JPP JOURNAL OF Pharmacy and Pharmacology
ROYAL PHARMACEUTICAL SOCIETY
JPP Journal of Pharmacy And Pharmacology
Research Paper

Neamine and 2-deoxystreptamine neomycin derivatives exhibit antinociceptive activity in rat models of phasic, incision and neuropathic pain

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Keywords
2-deoxystreptamine; aminoglycoside antibiotic; incision pain; neamine; neuropathic pain

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Received November 10, 2014
Accepted July 19, 2015

doi: 10.1111/jphp.12480

Abstract

Objectives To assess the antinociceptive activity of the neomycin derivatives neamine and 2-deoxystreptamine following intraspinal administration in rats.
Methods We used the tail-flick test and measured the threshold to mechanical stimulation in models of incisional and neuropathic pain.
Key findings The derivatives produced antinociception in the tail-flick test and reduced mechanical allodynia in models of incisional and neuropathic pain. The approximate ED_{50} in milligrams (confidence limits in parenthesis) in these tests were 1.35 mg (0.61; 2.95), 0.20 mg (0.14; 0.27) and 0.28 mg (0.12; 0.63) for neamine, and 1.05 mg (0.68; 1.60), 0.78 mg (0.776; 0.783) and 0.79 mg (0.46; 1.34) for 2-deoxystreptamine, respectively. Neamine was more potent than 2-deoxystreptamine in the incisional and neuropathic pain models, but they had similar potency in the tail-flick test. Tetra-azidoneamine, a neamine derivative in which free amino groups are replaced with azido groups, did not change the incisional mechanical allodynia. The reduction of incisional allodynia by neamine and 2-deoxystreptamine was transitorily antagonized by intrathecal administration of calcium chloride.
Conclusions The intraspinal administration of neamine and 2-deoxystreptamine is antinociceptive in rats. The presence of amino groups in the structure of these derivatives is fundamental to their antinociceptive effect, which may be due to a calcium antagonist activity.

Introduction

Aminoglycoside antibiotics, such as gentamicin, streptomycin, neomycin and kanamycin, produce antinociception in rodent models of experimental pain following intraperitoneal,[1] intracerebroventricular[2,3] or intraspinal administration.[3] The antinociceptive effect of intraspinal gentamicin was antagonized by intraspinal administration of calcium chloride,[3] thus leading to the idea that the effect of gentamicin depends on its calcium-antagonist property. Gentamicin and other aminoglycoside antibiotics act as antagonists of N-type calcium channels[4,5] and compete with calcium in several biological processes.[6] However, the chronic utilization of these compounds is frequently followed by oto-[7] and nephrotoxicity.[8] Moreover, the fast increase of the plasmatic concentration of aminoglycoside antibiotic produces a severe decline in blood pressure accompanied by neuromuscular blockade, both of which are competitively antagonized by calcium.[9,10]

Streptidine, an aminocyclitol derivative from streptomycin, produces a neuromuscular blockade with the same characteristics as the entire molecule of the antibiotic,[11] including a competitive interaction with calcium.[12] More recently, it was shown that neamine, but not 2-deoxystreptamine (aminoglycoside-aminocyclitol and aminocyclitol derivatives of neomycin, respectively) (Figure 1), retains the antibacterial and ototoxic properties of the entire antibiotic molecule.[13]

Discovery of new pain relieving drugs is necessary since many patients still suffer from pain mainly because the

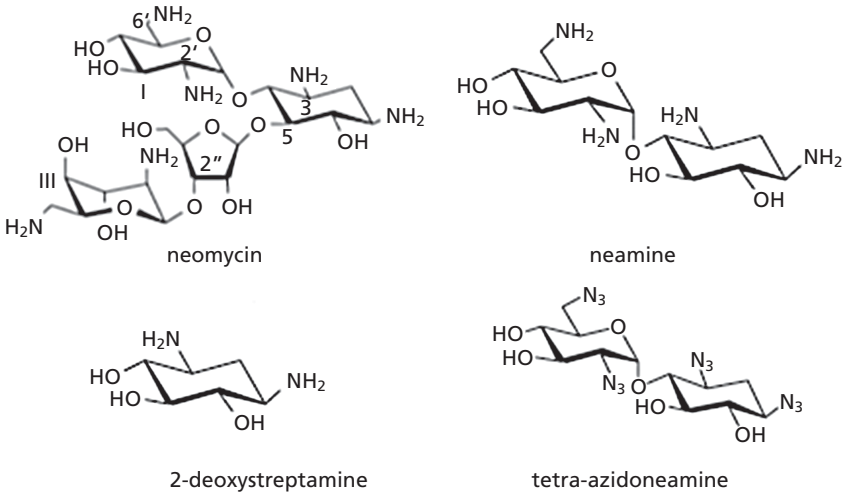

Figure 1 Chemical structures of neomycin and its derivatives neamine, 2-deoxystreptamine and tetra-azidoneamine.

