Effects of Different Dosage of Dexamethasone on Behavioral, Electrophysiological, and Histomorphological Recovery in a Chronic Sciatic Nerve Compression Model

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Abstract

Objective. To investigate the effects of different doses of topical dexamethasone (Dex) on sciatic nerves with simulated compressive neuropathy.

Methods. Thirty-two Wistar rats were divided into four groups of 8: Sham group: no compression of the sciatic nerve + no treatment; Saline: chronic compression of the left sciatic nerve for 4 weeks + saline; 0.8% Dex: chronic compression + 0.8 mg of Dex; 3.2% Dex: chronic compression + 3.2 mg of Dex. Two sponge strips soaked with saline or Dex were placed under and over the nerve for 30 min in both Dex groups. Mixed-nerve-elicited somatosensory evoked potentials (M-SSEPs) and compound muscle action potentials (CMAPs) were measured to verify the compressive neuropathy in post-treatment follow-up. Behavioral observations of thermal hyperalgesia tests were quantified before electrophysiological examinations. Treated and contralateral nerves were harvested for histomorphological analysis.

Results. M-SSEP and CMAP amplitudes significantly decreased and latencies were significantly prolonged on postcompression thermal hyperalgesia tests. Rats in both Dex groups showed significant improvement in both sensory and motor conductive values and in neurological function, as well as increased mean myelin diameter on the final histomorphological examination. For rats in the saline group, these parameters showed incomplete recovery compared with the Sham group and the precompression baseline. Moreover, the changes after Dex treatment were not dose-dependent.

Conclusions. Topical Dex reversed electrophysiological, behavioral, and structural changes in chronically compressed sciatic nerves. Differences between the beneficial effects of high-dose and low-dose Dex were nonsignificant.

Key Words. Dexamethasone; Local Application; Evoked Potentials; Sciatic Nerve; Rat

Introduction

Corticosteroid injections, a type of local anti-inflammatory therapy, is a widely used conservative care option available for different neuropathic pain conditions such as nerve entrapment syndrome [1,2], neuralgia [3,4], and neuroma [5,6]. When treating carpal tunnel syndrome (CTS), this procedure is generally considered safe; outcomes associated with this therapy vary, and
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some studies question its long-term efficacy [2,7–10]. Two systematic reviews report moderate-to-strong evidence for its short and moderate-term efficacy compared with surgical treatment [8,11].

Corticosteroid injections can reduce perineural inflammation and soft tissue swelling, and may stabilize the neural membrane, thus limiting the symptom-causing cross talk in ischemic nerve fibers [12]. However, treatment methods and doses vary, and knowledge of the possible adverse effects of corticosteroid on nerves is limited and unclear [7,12–14]. Ideally, the dosage should be adapted to achieve the maximal clinical efficacy, maintain the longest duration, and avoid potential toxicity, but comparative studies on the forms and doses of corticosteroids plus their clinical outcome are not plentiful, and their results are inconsistent [2,12,14].

Because of controversies about the efficacy and the optimal dose for treating CTS, we examined the functional, electrophysiological, and histological changes that occur after treating rats with chronic nerve compression (CNC) plus neuropathy with high and low doses of the corticosteroid dexamethasone (Dex). In addition to CNC, locally administered corticosteroids may be also useful for spinal conditions involving local chronic inflammation, for example, disc herniation, spinal stenosis, degenerative disc disease, and so forth.

We previously found that directly applying Dex to the sciatic nerves of healthy rats caused a dose-dependent and transiently deleterious effect on their neural conductive properties. We hypothesized that this impaired electrophysiological response was associated with neural dysfunction. However, the effects of corticosteroids and, in particular, the effect of different doses of corticosteroids on chronically compressed nerves is still unknown. In this study, we used a rat model of CNC to characterize any of the positive, negative or neuroprotective effects that topical application of different doses of Dex may have following chronic compression injuries. We wanted to illustrate the effects of these treatments on nerve regeneration and functional outcome.

Methods

Experimental Animals, Silastic Tubing on Sciatic Nerve, and Groups

The experiments were done in accordance with the guidelines on animal experiments in our university; the protocol was approved by the Animal Ethics Committee. Thirty-two 8-week-old male Wistar-rats weighing 250–330g were randomly assigned to one of four groups of 8. Saline (positive control), 0.8%, and 3.2% Dex groups of rats all underwent chronic compression of the sciatic nerve. CNC was achieved by wrapping one sciatic nerve with silastic tubing and ligated with 3 sutures. The nerves were then treated with saline, 0.8% Dex (Sigma-Aldrich, St. Louis, MO), and 3.2% Dex, respectively, 4 weeks after the operation. The sham group of rats underwent an operation that exposed the sciatic nerve; they were otherwise not treated (negative control). The rats were housed two per cage with controlled temperature, a 12/12-h light/dark cycle, and free access to standard rat chow and water.

