The effect of hypokalaemia on a neuromuscular blockade induced by pancuronium and its antagonism by neostigmine was studied in the cat anterior tibialis-peroneal nerve preparation using the constant infusion of pancuronium technique. Hypokalaemia was induced by chronic administration of chlorothiazide. The infusion rate of pancuronium required to maintain a 90% depression of twitch tension was reduced from 0.72 ± 0.06 μg kg⁻¹ min⁻¹ in the cats with a normal serum concentration of potassium (K⁺ = 4.4 ± 0.2 mmol litre⁻¹; n = 7) to 0.41 ± 0.07 μg kg⁻¹ min⁻¹ in the hypokalaemic cats (K⁺ = 2.3 ± 0.1 mmol litre⁻¹; n = 8). The dose of neostigmine necessary for 50% antagonism of the pancuronium-induced depression of twitch tension (ED₅₀) was 10.0 μg kg⁻¹ in the cats with a normal potassium concentration and 18.5 μg kg⁻¹ in hypokalaemic cats. We conclude that hypokalaemia decreases the dose of pancuronium required for neuromuscular blockade and increases the dose of neostigmine required for antagonism of the block.

The ratio of intracellular to extracellular potassium concentration is of prime importance in determining the postjunctional transmembrane potential. An acute decrease in extracellular potassium should hyperpolarize the cell, resist depolarization by acetylcholine and enhance the block from a non-depolarizing myoneural blocking drug such as pancuronium. However, Feldman (1963) and Cohen (1966) have suggested that changes in extracellular potassium concentration are reflected in changes in the intracellular potassium with little resultant change in the resting membrane potential. From their conclusions, one might predict that there would be no effect on the action of muscle relaxant drugs in patients receiving chlorothiazide therapy without potassium supplement. However, since the effect of the loss of potassium on relaxant requirement and neostigmine antagonism has not been studied, the predictions of Feldman (1963) and Cohen (1966) are speculative. Therefore, we examined, in cats, the effects of the loss of potassium, as a result of chlorothiazide administration, on a non-depolarizing neuromuscular blockade and its antagonism by neostigmine.

METHODS

Fifteen cats weighing 1.8–4.4 kg were studied. Seven of the cats received chlorothiazide 20 mg kg⁻¹ per day orally for 12 days before the study. The remaining cats were given potassium chloride, and a potassium-rich diet for 10 days. There was no difference in weight between the cats not receiving and those receiving chlorothiazide both before and after the diuretic therapy; there was no difference in age between the groups. Anaesthesia was induced with chloralose 80 mg kg⁻¹, and urethane 250 mg kg⁻¹, i.p. After performing tracheotomy, ventilation was controlled using a volume-cycled ventilator. PaCO₂ was maintained between 4.3 and 5.3 kPa and arterial pH between 7.35 and 7.43 units. Rectal and muscle temperatures were maintained between 37 and 38 °C by means of a heating blanket and lamp.

The tendon of the anterior tibialis muscle was freed, sectioned near its attachment and connected to a Grass FT.03 force-displacement transducer. The sciatic nerve was sectioned, the distal peroneal nerve was isolated and a supramaximal stimulus (2–10 V) of 0.3 ms duration and 0.1 Hz from a stimulator was applied through shielded platinum electrodes to the isolated nerve. The resulting force of muscle contraction was recorded continuously on a polygraph.

An i.v. bolus injection of pancuronium 20 μg kg⁻¹ was followed by an infusion of pancuronium 10 μg ml⁻¹ from an infusion pump at a rate that depressed twitch tension by a constant 90%. This infusion rate decreased progressively for the next 15–45 min, after which it remained constant, as determined by at least 15 min of observation. Further details of this constant-infusion technique have been described (Miller et al., 1974). While continuing this rate of pancuronium infusion, neostigmine 10, 20, 30 or 70 μg kg⁻¹ was
administered as an i.v. bolus. The resultant maximum antagonism of twitch depression was calculated as a percentage of the pre-existing 90% depression (for example, a peak increase to 40% of the pre-pancuronium twitch would be (40-10/90) or 33% antagonism). In addition, the times from the administration of neostigmine to its peak effect (onset time) and to 50% return to the pancuronium-depressed twitch height (duration of action) were measured. Subsequent doses of neostigmine were given after the twitch had returned to and remained stable at 90% depression for at least 15 min. Usually two doses of neostigmine were studied in each cat. In two cats only one dose was studied. The selection of the dose of neostigmine was randomized. At the end of each study, in those cats not receiving chlorothiazide pretreatment, chlorothiazide 35 mg kg⁻¹, was given i.v. during (n = 4) and in the absence of (n = 4) a partial pancuronium-induced neuromuscular blockade. This was done to determine the effect on the twitch tension of chlorothiazide administered acutely with and without pancuronium.