available drugs are effective but limited by side effects.[14] Here, we focused on the potential of neomycin derivatives as new analgesic drugs. It is currently accepted that establishing efficacy across a variety of models reduces translational risk associated with the target mechanism, and variability in potency or maximal effects can help inform both clinical trial design and patient population selection.[15] By this reason, this study examines comparatively whether neamine and 2-deoxystreptamine exhibit antinociceptive activity on three models of pain in rats. The rat tail-flick test, incision pain and neuropathic pain were used to determine the antinociceptive effects of the neomycin derivatives against acute, post-surgical and neuropathic pain, respectively. These methods are used widespread and are essential to progress in pain research.[16] The efficacy of the derivatives in each test was established. They were injected intrathecally because neomycin[17] and gentamicin[3] were more effective in rats by intrathecal than by intracerebroventricular and intraperitoneal routes. The changes in incision pain induced by tetra-azidoneamine, a neamine derivative in which the amino groups were replaced by azido groups (Figure 1), were also evaluated. The effect of intrathecal administration of calcium chloride against the antinociceptive activity of neamine and 2-deoxystreptamine in the incision pain was evaluated as well.

Materials and Methods

Animals

Male Wistar rats (140–160 g) from the main animal house of the Ribeirão Preto Campus of the University of São Paulo were maintained at a controlled temperature (23 ± 1ºC) with free access to food and water. The experiments were approved by the institutional ethics committee on animal research (No. 116/2012) and followed the guidelines of the Committee for Research and Ethical Issues of the International Association for the Study of Pain.[18]

Tail-flick test

Each animal was placed in a ventilated tube with the ventral surface of the tail between 4 and 6 cm from the tip laid across a wire coil maintained at room temperature (23ºC ± 2ºC). The coil temperature was then raised by the passage of electric current through it, and the tail-flick latency (TFL) was measured. Each trial was terminated after 6 s to minimize the probability of skin damage. TFL was measured at 5-min intervals until a stable baseline was obtained over three consecutive trials, 10 min after intrathecal injection and then at 10-min intervals for up to 60 min.

Incision pain test

Each animal was anaesthetized with isoflurane (2% for induction and 0.5% for maintenance) in oxygen flow via a loose-fitting, cone-shaped mask. A 1-cm longitudinal incision was made with a surgical blade through the skin and fascia of the plantar aspect of the right hind paw starting 0.5 cm from the proximal edge of the heel, as described elsewhere.[19] The wound was then closed with two 5-0 nylon sutures, and the animal allowed to recover in the home cage until the beginning of the experiment.

Chronic constriction injury model

Each animal was anaesthetized as above and subjected to chronic constriction injury (CCI) of the sciatic nerve as proposed elsewhere.[20] A single ligature of 100% cotton glace thread (Coats Corrente Ltda., São Paulo, Brazil) was placed around the sciatic nerve proximal to its trifurcation. The incision was then closed with silk sutures, and the animal was kept in recovery for 8–10 days.

Mechanical stimulation

Rats with hind paw incision or CCI were placed in a cage with a nylon mesh bottom, which allowed easy access to the paw plantar surface. The threshold for mechanical stimulation was measured with an electronic apparatus (IITC Electronic Equipment, Woodland Hills, CA, USA) consisting of a hand-held probe unit to which a rigid plastic tip (tip area 0.44 mm2) was connected. The plastic tip was applied with increasing force against the central area of each hind paw of CCI animals or sites near the heel, 1–2 mm adjacent to the medial border of the wound in the incised hind paw. The movement of the probe was stopped when withdrawal of the stimulated paw occurred. A single trial consisted of three applications of the tip, once every 5 s, to each hind paw. The mean of three readings was taken as the threshold for a particular time. The threshold was determined immediately before surgery (baseline) and 2 h after hind paw incision or 8–10 days after CCI. The threshold was measured again 10 min after intraspinal injection and then at 10-min intervals for up to 80 min. The mechanical threshold in non-operated hind paws was measured as well.

Intrathecal injection

The injections were performed in rats lightly anaesthetized as described above. A 1-inch, 25-G needle was transcutaneously introduced at the L5–L6 level into the subarachnoid space as described elsewhere.[21] Each rat received only one intrathecal injection (constant 5-μl volume) using this method. The animal was submitted to the algesimetric test 10 min later.