Experimental Protocol of Surgical Procedures of Silastic Tubing and Topically Applied Dex

Compression Procedure Using Silastic Tubing with Ligation

All surgical procedures were done using 50 mg/kg of intraperitoneal (i.p.) sodium pentobarbital anesthesia (Nembutal; Abbott, North Chicago, IL). The depth of anesthesia was determined by assessing the withdrawal reflex on tail pinch, and subsequent doses of pentobarbital were administered as necessary to maintain adequate anesthetic depth. The rats were intramuscularly (i.m.) premedicated with gentamycin (8 mg/kg) (Yung-Shin Pharmaceutical Industrial, Taichung, Taiwan). Core temperature was monitored with rectal probes connected to a multichannel thermometer (Portable Hybrid Recorder, model 3087; Yokogawa Hokushin Electric, Tokyo, Japan) and maintained within 37–38°C using a homeothermic animal blanket (Jerboa Scientific, New Taipei City, Taiwan) until the rats recovered. Rats were then placed in the prone position with extended hips. The surgical procedures were done under aseptic conditions. A 20-mm segment of the left sciatic nerve was then exposed using a gluteal-splitting approach. A 1-cm length of sterile biological inert silastic tube with an internal diameter of 1.2 mm was cut longitudinally and placed around the nerve. After the wrapping procedure, the tube and nerve were ligated with three 9/0 sutures (Ethilon; Ethicon, Somerville, NJ); this provided peripheral nerve compression without disturbing the visible extrinsic blood flow. In the Sham group, the contralateral sciatic nerve was exposed and carefully mobilized, but no tube was placed around it.

Topically Applied Dexamethasone

Four weeks after the previously compressed sciatic nerves were re-exposed. The induction of chronic compressive sciatic nerve neuropathy was electrophysiologically (using sensory and motor evoked potentials) and functionally (using a thermal hyperalgesia test) confirmed. The sciatic nerve with ligated silastic tubing was freed from its investing fascia and fibrosis with extreme care to avoid interfering with the circulation. Two small sponge strips (0.6 × 1.0 cm²) (Gelfoam; Pharmacia-Upjohn, Kalamazoo, MI) soaked with 0.8 or 3.2 mg of Dex were placed under and over the injured left sciatic nerve for 30 min as treatment for the 0.8 and 3.2 mg Dex groups. Each sponge strip was soaked and then diluted, if necessary, with saline to a total volume of 100 mL.
Dexamethasone & Chronic Sciatic Nerve Compression

Sensory and Motor Evoked Potentials

Two electrophysiological surveillance systems were set up and data were recorded immediately, 2 weeks, and 4 weeks after compression. Two weeks and 6 weeks after the topical saline treatment and thermal hyperalgesia test, electrophysiological data were recorded again.

Ascending Evoked Potentials Were Elicited from the Hind Paws

Spinal somatosensory evoked potentials (SSEPs) were recorded using bipolar needle electrodes. The recording cathode was placed in the thoracolumbar (T-L) interspinous ligament, a corresponding reference electrode was placed in subcutaneous tissue just proximal to the recording electrode, and a ground electrode was placed in the pelvic girdle ipsilateral to the side stimulated. Stimulation was delivered using subcutaneous needle electrodes placed just medial to both ankles adjacent to the tibial nerve. Square impulses 0.2 ms in duration with intensity 5 times greater than the threshold of visible potential were used. The stimulation rate was 5 impulses per second, with 20 repetitions. The recording was filtered for data from 1 to 5,000 Hz; recording time was 20 msec.

Descending Compound Muscle Action Potential

Compound Muscle Action Potential (CMAP) was recorded using monopolar myographic needle electrodes placed in each belly of the bilateral gastrocnemius muscles. The spinal cord was stimulated at T12-L3 using needle electrodes in the interspinous ligament. The acquisition parameters were similar to those for SSEP, but the presentation rate of stimulation decreased to 1/sec, and the recording was filtered for data from 1 to 2,000 Hz. A ground electrode was placed subcutaneously between the stimulus and the recording site. At least three sequential single-sweep runs (i.e., without averaging) with similar waveforms were recorded to check and verify the consistency of the response.

