derived originally from NIH, Taconic Farms Inc., Germantown, N.Y., USA), age 5

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