When two injections per rat were necessary, each rat was catheterized via a lumbar puncture as described elsewhere.[22] A 20-gauge Weiss needle was introduced through/the skin into the L5–L6 intervertebral space. A 12-mm length of polyethylene tubing (o.d. = 0.4 mm, dead space =10 μl) was introduced through the needle to protrude 1.5 cm into the subarachnoid space in the cranial direction. The needle was removed, and the tubing anchored to the back skin with a cotton thread suture. A surgical incision was then performed in the right hind paw as described above. The experiment was carried out 2 h later. All solutions contained 1% fast green dye to confirm the correct position of the catheter.

Statistical analysis

TFL (in seconds) and mechanical threshold (in grams) are reported as the mean ± SD. Comparisons between control (rats given saline intrathecally) and test groups were made by multivariate analysis of variance with repeated measures. The factors analysed included treatment, time and treatment × time interaction. In the case of the treatment × time interaction, one-way analysis of variance followed by the Bonferroni correction was performed for each time point.

The approximate dose producing antinociception in 50% of the rats in an experimental group (ED_{50}) and 95% confidence limits were calculated as proposed by Litchfield and Wilcoxon.[23] The Litchfield–Wilcoxon method was chosen because it allows the calculation of approximate ED_{50} using a small number of animals.[24] Antinociception in the tail-flick test was defined whenever TFL > baseline TFL + (5 × SD). Antinociception in the tests using mechanical stimulation was defined whenever the mechanical threshold was at least 50% of the baseline. The regression lines (not shown in the figures) for neamine and 2-deoxystreptamine in each model were compared with regard to the slope function ratio and corresponding factors as suggested elsewhere.[23] The potency ratio of neamine and 2-deoxystreptamine in each model was also calculated whenever the regression lines did not deviate significantly from parallelism.

Preparation of neamine

Neomycin trisulfate (1.0 g, 1.10 mmol) was dissolved in a saturated solution of methanol (130 ml) and HCl (pH < 1), and heated at 80°C for 3 h. After cooling to 0°C, the reaction was treated with ethyl ether (40 ml) to precipitate a white flocculent solid, which was filtered and washed with ethyl ether. The solid was dried under reduced pressure, and the resulting crude product was purified as described elsewhere.[25] The solid was dissolved in methanol (200 mg/10 ml) followed by titration with ethyl ether until start to precipitate a white insoluble fraction, which was removed in glass filter. This procedure was repeated until removal of impurity. Purity of the solution was monitored by TLC (methanol-ethyl acetate 2 : 1). The solution was evaporated to yield the product in 67%. ^{1}H NMR (400 MHz, D_2O): 1.14 (1H, q, *J* 12.8 Hz, H-2'ax); 1.91 (1H, dt, *J* 4.0; 12.8 Hz, H-2'eq); 2.66 (1H, ddd, *J* 4.0; 9.8; 12.8 Hz, H-1′); 2.71–2.84 (3H, m, H-2. H-3′, H-6a); 2.98 (1H, dd, *J* 2.7; 13.6 Hz, H-6b); 3.09 (1H, t, *J* 9.8 Hz, H-6′); 3.19–3.28 (2H, m, H-4, H-4′); 3.44 (1H, t, *J* 9.8 Hz, H-5′); 3.51 (1H, t, *J* 9.8 Hz, H-3); 3.72 (1H, ddd, *J* 2.7; 7.3; 9.8 Hz, H-5); 5.23 (1H, d, *J* 3.7 Hz, H-1).

Preparation of 2-deoxystreptamine

A solution of neamine chloride (0.155 g) in aqueous hydrobromic acid (HBr 48%, 3.2 ml) was heated at 130°C for 18 h. The solution became dark during this period. The mixture was then allowed to cool down to room temperature, evaporated under reduced pressure and repeatedly treated with water and evaporated three times to remove acid excess. The residue was dissolved in warm methanol (3.2 ml), filtered, and treated with activated charcoal for

3 min and filtered again. Finally, the system was filtered and washed with water. The obtained solution was dried under reduced pressure, yielding 2-deoxystreptamine (98%). ^{1}H NMR (400 MHz, D_2O): 1.66 (1H, q, *J* 12.3 Hz, H-2ax); 2.31 (1H, dt, *J* 4.2; 12.3 Hz, H-2eq); 3.14–3.22 (2H, m, H-1, H-3); 3.28 (1H, t, *J* 9.3 Hz, H-5); 3.39 (2H, t, *J* 9.3 Hz, H-6, H-4).

