TITLE: IN VITRO METABOLISM OF BW B1090U

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BW B1090U (BW1090), a new short-acting nondepolarizing muscle relaxant, is metabolized by pseudocholinesterase. The duration of neuromuscular blockade at equipotent doses is several-fold longer with BW1090 than with succinylcholine. We were, therefore, interested in determining the rate of metabolism of BW1090 at clinically relevant concentrations in buffer and in human plasma with normal and reduced pseudocholinesterase activity. These data were compared to the in vitro rate of metabolism for succinylcholine.

MATERIALS AND METHODS:

Five (300 ml) units of fresh human plasma were pooled and subdivided for subsequent dilutions. Serial dilutions (1:1, 1:2, 1:4, 1:8, and 1:16) of plasma and phosphate buffer (pH 7.4) were made. Aliquots (5 ml) of the pooled plasma and the various dilutions were assayed for pseudocholinesterase activity and dibucaine number using appropriate standard techniques.

BW1090 (final concentration 9.0 umol L-1 was added to 50 ml of phosphate buffer, to 50 ml of the various plasma dilutions, or to 5 ml phosphate buffer with acetylcholinesterase (EC 3.1.1.7.) (100 units/ml) or butyrlcholinesterase (EC 3.1.1.8) (100 units/ml). The solutions were mixed thoroughly with BW1090 and incubated at 37 C. At timed intervals (0, .5, 1, 1.5, 2, 3, 4, 5, 6, 7.5, 10, 15, 20, 25, 30, 45, and 60 minutes) 1.0 ml aliquots were withdrawn from the 50 ml tubes added to tubes containing 25 ul of a 7% solution of phenylmethylsulfonylfluride in dimethylformamide, a cholinesterase inhibitor, and eventually frozen to -70°C. Smaller aliquots were drawn from the 5 ml tubes. BW1090 concentration was determined at each time by a sensitive, specific high pressure liquid chromatographic assay modified from an atracurium assay. A plot of the logarithm of the BW1090 concentration over time was linear. A linear regression analysis of these data was used to estimate the slope of the line, the half-life (t 1/2, min) and the rate of metabolism of BW1090 (umol min^{-1} L^{-1}).

RESULTS:

The pooled undiluted human plasma had a pseudocholinesterase activity of 2900 Units/L; the activity decreased linearly with the serial dilutions. At physiological pH and temperature, there was no spontaneous hydrolysis of BW1090 in buffer alone over one hour. Acetylcholinesterase added to buffer did not catalyze the hydrolysis of BW1090 but butyrlcholinesterase did. The in vitro rate of BW1090 metabolism in normal plasma was 1.73 umol min^{-1} L^{-1} and the half life was 2.6 min; the rate of BW1090 metabolism decreased and the half-life increased in inverse proportion to the pseudocholinesterase activity (Table).

DISCUSSION:

This study documents that in vitro BW1090 is hydrolyzed by butyrlcholinesterase (pseudocholinesterase), but not by acetylcholinesterase (true cholinesterase); spontaneous hydrolysis of BW1090 appears to be minimal, if at all. The rate of BW1090 metabolism at the concentrations studied is directly related to pseudocholinesterase activity. In contrast, succinylcholine is metabolized by butyrlcholinesterase at a rate of 70-100 umol min^{-1} L^{-1} and undergoes spontaneous hydrolysis at a rate that averages 25 umol min^{-1} L^{-1} (1,2) These markedly different in vitro rates of metabolism for the two relaxants may partially explain the observed differences in duration of neuromuscular blockade. The in vivo situation is complicated for both drugs by distribution of the relaxant from plasma to the interstitial space so that the pharmacokinetic behavior cannot be entirely explained by hydrolysis in plasma. In vivo other mechanisms (eg. metabolism in the liver or renal excretion) may also contribute to the removal of BW1090.

TABLE

BW 1090

Pseudocholinesterase Activity (U/L)	Hydrolysis Rate*	t1/2 (min)
2900	1.73	2.6
1490	.92	4.9
760	0.54	8.3
440	0.31	14.3
180	0.15	31.1
0	0	0

*umol min^{-1} L^{-1}

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