ACUTE AND SUBCHRONIC NEUROMUSCULAR BLOCKING CHARACTERISTICS OF STREPTOMYCIN: A COMPARISON WITH NEOMYCIN*

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SUMMARY

The characteristics of the neuromuscular block produced by streptomycin in vivo were studied on the sciatic- anterior nerve-muscle preparation of eight anaesthetized cats. The lungs of the animals were ventilated mechanically and normocarbia was maintained. During acute exposure to streptomycin (within 2 h), ED_{50} for blockade of the twitch was 56 (SEM ± 5) mg kg^{-1} of the base. The characteristics of block were similar to those of neomycin-induced block in some aspects. There was absence of train-of-four fade and tetanic fade, partial sparing of the responses elicited at 10 Hz and 20 Hz, and total sparing of the 50 Hz tetanus, as well as the post-tetanic twitch. In contrast to neomycin-induced neuromuscular block, however, post-tetanic exhaustion was not observed and prolonged exposure to streptomycin (22-28 h) did not change the characteristics of the block. We conclude that, despite their chemical similarities, streptomycin and neomycin block neuromuscular transmission differently.

The unique characteristics of the neuromuscular block produced by neomycin have been described previously (Lee et al., 1976) and the difference between the block induced, in vivo, by the aminoglycoside antibiotics and the polypeptide antibiotics has been stressed (Lee et al., 1976; Lee, Chen and Nagel, 1977). Recently Wright and Collier (1977) reported differences in vitro between neomycin and streptomycin, the two most important aminoglycosides with neuromuscular-blocking properties, in their predilection for the prejunctional and the post-junctional structures. Thus, not only dissimilar but also similar antibiotics may block neuromuscular transmission by inherently different mechanisms. The purpose of this present study was to determine whether the characteristics of the neuromuscular block produced by streptomycin differed from those of neomycin in vivo also.

METHODS

The methods used by Flacke (1972) and by ourselves (Lee et al., 1976; Lee, Chen and Nagel, 1977) in our previous studies on antibiotics have been combined and modified. Eight healthy cats of 2.0-3.2 kg (mean 2.4 kg; SD 0.4) were anaesthetized with an intraperitoneal injection of alpha-chloralose 60 mg kg^{-1} and pentobarbitone 10 mg kg^{-1}. Tracheotomy was performed and mechanical ventilation was instituted and adjusted to produce normocarbia. The sciatic-tibialis anterior nerve-muscle preparation, which has been described previously (Lee et al., 1976) was used. The sciatic nerve was stimulated at 0.1 Hz (twitch). This was interrupted only for the application of other modes of stimulation as follows: 2 Hz for 2 s (train-of-four), 10 Hz for 2 s, 20 Hz for 2 s (partial tetanization), 50 Hz for 5 s and 100 Hz for 1 s (tetanus). The stimulus was 0.1 ms square electric pulse of supramaximal voltage generated by a Grass S88 stimulator. The presence or absence of tetanic fade was determined by the 50-Hz tetanus. The first post-tetanic twitch was elicited 10 s after the end of the 50-Hz tetanus. Subsequent post-tetanic twitches were followed to determine the time course of facilitation and possible exhaustion. Following the determination of baseline (control) values streptomycin sulphate, dissolved in 0.89% saline, was injected i.v. To determine the dose required, appropriate bolus doses of streptomycin were injected until an almost total block of the twitch was established. Cumulative ED_{50} and ED_{95} were determined. Spontaneous recovery was timed. Following this, a 50% block of the twitch was established repeatedly by additional bolus injections of streptomycin, and the effects on
the various modes of stimulation observed. After each tetanus, recovery of the twitch from the post-tetanic facilitation was awaited before the response to another mode of stimulation was examined. On completion of these studies, reversal of the block was tested by the injection of edrophonium chloride 0.2 mg kg\(^{-1}\) to the cat at a point of 80% block of the twitch. The effect of calcium chloride 10 mg kg\(^{-1}\) was examined similarly 1 h later. The cat was rested until the next morning. Overnight, an additional amount of streptomycin sulphate was infused into three cats over a period of 5–8 h to increase the total exposure to the drug. Next morning, the acute experiment was repeated, with the exception that 4-aminopyridine 0.6 mg kg\(^{-1}\) was used instead of edrophonium or calcium chloride. For general support, each cat received an infusion of lactated Ringer’s solution with 5% dextrose at the rate of 20 ml kg\(^{-1}\) during the initial preparation and period of stabilization, 20 ml kg\(^{-1}\) during the acute session, 30 ml kg\(^{-1}\) overnight, and another 20 ml kg\(^{-1}\) during the subchronic session on the 2nd day. For maintenance of anaesthesia, each cat received an additional 70 mg kg\(^{-1}\) of alpha-chloralose overnight. Throughout the experiment the body temperature of the cat was maintained within 1 °C of its normal value (37–38.5 °C) by a warming mattress and a heating lamp.