Thermal Hyperalgesia Test

The thermal hyperalgesia test was done using an infrared heat stimulus device (Plantar Test [Hargreaves Apparatus]; Ugo Basile, Comerio, Italy) as previously described [15]. The test was run 5 times: precompression, postcompression 2 and 4 weeks, and post-treatment 2 and 6 weeks. The rats were acclimated to the machine for 10 min, and then their hind paws were thermally stimulated. There was at least a 3-min interval between paws. Each paw was stimulated four or five times, and the mean value in seconds was then calculated as the thermal nociceptive threshold.

Tissue Harvesting and Histological Examination

After the final functional and electrophysiological tests were done, all the rats were injected with an overdose of pentobarbital (100 mg/kg) and then transcardially perfused with 0.1 M phosphate-buffered saline/4% paraformaldehyde. The sciatic nerve was harvested 5-mm from the epicenter proximally and distally and then immersed in 4% paraformaldehyde overnight. Tissue samples were fixed with 1% osmium tetroxide for 2 h and then dehydrated with graded alcohol and embedded in resin. One-micrometer thick sections were collected and the myelin was observed (magnification: 200×) under a microscope (Axio Imager 2; Carl Zeiss Microscopy GmbH, Oberkochen, Germany). Five random views of a sciatic nerve cross-section were photographed. The mean diameter and number of myelin sheaths were manually calculated by 2 researchers blinded to each other's results.

Statistical Analysis

Statistical analyses of body weight, electrophysiological examinations, and thermal hyperalgesia tests were done using repeated-measure two-way analysis of variance (ANOVA) with a posthoc Kruskal-Wallis test, for myelin sheath mean diameter and number were done using one-way ANOVA and then the Kruskal-Wallis test. Significance was set at \( P < 0.05 \).

Results

General Postoperative Conditions

The general condition of all the rats was good. They all showed steady body-weight gains throughout the three-month period of observation. There was no significant difference between the three experimental groups: body weights were not significantly affected by the constriction operation or by topical steroid treatment.

Electrophysiological Findings

M-SSEP at the thoracolumbar junction interspinous ligament was consistent and stable; it showed a major negative wave preceded by a small positive wave. CMAP was also consistent from gastrocnemius muscles. There were no significant differences between right and left lower limbs in amplitude and latency, nor were there differences between the control and experimental groups in basic preoperative or immediately postoperative recordings (Figures 1–3). However, 2 and 4 weeks postoperatively, M-SSEP and CMAP amplitude was significantly lower and latency significantly longer for the three experimental groups (Tables 1–4), which indicated consistent neural damage.

At 2 and 6 weeks post-treatment, rats in both Dex groups showed that the amplitude and latency improved significantly at most parameters. In contrast, the Saline group showed no significant post-treatment change.
However, compared with precompression baseline levels, the significantly lower amplitude and longer latency persisted in all three groups. Moreover, differences between the three experimental and Sham groups were significant, but they were not significant between the Dex groups. These findings suggested incomplete but significant recovery of neural conduction of both sensory and motor tracts in the two Dex groups, but not in the Saline group.

Figure 1 Representative tracings of M-SSEP and CMAP from the Group 3.2% Dex. Responses were elicited from the experimental side with dexamethasone (left) and the control side with saline (right) at various time points before and after treatment. Note there were no significant differences in both sides.

Thermal Hyperalgesia Test
A thermal hyperalgesia test showed significant sciatic functional impairment in all three experimental groups compared with preoperative levels. The paw withdrawal latency to noxious thermal stimuli was significantly lower in three experimental groups than in the Sham group (Table 5) 2 and 6 weeks postcompression, which is consistent with compression-induced thermal hyperalgesia. Rats in the Saline group showed progressive and significant decreases in thermal hyperalgesia for the entire 10 weeks of follow-up. Rats in the 0.8% Dex group showed a significant difference (Table 5) postcompression 6 week (=Post-treat 6 wks) but did not achieve statistical significance in the high dexamethasone dose.

Histological Observation
Pathology tests of the tissue from the two Dex group rats showed fewer morphological changes than did tests of tissue from Saline group rats. There were numerous small diameter myelin basic protein (MBP) cells (Figure 4A, arrowhead) and evidence of
demyelination (Figure 4A, arrow). The number of MBP cells was not significantly different between the four groups (Saline: 819.0 ± 178.9, 0.8% Dex: 679.8 ± 211.3, 3.2% Dex: 1050.0 ± 338.0, Sham: 7930.8 ± 380.0). The mean diameter of the myelin sheaths surrounding the neurons in the tissue from the rats in both Dex groups was greater than that in the Saline group (0.8% Dex: 6.96 ± 0.60, 3.2% Dex: 6.56 ± 0.82, and Saline: 4.66 ± 0.45). The difference between the two Dex groups was not significant. Although the mean diameters in both Dex groups were less than that in the Sham group, the difference was not significant, but the difference between the Saline and Sham groups was significant (Figure 5).