Preparation of tetra-azidoneamine

A solution of neamine chloride (0.15 g, 0.35 mmol) in water (6 ml) was treated with sodium carbonate (0.38 mg, 2.3 mmol) then copper sulfate (2.8 mg, 0.02 mmol) and methanol (2.0 ml). Finally, trifluoromethanesulfonyl azide solution (prepared procedure described below) was added. Then, methanol was added to make a homogenous solution, which was stirred at room temperature for 24 h. After that, solvents were evaporated and residue partitioned in ethyl acetate and water. To eliminate copper salts, the organic layer was washed with EDTA solution (10%). Organic layer was dried under $MgSO_4$, filtered and evaporated, yielding neamine tetra-azide in 71%.

Preparation of trifluoromethanesulfonyl azide (TfN_3)

A solution of sodium azide (1.20 g, 0.18 mmol) in water (3 ml) was cooled to 0°C, and dichloromethane (5 ml) was added, followed by dropwise addition of trifluoromethanesulfonic anhydride (Tf_2O) (600 µL) during 5 min. After 2 h, the organic layer was separated and aqueous layer extracted with dichloromethane (5 ml × 2). Organic layers were combined and washed with sodium carbonate solution (Na_2CO_3). ^{1}H NMR (400 MHz, DMSO-d_6/D_2O): δ 1.40 (1H, q, *J* 12.3 Hz, H-2'ax); 2.29 (1H, dt, *J* 4.0; 12.3 Hz, H-2'eq); 3.21 (1H, dd, *J* 3.7; 10.6 Hz, H-2); 3.24–3.35 (1H, m, H-1′, H-3′); 3.45–3.62 (6H, m, H-6a, H-6b, H-4′, H-4, H-6′, H-5′); 3.79 (1H, dd, *J* 8.8; 10.6 Hz, H-3); 4.08 (1H, ddd, *J* 2.7; 6.3; 8.8 Hz, H-5), 5.66 (1H, d, *J* 3.7 Hz, H-1).

Results

The antinociceptive effects of intrathecal injection of neamine and 2-deoxystreptamine on the tail-flick test

The time course of the changes induced in the TFL by intrathecal neamine (0.2–0.8 mg) and 2-deoxystreptamine (0.5–2.0 mg) is shown in Figure 2a and 2b, respectively. The effects of the smaller doses of each derivative did not differ from the effect of saline (10 µl). However, higher doses produced a significant increase in the TFL compared with the effect of saline. The peak effect was reached 30–40 min after administration of neamine and 20 min after administration of 2-deoxystreptamine. The duration of the effect of

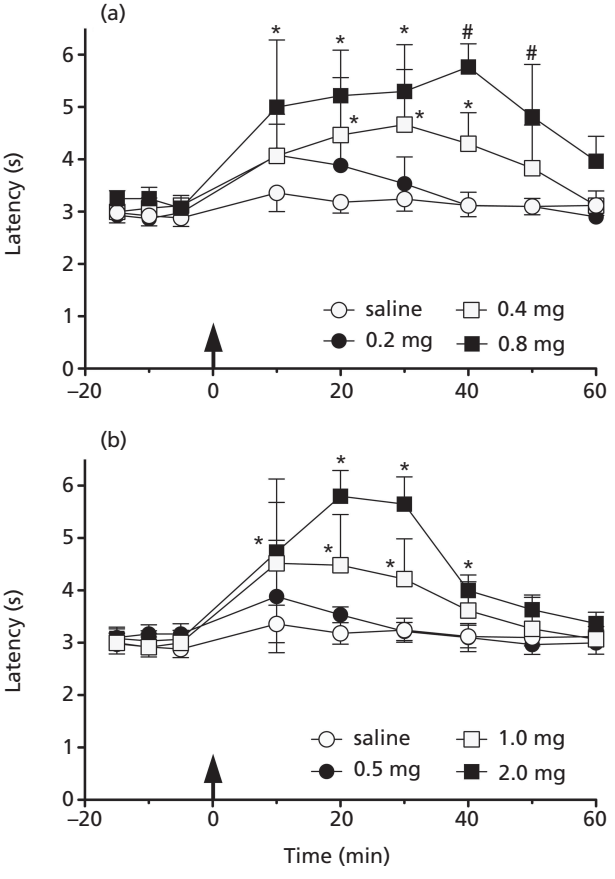

Figure 2 The time course of the effects of intrathecal administration (arrow) of saline (10 µl), neamine or 2-deoxystreptamine on the tail-flick latency of rats. Points are the mean (± SD) of six rats/curve. Doses of neamine (a) and 2-deoxystreptamine (b) are given in mg/10 µl. $P < 0.05$ vs saline-treated rats (*) or vs all the remaining groups (#).

neamine, but not 2-deoxystreptamine, was also dose-dependent. No animal showed gross disturbance of motor coordination, which was judged by visual inspection after intrathecal administration. The curves in Figure 2a and 2b were significantly different in terms of treatment $F_{3,20} = 28.0$ and 14.4, respectively) and time ($F_{8,160} = 27.8$ and 36.5, respectively) and have a significant treatment × time interaction ($F = 57.8$ and 36.9, respectively) ($P < 0.0001$ in all cases).