RESULTS

The total amount of streptomycin base (mean ± SEM) given to each cat was 194 ± 18 mg kg\(^{-1}\) during the acute session and 142 ± 12 mg kg\(^{-1}\) during the 2nd day. Three cats received additional 178 mg kg\(^{-1}\), 246 mg kg\(^{-1}\) and 865 mg kg\(^{-1}\), respectively overnight. During the acute session ED\(_{50}\) for the twitch was 56 ± 5 mg kg\(^{-1}\), and ED\(_{95}\) was 88 ± 8 mg kg\(^{-1}\). The neuromuscular block was of immediate onset, each incremental dose reaching its full effect in 2–3 min. The time to 50% recovery (from 75% block to 25%) was 5.2 ± 0.5 min. Full recovery from 80% block required 15–20 min. On the 2nd day, ED\(_{50}\) and ED\(_{95}\) were 43 ± 4 mg kg\(^{-1}\) and 72 ± 7 mg kg\(^{-1}\) of the base, respectively. Both values represented a slight but statistically significant increase in sensitivity following prolonged exposure (\(P<0.02\)). The 50% recovery time was 5.9 ± 0.8 min, a value not significantly different from that of the 1st day (\(P>0.2\)).

In the presence of a 50% block of the twitch, train-of-four twitches and tetanus did not fade. The post-tetanic twitch was augmented markedly. There was no secondary post-tetanic exhaustion. Muscle responses evoked by the trains of stimuli with a stimulus frequency greater than 0.1 Hz retained more than 50% of their respective controls. The greater the stimulus frequency, the more was the response spared from block (fig. 1). The 50-Hz and the 100-Hz tetanic responses equalled or exceeded their respective controls. The post-tetanic twitch was spared also. Sparing of these responses from block in the presence of a diminished twitch resulted in more than doubling of the tetanus (50 Hz) to twitch and the post- to pre-tetanic twitch ratios during block. These ratios were 10.1 ± 0.8 and 4.3 ± 0.2 respectively during a 50% block, as opposed to 4.3 ± 0.4 and 1.9 ± 0.4 respectively, at control. Following prolonged exposure, there continued to be no train-of-four or tetanic fade. The tetanus to twitch ratio and other indicators of the nature of block remained virtually unchanged (fig. 1).

FIG. 1. Characteristics of neuromuscular block during acute and subchronic exposure to streptomycin in the cat. Height of columns represents the relative force of contraction of the twitch or of the tetanus at its peak. “Acute” session was within 1–2 h, “subchronic” after 22–28 h of exposure to streptomycin in doses sufficient to produce neuromuscular blockade. “Train-of-four” refers to the fourth of a train of tetanic responses evoked at 2 Hz. Note that 10-Hz and 20-Hz responses were relatively spared, while 50-Hz and 100-Hz responses were totally spared, despite 50% block of the single twitch. The fourth twitch of a train-of-four equalled the single or its first twitch and the 50-Hz tetanus maintained its initial force at 5 s, without fade. The characteristics of block remained essentially unchanged after prolonged exposure.
Edrophonium chloride 0.2 mg kg\(^{-1}\) and calcium chloride 10 mg kg\(^{-1}\), tested during the 1st day, increased the twitch from 20\% of control (80\% block) to 69 \pm 4\% and 70 \pm 3\% of control, respectively. 4-Aminopyridine 0.6 mg kg\(^{-1}\), tested on the 2nd day, restored the twitch to control within 8 \pm 2 min of injection. The reversal was followed by a prolonged overshoot of the twitch response. At its peak, which occurred 71 \pm 15 min after the injection of 4-aminopyridine, the twitch response averaged 138 \pm 10\% of control.