Discussion

In a model of simulated chronic sciatic nerve compressive neuropathy, we found that topical dexamethasone (Dex), a corticosteroid, significantly inhibited thermal hyperalgesia, improved both sensory and motor conduction, and increased the mean myelin diameter of the rat sciatic nerve.

There are many chronic sciatic nerve compression animal models. Some apply direct compression to the nerve with a balloon [16], a constricting band [17–19], or another artificial device [20]. Some use only tight or loose ligation [21,22]. Chronic compression models in the rat show progressive epineural and perineural fibrosis and thinning of the myelin based on the duration of compression duration. The changes seen in rats are identical to those seen in human beings [19]. These studies are not only helpful for characterizing the effect of compression on a nerve and identifying potential mechanisms; they are also used to investigate the therapeutic effects of medications for compression neuropathy.
In our experiment, we used a locally ligated silastic-tubing induced entrapment model [23], first, because it met the criteria for mimicking the pathogenesis and clinical entrapment neuropathy of carpal tunnel and cubital tunnel syndromes [24,25], second, because the experimental findings in our recent study [26] showed progressive and consistent neurological dysfunction with a decline in amplitude and a prolongation of latency [26], and third, because the model required only a topical application of corticosteroid and minimal surgery.

Although most of the corticosteroid injections studies supported the potential efficacy of this treatment, the correlation between the improvement in clinical presentations, electrophysiological changes, or both, and the appropriate dose of corticosteroid is controversial [7–11]. The relative inconsistency in validation may be the result of the complexity of the pharmacological mechanism of corticosteroids and of different pre-existing preoperative neuropathy of the patients. Therefore, in addition to clinical trials and other clinical observations and a well-designed experiment was needed to interpret the electrophysiological, functional, and histological changes after different doses of corticosteroids, but none had ever been reported. We found that after CNC in both the low- and high-dose-treated groups, amplitudes increased and latency decreased in SSEP and CMAP, neurological function was less impaired, there were fewer myelinated axons, and myelin sheaths were thicker and had a larger mean diameter than did the experimental groups without corticosteroid treatment.

Corticosteroids promote functional recovery from neuropathic pain, which may be more related to their complex mechanisms than to their ability to decrease perineural

Figure 3 Representative tracings of M-SSEP and CMAP from the Group Control (saline). Responses were elicited from the experimental side with saline (left) and the control side with saline (right) at various time points before and after treatment. Note there were no significant differences in both sides.
Table 1  Comparison of amplitude changes of M-SSEPs between Sham and Experimental groups

<table>
<thead>
<tr>
<th>Time Point</th>
<th>Saline</th>
<th>0.8% Dex</th>
<th>3.2% Dex</th>
<th>Sham</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td></td>
</tr>
<tr>
<td>Precom</td>
<td>0.96 ± 0.08^1 (0.83–1.11)</td>
<td>0.93 ± 0.13^1 (0.75–1.08)</td>
<td>0.95 ± 0.12^1 (0.81–1.00)</td>
<td>0.96 ± 0.04^1 (0.91–1.01)</td>
<td>ns</td>
</tr>
<tr>
<td>Postcom 2 wks</td>
<td>0.63 ± 0.12a^2 (0.51–0.79)</td>
<td>0.69 ± 0.25a^3 (0.26–0.82)</td>
<td>0.62 ± 0.26a^3,5 (0.26–0.85)</td>
<td>1.00 ± 0.08b (0.89–1.05)</td>
<td>0.009</td>
</tr>
<tr>
<td>Postcom 4 wks</td>
<td>0.60 ± 0.11a^2 (0.47–0.72)</td>
<td>0.57 ± 0.12a^2,3 (0.51–0.76)</td>
<td>0.52 ± 0.17a^2,3 (0.24–0.73)</td>
<td>0.96 ± 0.05b (0.90–1.00)</td>
<td>0.002</td>
</tr>
<tr>
<td>Post-treat 2 wks</td>
<td>0.63 ± 0.11a^2 (0.48–0.75)</td>
<td>0.70 ± 0.12a^2,3,4 (0.52–0.85)</td>
<td>0.71 ± 0.15a^2,3,5 (0.50–1.08)</td>
<td>1.00 ± 0.07b (0.90–1.10)</td>
<td>0.005</td>
</tr>
<tr>
<td>Post-treat 6 wks</td>
<td>0.63 ± 0.11a^2 (0.50–0.78)</td>
<td>0.80 ± 0.12a^1,4 (0.64–0.98)</td>
<td>0.71 ± 0.17a^2,3 (0.50–0.93)</td>
<td>1.02 ± 0.08b (0.91–1.10)</td>
<td>0.002</td>
</tr>
</tbody>
</table>

P-value 0.001 0.004 0.003 ns

Dex = dexamethasone; SD = standard deviation; RC = range of change; Precom = precompression; Postcom = postcompression.