The estimated ED_{50} of neamine and 2-deoxystreptamine in the test were 1.35 mg (confidence limits: 0.61 and 2.95) and 1.05 (confidence limits: 0.68 and 1.60), respectively. The regression lines obtained for neamine and 2-deoxystreptamine did not deviate significantly from parallelism (slope ratio = 1.74 < slope ratio factor = 4.0), nor did they differ significantly regarding the calculated potency in this test (potency ratio = 1.28; confidence limits: 0.52 and 3.13).

The antinociceptive effects of intrathecal injection of neamine and 2-deoxystreptamine on incision allodynia

None of the rats had spontaneous pain 2 h after surgery. However, all rats had a significant reduction of the mechanical threshold of the incised hind paw measured 2 h after surgery. The time course of the changes induced by intrathecal neamine (0.1–0.8 mg) and 2-deoxystreptamine (0.5–2.0 mg) in the mechanical threshold is shown in Figure 3a and 3b, respectively. The effects of the smaller doses of neamine did not differ from the effect of saline (10 µl). However, the higher doses of neamine and all doses of 2-deoxystreptamine produced a significant increase of the mechanical threshold compared with the effect of saline. The duration of the effects of neamine and 2-deoxystreptamine was also dose-dependent. No animal showed gross disturbance of motor coordination, which was judged by visual inspection after intrathecal administration. The curves in Figure 3a and 3b were significantly different in terms of treatment ($F_{3,20} = 48.1$ and 231.6, respectively) and time ($F_{9,180} = 475,2$ and 1508.0, respectively) and have a significant treatment × time interaction ($F_{27,180} = 44.18$ and 152.2, respectively) ($P < 0.0001$ in all cases).

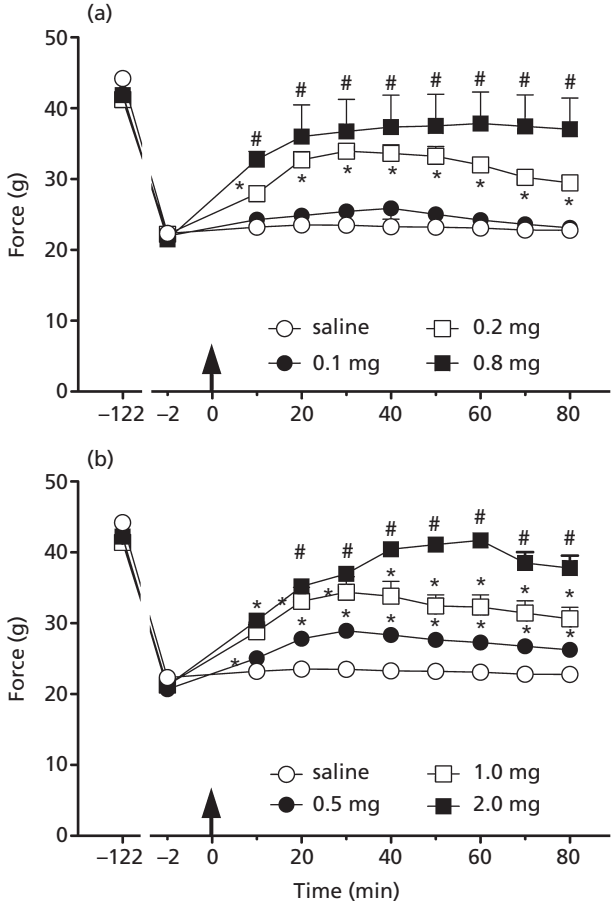

Figure 3 The time course of the effects of intrathecal administration (arrow) of saline (10 µl), neamine or 2-deoxystreptamine on the mechanical threshold (in grams) of rats before and 2 h after a surgical incision of a hind paw. Points are the mean (± SD) of six rats/curve. Doses of neamine (a) and 2-deoxystreptamine (b) are given in mg/10 µl. $P < 0$ .05 vs saline-treated rats (*) or vs all the remaining groups (#).

The estimated ED_{50} of neamine and 2-deoxystreptamine were 0.20 mg (confidence limits: 0.14 and 0.276) and 0.78 mg (confidence limits: 0.776 and 0.783), respectively. The regression lines obtained for neamine and 2-deoxystreptamine did not deviate significantly from parallelism (slope ratio = 1.38 < slope ratio factor = 3.48). Neamine was 3.9 (confidence limits: 1.39 and 11.48) times more potent than 2-deoxystreptamine. No change in the mechanical threshold of the non-incised hind paw of saline-, neamine- or 2-deoxystreptamine-treated rats was detected throughout the period of experimentation (data not shown).