RESULTS

The infusion rate of pancuronium required to maintain a constant 90% depression of twitch tension was related directly to the serum concentration of potassium (r = 0.72; (P < 0.05) (fig. 1). Although hypokalaemia increased the dose of neostigmine required to antagonize the neuromuscular block (P < 0.05) (fig. 2), hypokalaemia did not prevent the complete antagonism of pancuronium-induced blockade by larger doses of neostigmine (70 µg kg⁻¹). Changes in the potassium concentration did not alter

![Graph](https://example.com/graph1.png)

**Fig. 1.** Relationship between infusion rate of pancuronium necessary to maintain 90% depression of twitch tension and serum potassium concentrations. Each dot represents the infusion rate and potassium concentration from one cat. The line represents analysis of linear regression.

![Graph](https://example.com/graph2.png)

**Fig. 2.** Effect of serum potassium concentration on dose of neostigmine needed to antagonize pancuronium-induced depression of twitch tension. The lines represent analysis of linear regression. The dots and brackets represent mean ± SEM respectively. The numbers in parenthesis represent number of cats studied at each dose.
HYPOKALAEMIA, PANCURONIUM AND NEOSTIGMINE

Fig. 3. Effect of serum potassium concentration on time to peak antagonism of neostigmine action. The dots and brackets represent mean ± SEM respectively. The lines are drawn only to aid visual understanding.

Fig. 4. Effect of serum potassium concentration on duration of action of neostigmine. The dots and brackets represent mean ± SEM respectively. The lines are drawn only to aid visual understanding.

The decrease in the requirement for pancuronium and the parallel shift to the left of the neostigmine dose–response curve in cats receiving chlorothiazide may be a result of hypokalaemia, hypocalcaemia, or a direct neuromuscular depressant effect of chlorothiazide itself. The importance of hypokalaemia is suggested by the direct relationship between plasma potassium concentrations and the infusion rate of pancuronium required to maintain a 90% depression of twitch tension. Although plasma calcium concentrations were reduced, no correlation existed between the required infusion rates of pancuronium and the calcium concentrations. Unfortunately we could not measure ionized calcium concentrations which may have been decreased although the pH was the same between the two groups. We were unable to detect any direct depressant effect of chlorothiazide on twitch tension or a partial pancuronium neuromuscular block. The state of hydration was comparable in both groups of cats as shown by similar plasma sodium concentrations and haematocrit values.

One might speculate that chlorothiazide administration of longer duration than that used in this study would not affect a pancuronium neuromuscular block and its antagonism by neostigmine as much as we observed. Using the reasoning of Feldman (1963) and Cohen (1966), the transmembrane potential may become normal in spite of low potassium concentrations in both the extra- and intracellular compartments. Although a 200–400 mmol litre\(^{-1}\) deficit of total body potassium may be indicated by a serum potassium concentration of less than 3.0 mmol litre\(^{-1}\) (Huth, Squires and Elkington, 1959), the serum concentration provides no information about the transmembrane potential. Although sagging ST segments, depression of T waves and elevation of U waves in the e.c.g. indicate abnormal transmembrane potentials of cardiac muscle, this may not be a reflection of skeletal muscle potentials. Certainly, prolonged chlorothiazide therapy will result in continued loss of potassium. Since skeletal muscle transmembrane potentials cannot be determined clinically, we believe it is reasonable to conclude that patients with hypokalaemia from chlorothiazide therapy may require less pancuronium to produce

Chlorothiazide 35 mg kg\(^{-1}\) i.v. did not affect twitch tension without pancuronium or during a 40–60% neuromuscular blockade induced with pancuronium.

