Title: RESISTANCE TO d-TUBOCURARINE IN CATS FOLLOWING 72 HOURS OF CONTINUOUS PARALYSIS

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Introduction. Resistance to nondepolarizing muscle relaxants in man and animals has been reported following upper motoneuron injury,[1] burns[2] and disuse atrophy.[3] In addition, the clinical observation has been made that patients in the ICU setting requiring mechanical ventilation appear to need progressively larger doses of nondepolarizing muscle relaxants over time to permit them to tolerate the ventilator.[4] This study in cats was designed to learn if there was any change in the plasma concentration-response curves of these animals following prolonged paralysis with d-tubocurarine (dTc).

Methods. Five adult male cats were studied. The animals were anesthetized with halothane in oxygen and intubated. An intravenous catheter was inserted the halothane discontinued, and the animal placed on a mechanical ventilator delivering filtered room air. Ventilation was adjusted to maintain an arterial PCO_2 between 30-36 mmHg. Sedation was maintained with iv diazepam, 5 mg at 12 hr intervals; iv ketamine in 25 mg increments to a maximum dose of 175 mg was given to control any movements as the paralysis decreased. A gastric feeding tube and an indwelling bladder catheter were placed. Blood pressure was measured by means of an arteriosound® over the popliteal artery. Body temperature was kept at 37 ± .5° C with a heat lamp and blankets. The nutritional status of the animal was maintained with feedings of kitten milk formula through the gastric tube. Ringer's lactate and Normosol-R were infused to maintain a urine output of 3 ml/kg/hr. Oxacillin was given to prevent pulmonary infection. One of the animal's front paws was firmly attached to a metal board. Neuromuscular transmission was assessed by measuring twitch of the paw with a Grass force-displacement transducer (FT-3). The ulnar nerve was stimulated at the olecranon with supramaximal stimuli from a Grass stimulator (Model S8) in conjunction with a stimulus isolation unit. Responses to single stimuli of 0.15 ms duration delivered at a frequency of 0.1 Hz were recorded. Train-of-four stimulus (TOF) was also recorded, and when T_2 of the TOF was the same height as T_1, it was presumed that the single stimulus had returned to its control value. At this point and at least 1 hour after discontinuance of the halothane, dTc 0.3 mg/kg iv was given. Additional increments of dTc 0.1 mg/kg were given until 95 percent paralysis was obtained. As the twitch returned, heparinized blood samples of 0.25 ml were obtained. The plasma was separated and frozen until analyzed by radioimmunoassay. When twitch had returned to its control value, an iv infusion of dTc was started to maintain continuous paralysis, employing a Harvard infusion pump. The study aimed for 100 percent paralysis of twitch, that is, twitch would be abolished, but a tetanic stimulus (50 Hz) would cause slight movement of the paw. After 72 h of paralysis, the dTc infusion was stopped and once again blood samples were drawn as twitch returned. Plasma concentration-response (percent paralysis) curves before and after 72 h paralysis for each animal were derived from the Hill equation, employing a nonlinear regression program. The resulting sigmoid curves were then used to calculate the expected concentrations of dTc at 5, 25, 50, 75 and 95 percent paralysis. Comparisons between plasma concentrations at the various percent paralysis before and after 72 h of paralysis were made by applying Student's t-test for paired data (two-tailed). Parallelism between the regression lines before and after 72 h paralysis was examined by analysis of covariance.

Results. The data derived from the plasma concentration-response curves before and after 72 h paralysis are presented in Table 1. After 72 h, a significantly higher concentration of dTc was required to produce paralysis in the animal when compared with the dose required initially. The log plasma concentration-response lines between 20 to 80 percent paralysis before and after 72 h paralysis are parallel.

Discussion. This study suggests that prolonged paralysis with dTc caused the animals to become resistant to the relaxant, and provides some evidence to substantiate the earlier clinical observations in man. Any explanation of the cause of this observation is conjectural, but Berg and Hall chronically infused dTc intraperitoneally in rats and found that within three days there was an increased number of extrajunctional acetylcholine (Ach) receptors in the rat's diaphragm.[5] In addition, Fambrough has pointed out that lack of muscle activity leads to the appearance of extrajunctional Ach receptors.[6] Bowman has stated that muscle activity, perhaps by the production of cyclic guanosine monophosphate could suppress extrajunctional Ach receptor formation.[7] Lack of muscle activity would decrease cyclic GMP production and permit cyclic AMP to stimulate the energy-dependent process responsible for synthesis of Ach receptors. The stimulation of production of extrajunctional Ach receptors by blocking neuromuscular transmission postsynaptically appears to be a possible explanation for the observation that an increased plasma concentration of dTc is required to produce paralysis in the cat after prolonged paralysis.

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Table 1. Plasma Concentration-Response Relationships of d-Tubocurarine in Cats (n = 5)

Percent Paralysis	Initial Concentrations µg/ml, Mean±SE	Concentrations after 72 h Paralysis, µg/ml Mean±SE	P
95	2.92 ± .1	7.84 ± .2	<.001
75	2.01 ± .05	4.79 ± .2	<.001
50	1.62 ± .05	3.62 ± .3	<.005
25	1.29 ± .07	2.73 ± .3	<.005
5	.90 ± .07	1.80 ± .2	<.025