The antinociceptive effects of intrathecal injection of neamine and 2-deoxystreptamine on neuropathic pain

None of the rats exhibited autotomy or any motor abnormality after surgery. All animals had a significant reduction of the mechanical threshold measured 8–10 days after surgery in the operated hind paw compared with preoperative thresholds. None of the rats had spontaneous pain during this period. The time course of the changes induced in the mechanical threshold of operated rats by intrathecal neamine (0.2–0.8 mg) and 2-deoxystreptamine (0.5–2.0 mg) is shown in Figure 4a and 4b, respectively. All doses of neamine and 2-deoxystreptamine produced a significant and dose-dependent increase of the mechanical threshold compared with the effect of saline (10 µl). The effects of different doses of neamine and 2-deoxystreptamine remained significantly above those of the control throughout the period of observation. No animal showed gross disturbance of motor coordination, which was judged by visual inspection after intrathecal administration. The curves in Figure 4a and 4b were significantly different in terms of treatment ($F_{3,20} = 552.4$ and 1358.0, respectively) and time ($F_{9,180} = 1263.0$ and 2877.0, respectively) and have a significant treatment × time interaction ($F_{27,180} = 116.9$ and 545.8, respectively) ($P < 0.0001$ in all cases).

The estimated ED_{50} of neamine and 2-deoxystreptamine were 0.28 mg (confidence limits: 0.12 and 0.63) and 0.79 mg (confidence limits: 0.46 and 1.34), respectively.

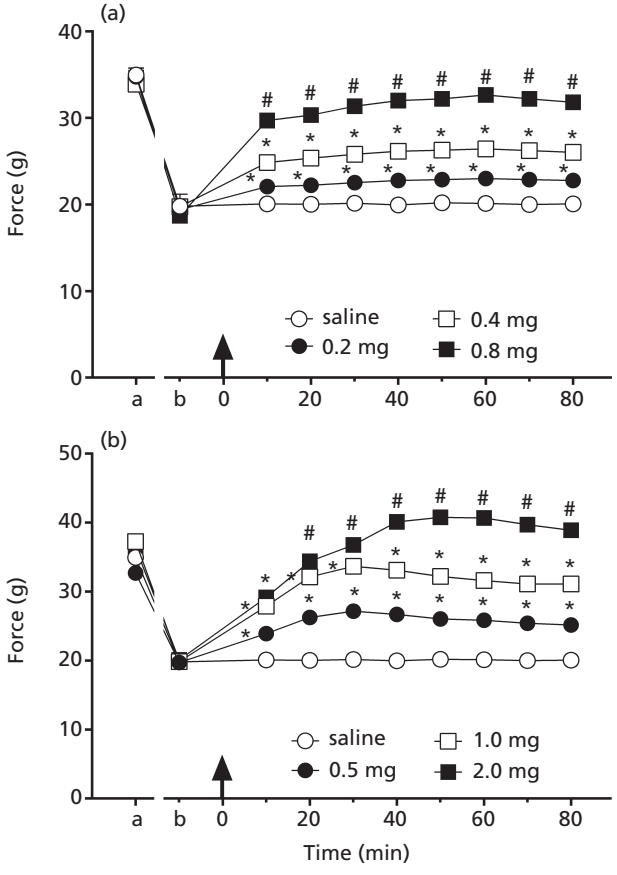

Figure 4 The time course of the effects of intrathecal administration (arrow) of saline (10 μl), neamine or 2-deoxystreptamine on the mechanical threshold (in grams) of rats before (a) and 8–10 days after (b) the chronic ligature of the sciatic nerve. Points are the mean (± SD) of six rats/curve. Doses of neamine (a) and 2-deoxystreptamine (b) are given in mg/10 μl. $P < 0.05$ vs saline-treated rats (*) or vs all the remaining groups (#).

The regression lines obtained for neamine and 2-deoxystreptamine did not deviate significantly from parallelism (slope ratio = 1.28 < slope ratio factor = 4.0). Neamine was 2.82 (confidence limits: 1.07 and 7.44) times more potent than 2-deoxystreptamine. No change in the mechanical threshold of the non-operated hind paw of saline-, neamine- or 2-deoxystreptamine-treated rats was detected throughout the period of experimentation (data not shown).

The effects of intrathecal injection of tetra-azidoneamine on the incision allodynia

Tetra-azidoneamine (3 mg/10 μl) or 5% DMSO (10 μl) was injected intrathecally in groups of six rats each 2 h after surgical incision of a hind paw. Neither this neamine derivative nor the vehicle produced a significant change in the post-incision mechanical allodynia (Figure 5).

