EFFECT OF HALOFENATE ON THE SERUM BINDING OF PHENYTOIN

Halofenate free acid, the plasma form of the hypolipidemic drug halofenate (MK-185), reduces the plasma protein binding of salicylic acid, aspirin, chlorothiazide, and tolbutamide (Hucker, Stauffer & White, 1972), and may displace propranolol from plasma protein binding sites (Huffman, Azarnoff, Shoeman & Dujovne, 1976). Halofenate free acid also displaces endogenous thyroxine (Karch, Morgan, Kubasik & Sine, 1976; Davis, Hsu, Bianchine & Morgan, 1972) and uric acid (Schlosstein, Kippen, Whitehouse, Bluestone, Paulus & Klinenberg, 1973) from their plasma protein binding sites. Phenytoin is highly bound to plasma proteins in man (Barth, Alvan, Borga & Sjoqvist, 1976), and plasma phenytoin binding is reduced by salicylic acid, phenylbutazone (Lunde, Rane, Yaffe, Lund & Sjoqvist, 1970), renal disease (Odar-Cederlof, Lunde & Sjoqvist, 1970; Reidenberg, Odar-Cederlof, von Bahr, Borga & Sjoqvist, 1971; Hooper, Bochner, Eadie & Tyrer, 1974), and hepatic disease (Hooper, Bochner, Eadie & Tyrer, 1974; Affrime & Reidenberg, 1975). Phenytoin therapy is frequently monitored by the serum total phenytoin level, and reduced plasma protein binding complicates the interpretation of serum total phenytoin levels in patients (Odar-Cederlof *et al.*, 1970). If halofenate free acid reduces the plasma protein binding of phenytoin, then serum total phenytoin levels might be difficult to interpret when halofenate and phenytoin are taken concurrently. This study evaluates the effect of halofenate free acid on the serum binding of phenytoin.

Fresh serum was obtained from healthy volunteers. Serum phenytoin binding was measured by ultrafiltration at 37°C with therapeutic concentrations of added phenytoin (10 and 20 µg/ml) and 0, 250, 500, or 1000 µg halofenate free acid/ml serum. The halofenate free acid concentrations are consistent with the serum levels obtained with therapeutic doses of halofenate (Karch *et al.*, 1976). 5,5-diphenylhydantoin-4-[¹⁴C] (10 µl of 495 µM) (specific activity 50.5 Ci/mol; New England Nuclear, Boston, MA, USA) was added as a

tracer to 5 ml serum samples, and after gentle mixing duplicate 200 µl aliquots were removed for liquid scintillation counting. After 30 min equilibration the serum samples were ultrafiltered at 37°C in an Amicon Ultrafiltration Cell (model MMC, Amicon Corp., Lexington, MA, USA) with an XM-50 membrane (Amicon) under a gas (98% nitrogen, 2% carbon dioxide) pressure of 40 psi. The first 1.0 ml filtrate was discarded, and duplicate 200 µl aliquots of the next 0.5 ml filtrate were removed for liquid scintillation counting. The serum and filtrate samples (200 µl) were added to 0.8 ml distilled water and 10 ml ScintiVerse (Fisher Scientific Co., Fairlawn, NJ, USA), and counted with a Packard Tricarb Liquid Scintillation Counter (Model 3385, Packard Instrument Co., Downers Grove, IL, USA). The percent free phenytoin was calculated for each sample as: (DPM filtrate/DPM serum) × 100, and statistical comparison of the mean values for each group was evaluated by one-way analysis of variance.

The effect of halofenate free acid on the serum binding of phenytoin is summarized in Table 1. Halofenate free acid increased the percent free phenytoin, i.e. reduced the serum binding of phenytoin, at each level of added halofenate free acid, and the effect was progressively larger as the concentration increased from 250–1000 µg halofenate free acid/ml serum. There was no significant difference in the magnitude of the effect with serum phenytoin concentrations of 10 or 20 µg/ml, and additions of 1000 µg halofenate free acid/ml serum increased the free phenytoin level by about 140%.

The percent free phenytoin measured in the fresh serum samples from healthy individuals without added halofenate free acid agrees with the values reported from other laboratories (Lunde *et al.*, 1970; Hooper *et al.*, 1974). The addition of halofenate free acid in concentrations achieved in serum during halofenate therapy caused a significant reduction in the serum binding of phenytoin, and the magnitude of the effect was proportional to the halofenate free acid con-

Table 1 Effect of halofenate free acid on the serum binding of phenytoin in man. Values are % free phenytoin (mean ± s.e. mean, *n* = 8)

Phenytoin (µg/ml)	Halofenate free acid concentration (µg/ml)			
	0	250	500	1000
10	9.2 ± 0.1	10.5 ± 0.2	13.4 ± 0.6*	22.1 ± 1.5**
20	10.8 ± 0.5	12.1 ± 0.7	15.2 ± 3.6*	26.6 ± 2.1**

* *P* < 0.005 compared with the 0 and 250 µg/ml halofenate free acid samples.

** *P* < 0.001 compared with the 0, 250 and 500 µg/ml halofenate free acid samples.

centration. Since phenytoin therapy is frequently monitored by serum total (free plus bound) phenytoin levels, and reduced phenytoin serum binding complicates the interpretation of the serum total phenytoin level in patients (Odar-Cederlof *et al.*, 1970), then the reduced serum binding of phenytoin caused by halofenate free acid may complicate therapy when halofenate and phenytoin are taken concurrently.

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