The mean arterial pressure, which was 99 \pm 10 mm Hg before the injection of streptomycin, decreased to 87 \pm 6 mm Hg 2 min after the injection of the first 30 mg kg\(^{-1}\) of streptomycin. The mean arterial pressure, which averaged 93 \pm 4 mm Hg immediately before the injection of 4-aminopyridine, increased to 118 \pm 8 mm Hg 5 min after administration.

**DISCUSSION**

The present study revealed the similarities and the dissimilarities between the neuromuscular blocking characteristics of streptomycin and those of neomycin. The similarities were the absence of fasciculation, the absence of train-of-four and tetanic fade, and the sparing of the partial and the full tetanus in the presence of depression of the twitch. These features of the block produced by aminoglycosides were obviously not like those of tubocurarine. The dissimilarities were twofold and were both qualitative. With neomycin-induced neuromuscular block a brief post-tetanic facilitation was followed quickly by exhaustion of the twitch (Lee et al., 1976), whereas this was not shared by streptomycin. Another difference was the absence of the time-dependent changes in the nature of block following prolonged exposure to streptomycin. With neomycin, Flacke (1972) observed an alteration of the block to a "curare-like" pattern after 24 h of subchronic exposure (130–350 mg kg\(^{-1}\)).

A study of this type enables proper delineation of each type of neuromuscular block, for its own interest and for the use of such knowledge in the differential diagnosis of clinical problems. It permits the matching and interpretation of the various signs of block observed in vivo with the various mechanisms of action observed in vitro. In vitro, neomycin decreases predominantly the influx of the calcium ion into the motor nerve terminal, which in turn decreases the output of acetylcholine. By comparison, streptomycin is more effective in reducing the muscle motor endplate response to acetylcholine, although it decreases the output of the transmitter (Wright and Collier, 1977). The common features of streptomycin- and neomycin-induced neuromuscular block can be attributed plausibly to their well-known "magnesium-like" anti-release activity (Vital Brazil and Prado-Franceschi, 1969; Pittinger and Adamson, 1972). Both high stimulus frequency and post-tetanic facilitation tend to overcome this type of block (del Castillo and Katz, 1954). The differing features are more difficult to explain. Presumably neomycin has a transmitter-depleting effect. With pre-existing depletion, the post-tetanic state of facilitated release may deplete further the transmitter supply, so that secondary exhaustion follows initial facilitation. The time-dependent and changing nature of neomycin-induced neuromuscular block has been attributed also to its prejunctural effect (Flacke, 1972). With streptomycin, it appears that neither tetanic stimulation nor prolonged exposure results in a depletion of transmitter of comparable magnitude.

In conclusion, streptomycin-induced neuromuscular blockade which have clinical relevance could be divided into depolarizing and non-depolarizing categories. Recently, we have realized that not all non-depolarizing agents block similarly or cause a fade (Lee, Chen and Katz, 1977); neither do all antibiotics, all aminoglycosides or all prejunctional blockers (personal observations) block with similar characteristics. In the future, improved techniques of examination may reduce further the gap between the diversity of knowledge of the mechanism of action of various neuromuscular blocking drugs and what can be demonstrated in vivo. For example, we have observed that, while alpha-bungarotoxin neuromuscular block (post-junctional) has no effect on the refactoriness of neuromuscular transmission, beta-bungarotoxin (pre-junctional) reduced it markedly. In this respect, polymyxin B behaves like alpha-bungarotoxin. Streptomycin behaves like beta-bungarotoxin. Neomycin resembles beta-bungarotoxin except that the post-tetanic facilitation reduces the refractoriness while the secondary post-tetanic exhaustion enhances the effect further. We feel that this observation might help to confirm the pre- or post-junctional mechanism of action of these neuromuscular blocking antibiotics in vivo and eventually provide a method of differential diagnosis which would have clinical applicability.