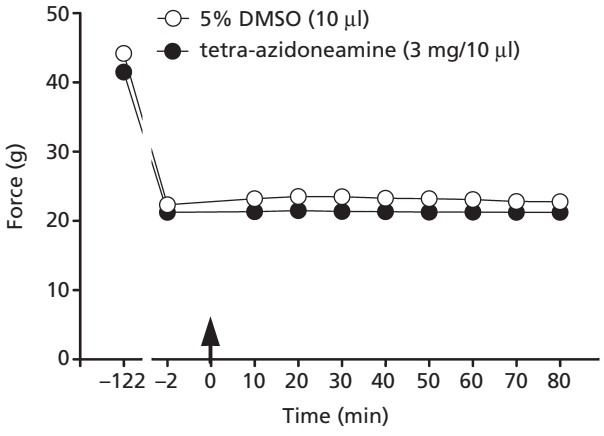

Figure 5 The time course of the effects of intrathecal administration (arrow) of DMSO (10 μl), or tetra-azidoneamine (3 mg/10 μl) on the mechanical threshold (in grams) of rats before and 2 h after a surgical incision of a hind paw. Points are the mean (± SD) of six rats/curve.

Antagonism of the effects of intrathecal injection of neamine and 2-deoxystreptamine on incision allodynia by intrathecal calcium chloride

All rats exhibited a significant reduction of the mechanical threshold of the incised hind paw measured 2 h after surgery. The intrathecal injection of saline did not produce significant changes in the incision allodynia. Neamine (0.8 μg, Figure 6a) and 2-deoxystreptamine (2.0 μg, Figure 6b) produced a significant increase in the mechanical threshold compared with the effect of saline. A further intrathecal injection of calcium chloride (4 and 8 μg) 39 min after the derivatives produced a transitory and dose-dependent reduction of the antiallodynic effect of neamine and 2-deoxystreptamine. Calcium chloride (8 μg) injected 39 min after saline did not produce effects different from baseline values. The curves in Figure 6a and 6b were significantly different in terms of treatment ($F_{2,15} = 702.8$ and 910.0, respectively) and time ($F_{7,105} = 458.0$ and 1755.0, respectively) and have a significant treatment × time interaction ($F_{14,105} = 114.8$ and 495.3, respectively) ($P < 0.0001$ in all cases).

Discussion

The presented results demonstrate that intraspinal administration of the neomycin derivatives neamine and 2-deoxystreptamine in rats has an antinociceptive effect in

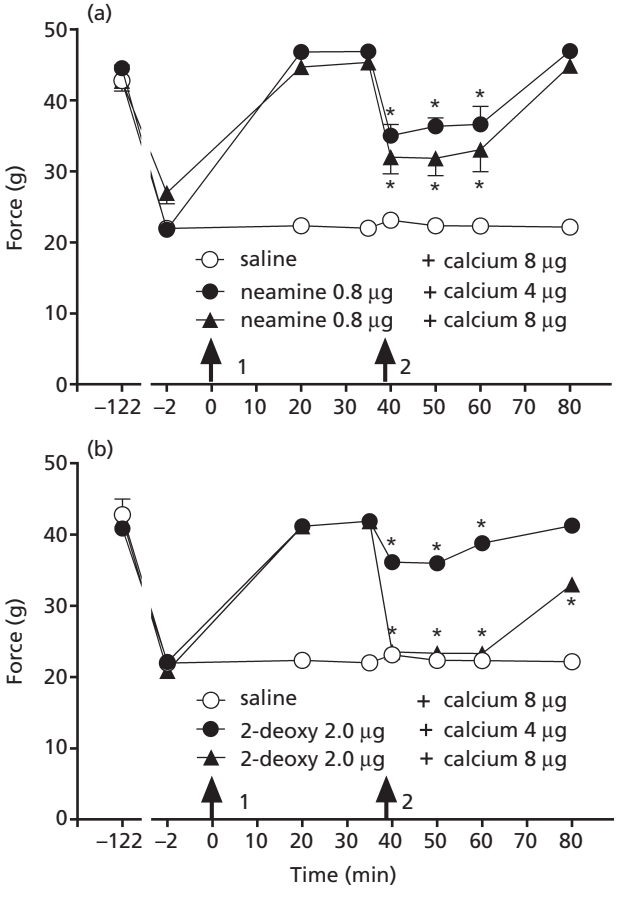

Figure 6 Reduction of the antiallodynic effect of neamine (a) and 2-deoxystreptamine (b) by intrathecal administration of calcium chloride in rats. Thresholds (in grams) to mechanical stimulation were measured before and 2 h after a surgical incision of a hind paw. Saline, neamine or 2-deoxystreptamine (2-deoxy) was injected (arrow 1) 39 min before calcium chloride (arrow 2). Points are the mean (± SD) of six rats/curve. $*P < 0.05$ vs threshold measured at time t = 30 min.

