PLASMA PROTEIN BINDING OF PHENYTOIN IN 100 EPILEPTIC PATIENTS

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The plasma protein binding of phenytoin was investigated in 100 epileptic patients, using equilibrium dialysis at 37°C. The unbound fractions of phenytoin in plasma formed a skewed distribution, with a range of 9.7 to 24.7% and a median value of 12.3%. Most (80%) patients appeared to form one group with free phenytoin fractions from 9.7 to 14.5%, while the remainder formed a group with elevated free fractions (> 14.5%). The total and unbound plasma concentrations of phenytoin were strongly correlated ($r = 0.95$, $P < 0.0001$). There was a weak correlation between increasing age and the unbound phenytoin fraction ($r = 0.28$, $P < 0.01$). The results indicate that measurement of the total phenytoin concentration in plasma should usually provide a reliable index of anticonvulsant effect. However, determination of the unbound phenytoin fraction would be beneficial in the management of those patients in whom this fraction may be elevated, due to interacting drugs or biochemical abnormalities.

Introduction

Phenytoin is known to be extensively (approximately 90%) bound to plasma proteins, and the cerebrospinal fluid level of phenytoin reflects the unbound (free) concentration, rather than the total phenytoin concentration, in plasma (Lund *et al.*, 1972; Schmidt & Kupferberg, 1975). One would therefore expect the free plasma level to be a more direct measure of biological effect than the total phenytoin level (Eadie, 1976). In fact, Booker & Darcey (1973) have shown that the toxic effects of phenytoin correlate best with the free level of drug in plasma.

Clinically, total plasma phenytoin levels are used to monitor dose-effectiveness, but this is only reliable if the inter-patient variability in the free fraction of phenytoin is small. Controversy has existed, however, about the actual variability in phenytoin plasma protein binding (Richens, 1979). This report details the inter-individual variation in 100 epileptic patients being treated with phenytoin.

Methods

For 7 months, each plasma phenytoin determination at the Royal Hobart Hospital was followed by a protein binding measurement, except when insufficient plasma remained. The unselected donor patients comprised 45 females and 55 males, aged 5 to 90 years. Most were receiving other drugs, including anticonvulsants, in addition to phenytoin. Total

phenytoin plasma levels were measured by EMIT (Syva, Palo Alto, CA, USA). Phenytoin binding to plasma proteins was measured by equilibrium dialysis at 37°C (Ehrnebo *et al.*, 1971) using 5,5-diphenyl-[4-¹⁴C]-hydantoin] (New England Nuclear). The method had a coefficient of variation of 2.2% ($n = 10$).

Results

The unbound phenytoin fractions ranged from 9.7 to 24.7%, with a median value of 12.3%. A probability plot (David, 1970) for the unbound phenytoin fraction (Figure 1) showed two principal segments, indicating the presence of two separable groups of patients. The combined sample formed a positively skewed distribution of unbound phenytoin fractions; the coefficient of skewness (Snedecor & Cochran, 1980) was 1.81 ($P < 0.01$). The free phenytoin concentration, calculated from the total plasma concentration and the unbound fraction, was strongly correlated with the total phenytoin concentration in plasma ($r = 0.95$, $P < 0.0001$). However, the unbound phenytoin fraction did not vary with the total plasma concentration of phenytoin ($r = -0.07$, $P > 0.50$). A weak correlation existed between the unbound fraction of phenytoin and the age of the patient ($r = 0.28$, $P < 0.01$). The unbound phenytoin fraction did not differ significantly between the sexes (Mann-Whitney $U = 1217.5$, $z = 0.14$, $P > 0.5$).

Figure 1 Probability plot of % phenytoin unbound in plasma for 100 patients.

Discussion

We found a 2.5-fold variation in the unbound plasma fraction of phenytoin for the study patients. This variation is similar to that reported by Barth *et al.* (1976), who investigated the plasma protein binding of phenytoin in 63 patients, using an ultrafiltration technique at room temperature. These results contrast with the much larger inter-individual variation reported in earlier studies (Svensmark *et al.*, 1960; Triedman *et al.*, 1960; Viukari & Tammisto, 1969; Booker & Darcey, 1973; Bochner *et al.*, 1974; Hooper *et al.*, 1974). Imperfect ultrafiltration membranes used in several of these studies may account for most of this discrepancy (Porter & Layzer, 1975; Barth *et al.*, 1976).

Twenty patients with unbound phenytoin fractions above 14.5% formed a distinct group. There was no apparent reason for the elevated unbound fraction in

two of these patients, although their serum albumin levels were not available. Likely explanatory details for the other 18 patients are given in Table 1. Valproate, warfarin, phenylbutazone, and salicylates are highly protein bound drugs capable of displacing phenytoin from its binding sites on albumin (Kober *et al.*, 1980; Sjöholm *et al.*, 1979; Lunde *et al.*, 1970). Liver disease, renal disease, and hypoalbuminaemia can also elevate the unbound fraction of phenytoin in plasma (Hooper *et al.*, 1974; Porter & Layzer, 1975), and hyperthyroidism decreases the protein binding of warfarin (Feely *et al.*, 1981). None of the major group of 80 patients was receiving drugs known to compete with phenytoin for albumin binding sites, or had liver, thyroid, or renal disease. A weak, but statistically significant, correlation existed between the age of the patient and the unbound phenytoin fraction. Elevated unbound plasma fractions of phenytoin in elderly patients, probably due to falling serum albumin levels with advancing age, have been reported elsewhere (Hooper *et al.*, 1974; Hayes *et al.*, 1975; Barth *et al.*, 1976).

Table 1 Interacting drugs and attendant medical conditions in 18 patients with low phenytoin protein binding

Drugs/Conditions	Number of patients
Valproic acid	5
Valproic acid + renal failure	1
Valproic acid + warfarin	1
Warfarin	2
Phenylbutazone	1
Aspirin	1
Aspirin + hypoalbuminaemia	1
Alcoholic liver disease	4
Hypoalbuminaemia	1
Hyperthyroidism	1

The results imply that for most patients measuring total phenytoin concentrations in plasma should provide a reliable index of anticonvulsant effect.

In as many as one in five patients, however, the unbound plasma fraction of phenytoin was elevated so that lower than normal total phenytoin concentrations in plasma produce optimum anticonvulsant effects. Dosage adjustments in these patients should be based on free, rather than total, phenytoin plasma levels.

Measurement of free phenytoin levels is clearly preferable for patients in whom decreased phenytoin binding is expected: the elderly; those with hepatic or renal disease; those taking displacing drugs; and patients with hyperthyroidism or hypoalbuminaemia.

Our data indicate that nearly all patients with decreased phenytoin binding had one or more of these complicating factors, but their predictive value

is not perfect since we found unexpectedly high phenytoin fractions in two other patients. Routine measurement of free drug concentrations is the only completely reliable way to monitor phenytoin therapy.

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