In conclusion, streptomycin-induced neuromuscular block in the cat is characterized by the absence of fade and a selective sparing of the tetanus and...
the post-tetanic twitch. In these respects, it is similar to the block produced by neomycin, another aminoglycoside antibiotic. However, during block the post-tetanic facilitation was not followed by a secondary post-tetanic exhaustion and there were no time-dependent changes in the characteristics of the block following prolonged exposure. Based on these qualitative differences, we conclude that even antibiotics of similar chemical structure may block neuromuscular transmission differently. Prejunctional effects appear to play a less important role in a block induced by streptomycin as compared with neomycin.

REFERENCES


CARACTERISTIQUES DU BLOCAGE NEUROMUSCULAIRE AIGU ET SOUS-CHRONIQUE PAR LA STREPTOMYCINE: COMPARAISON AVEC LA NEOMYCINE

RESUME

Les caracteristiques du blocage neuromusculaire produit par la streptomycine in vivo ont ete etudies sur une preparation de nerf-muscle sciatique-tibial anterieur de huit chats anesthesies. Les poumons des animaux etaient ventilies par des moyens mecaniques et la normocarbie a ete maintenue. Pendant l'exposition aigue a la streptomycine (et en l'espace de 2 h) l'EDs60 pour le blocage de la crispation a ete de 56 mg kg-1 de la base (erreur type des moyennes ± 5). Les caracteristiques du blocage ont ete similaires sous certains aspects a celles d'un blocage provoque par la neomycine. Il y a eu une absence d'attenuation de la serie de quatre crispations et d'attenuation tetanique, une moderation partielle des reactions decouvertes a 10 Hz et a 20 Hz et une moderation totale du tetan a 50 Hz de meme que de la crispation post-tetanique. Par contraste au blocage neuromusculaire provoque par la neomycine, l'epuisement post-tetanique n'a cependant pas ete observe et l'exposition prolongee a la streptomycine (22 a 28 h) n'a pas modifie les caracteristiques du blocage. Nous en concluons qu'en depot de leurs similarites chimiques, la streptomycine et la neomycine bloquent la transmission neuromusculaire de manieres differentes.

AKUTE UND SUBCHRONISCHE NEUROMUSKULARE BLOCKIERUNGSCARACTERISTIKEN VON STREPTOMYZIN: EIN VERGLEICH MIT NEOMYZIN

ZUSAMMENFASSUNG


CARACTERISTICAS DE BLOQUEO NEUROMUSCULAR AGUDO Y SUBCRONICO DE ESTREPTOMICINA: COMPARACION CON NEOMICINA

SUMARIO

Se estudiaron las características del bloqueo neuromuscular producido por estreptomicina in vivo en una preparación de nervio-músculo ciático-tibial anterior de ocho gatos anestesiados. Los pulmones de los animales se ventilaron mecánicamente y se mantuvo normocarbia. Durante la exposición aguda a la estreptomicina (dentro de 2 h), el EDs60 para el bloqueo de sacudida muscular fue de 56 (SEM ± 5) mg kg-1 de la base. Las características del bloqueo fueron semejantes en ciertos aspectos al bloqueo inducido por neomicina. Hubo ausencia de debilitamiento de "serie de cuatro" y debilitamiento tetánico, conservación parcial de las reacciones producidas a 10 Hz y 20 Hz, y conservación total del tetán de 50 Hz, además de la sacudida post-tetánica. Sin embargo, en contraste con el bloqueo neuromuscular inducido por neomicina, no se observó agotamiento post-tetánico y la prolongada exposición a estreptomicina (22-28 h) no cambió las características del bloqueo. Concluímos que, a pesar de su semejanza química, la estreptomicina y la neomicina bloquean la transmisión neuromuscular de forma diferente.