the tail-flick test and models of incisional and neuropathic pain. The effect was not accompanied by changes of motor coordination in all cases. The approximate ED_{50} (confidence limits in parenthesis) were 1.35 mg (0.61 and 2.95), 0.20 mg (0.14 and 0.27) and 0.28 mg (0.12 and 0.63), respectively, and for 2-deoxystreptamine they were 1.05 mg (0.68 and 1.60), 0.78 mg (0.776 and 0.783) and 0.79 mg (0.46 and 1.34), respectively. The effects of both derivatives lasted less than 60 min in the tail-flick test but reduced incisional and neuropathic pain for at least 80 min. The mechanical threshold measured in the non-incised and non-operated hind paw after intrathecal injection did not change throughout the period of observation. Therefore, the reduction of incisional or neuropathic pain is not due to an overall antinociceptive effect produced by the derivatives.

Aminoglycoside antibiotics are antinociceptive following intracerebroventricular administration in mice (20–80 μg; 40–160 μg and 80–320 μg, for neomycin, gentamicin and kanamycin, respectively)[2] and rat (ED_{50} = 50.12 μg, for gentamicin)[3] tail-flick test. Neomycin is antinociceptive in the rat tail-flick (ED_{50} = 9.22 μg; confidence limits 6.98 and 12.17) and incision pain tests (ED_{50} = 83.17 μg; confidence limits 51.6 and 133.9).[17] Neomycin applied *in situ* is also antinociceptive against formalin- or carrageenan-induced inflammatory pain rats, but much higher doses of the antibiotic were necessary (34 and 58 mg, respectively).[26] The intraspinal administration of morphine was also antinociceptive in the rat tail-flick test (ED_{50} = 0.29 μg; confidence limits 0.19 and/0.47).[22] Thus, the neomycin derivatives seem to be weaker than neomycin and morphine regarding antinociception. Neamine was more potent than 2-deoxystreptamine in the incision and neuropathic pain models, but these derivatives had similar potency in the tail-flick test.

Tetra-azidoneamine, a neamine derivative in which free amino groups are replaced with azido groups, did not change the mechanical allodynia displayed by rats 2 h after the surgical incision of a hind paw. We conclude that the presence of free amino groups in the structure of neomycin and its derivatives is fundamental for its antiallodynic effect. However, the aminoglycoside unit seems to be very important for the antinociceptive potency of these compounds.

We have also shown that the reduction of incisional allodynia by intrathecal neamine and 2-deoxystreptamine was antagonized by further intrathecal administration of calcium chloride in a transitory and dose-dependent manner. Intrathecal gentamicin-induced antinociception in the rat tail-flick test is also dose-dependently antagonized by intrathecal administration of calcium chloride.[3] We conclude that the effect of neamine and 2-deoxystreptamine may be due to a calcium antagonist property, a mechanism known to alter the effects of aminoglycoside antibiotics on various peripheral tissues.[6]

The earlier demonstration that intrathecal aminoglycoside antibiotics produce analgesia in rodents[3] is an interesting finding, suggesting the eventual use of these drugs in clinical pain management. Neamine and 2-deoxystreptamine seem to be less potent than neomycin and morphine. However, the present data showing that neomycin derivatives that do (neamine) or do not (2-deoxystreptamine) exhibit ototoxicity retain the antinociceptive property of the entire molecule may promote these derivatives as alternative targets for further studies on the management of clinical post-surgical and neuropathic pain.

Conclusions

Our results demonstrate that neamine and 2-deoxystreptamine, but not tetra-azidoneamine, have an antinociceptive property in different models of pain in rats. Thus, the presence of free amino groups in the structure of neomycin derivatives is fundamental to their antiallodynic effect. The reduction of incisional mechanical allodynia by intrathecal neamine and 2-deoxystreptamine was antagonized by intrathecal administration of calcium chloride in a transitory and dose-dependent manner. Therefore, the antiallodynic effect of the derivatives may be due to their action as calcium antagonists.

Declarations

Conflict of interest

The Author(s) declare(s) that they have no conflicts of interest to disclose.

Funding

This study was supported by FAPESP. ACR was the recipient of an FAPESP fellowship.

Acknowledgement

The authors greatly appreciate the technical assistance of M.A. Carvalho, P.R. Castania and I.A. Daniel.

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