DISCUSSION

The onset time and duration of action of neostigmine (figs 3 and 4).

Serum sodium, calcium and chloride concentrations were 152 ± 1.4 (SEM), 7.9 ± 0.4 and 118 ± 0.8 mmol litre\(^{-1}\) in the cats pretreated with chlorothiazide and 149 ± 1.7, 8.9 ± 0.6 and 116 ± 1.5 mmol litre\(^{-1}\) in the untreated cats. Although serum calcium concentrations were decreased in the chlorothiazide-treated cats, the decrease was not significant and did not correlate with the required pancuronium infusion rates (\(r = 0.15\)). The haematocrit was 45 ± 1 in cats not receiving and 47 ± 2 in cats receiving chlorothiazide.
neuromuscular blockade and that more neostigmine will be required to antagonize the block. Since chlorothiazide itself does not affect pancuronium and neostigmine, we believe also that it is reasonable to apply the results of this study to hypokalaemic states from other causes.

ACKNOWLEDGEMENTS
This study was supported by USPHS Grant 1 PO1 GM15571–10.

The authors are very grateful to Edmond I. Eger II, M.D., and Dorothy M. Urban for their advice in the preparation of this manuscript.

REFERENCES

HIPOPOTASSEMIA DE INDUCCION DIURETICA, BLOQUEO NEUROMUSCULAR DE PANCURONIO Y SU ANTAGONISMO MEDIANTE NEOESTIGMINA

On a etudié, sur une préparation de nerf tibial antérieur et de nerf péronier prélevés sur un chat, l'effet de l'hypokaliémie sur un blocage neuromusculaire provoqué par le pancuronium et son antagonisme par la néostigmine, à l'aide d'une technique basée sur l'infusion constante de pancuronium. L'hypokaliémie a été provoquée par l'administration chronique de chlorothiazide. Le taux d'infusion du pancuronium nécessaire pour maintenir une dépression de 90% de la tension de la crispation a été abaissée de 0,72 ± 0,06 μg kg⁻¹ mn⁻¹ sur les chats ayant une concentration normale de potassium dans le sèrum (K = 4,4 ± 0,2 mmol litre⁻¹; N = 7) à 0,41 ± 0,07 μg kg⁻¹ min⁻¹ sur les chats hypokaliémiques (K = 2,3 ± 0,1 mmol litre⁻¹; N = 8). La dose de néostigmine qu'il a fallu pour obtenir un antagonisme de 50% de la dépression de la tension de la crispation (ED₅₀) provoquée par le pancuronium a été de 10 μg kg⁻¹ sur les chats ayant une concentration normale de potassium et de 18,5 μg kg⁻¹ sur les chats hypokaliémiques. Nous en concluons que l'hypokaliémie réduit la dose de pancuronium qu'il faut pour obtenir le blocage neuromusculaire et augmente la dose de néostigmine qu'il faut pour arriver à l'antagonisme du blocage.

SUMARIO
Se estudió el efecto de hipopotasemia sobre un bloqueo neuromuscular inducido por pancuronio y su antagonismo por neostigmina en la preparación de nervio tibial-peroneal, empleando la técnica de la constante infusión de pancuronio. La hipopotasemia fue inducida mediante la crónica administración de clorotiacida. La rapidez de infusión del pancuronio necesaria para mantener la depresión de 90% de la tensión de contracción se redujo de 0,72 ± 0,06 μg/kg/min en los gatos con una concentración de potasio en el suero normal (K = 4,4 ± 0,2 mmol/litro; N = 7) a 0,41 ± 0,07 μg/kg/min en los gatos hipopotasémicos (K = 2,3 ± 0,1 mmol/litro; N = 8). La dosis de neostigmina necesaria para el antagonismo de 50% de la depresión inducida por pancuronio en la tensión de contracción (ED₅₀) fue de 10,0 μg/kg en los gatos con una concentración de potasio normal y de 18,5 μg/kg en los gatos hipopotasémicos. Concluimos que la hipopotasemia disminuye la dosis de pancuronio necesaria para el bloqueo neuromuscular y aumenta la dosis de neostigmina necesaria para el antagonismo del bloqueo.