*Relative values (mean ± SD) of latency with percentage of saline or different doses of dexamethasone or saline side of the same rat.

a–c,1–4 Different superscript letters and numbers indicate a significant difference across groups or time points (Kruskal-Wallis test).

Table 2  Comparison of amplitude change of CMAP between Sham and Experimental groups

<table>
<thead>
<tr>
<th>Time Point</th>
<th>Saline</th>
<th>0.8% Dex</th>
<th>3.2% Dex</th>
<th>Sham</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SD* (RC)</td>
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<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td></td>
</tr>
<tr>
<td>Precom</td>
<td>1.00 ± 0.07^1 (0.88–1.08)</td>
<td>0.96 ± 0.10^1 (0.77–1.09)</td>
<td>0.99 ± 0.04^1 (0.92–1.04)</td>
<td>0.99 ± 0.13 (0.88–1.23)</td>
<td>ns</td>
</tr>
<tr>
<td>Postcom 2 wks</td>
<td>0.65 ± 0.16a^2 (0.32–0.82)</td>
<td>0.62 ± 0.14a,2,3 (0.48–0.86)</td>
<td>0.54 ± 0.16a,2,3 (0.16–0.67)</td>
<td>1.08 ± 0.09b (1.00–1.17)</td>
<td>0.001</td>
</tr>
<tr>
<td>Postcom 4 wks</td>
<td>0.61 ± 0.17a,2 (0.33–0.81)</td>
<td>0.51 ± 0.20a,2 (0.21–0.78)</td>
<td>0.51 ± 0.17a,2 (0.43–0.88)</td>
<td>0.99 ± 0.10b (0.89–1.15)</td>
<td>0.002</td>
</tr>
<tr>
<td>Post-treat 2 wks</td>
<td>0.53 ± 0.19a^2 (0.25–0.75)</td>
<td>0.66 ± 0.26a^2,3 (0.19–0.88)</td>
<td>0.69 ± 0.20a,2,3 (0.31–0.93)</td>
<td>0.98 ± 0.08b (0.88–1.08)</td>
<td>0.005</td>
</tr>
<tr>
<td>Post-treat 6 wks</td>
<td>0.68 ± 0.16a^2 (0.33–0.77)</td>
<td>0.77 ± 0.10a,1,4 (0.67–1.00)</td>
<td>0.78 ± 0.20a^2 (0.66–1.07)</td>
<td>1.03 ± 0.09b (0.93–1.17)</td>
<td>0.005</td>
</tr>
</tbody>
</table>

P-value 0.01 0.001 0.00 ns

Dex = dexamethasone; SD = standard deviation; RC = range of change; Precom = precompression; Postcom = post-compression.

*Relative values (mean ± SD) of latency with percentage of saline or different doses of dexamethasone or saline side of the same rat.

a–c,1–4 Different superscript letters and numbers indicate a significant difference across groups or time points (Kruskal-Wallis test).
### Table 3  Comparison of latency change of M-SSEPs between Sham and Experimental groups

<table>
<thead>
<tr>
<th>Group</th>
<th>Saline</th>
<th>0.8% Dex</th>
<th>3.2% Dex</th>
<th>Sham</th>
<th>P-value</th>
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<tbody>
<tr>
<td>Time Point</td>
<td>Mean ± SD* (RC)</td>
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<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td>P-value</td>
</tr>
<tr>
<td>Precom</td>
<td>1.03 ± 0.05 (0.97–1.12)</td>
<td>1.03 ± 0.12 (0.87–1.15)</td>
<td>1.03 ± 0.06 (0.96–1.13)</td>
<td>1.00 ± 0.05 (0.94–1.05)</td>
<td>ns</td>
</tr>
<tr>
<td>Postcom 2 wks</td>
<td>1.30 ± 0.11 (1.17–1.44)</td>
<td>1.20 ± 0.08 (1.17–1.28)</td>
<td>1.34 ± 0.18 (1.16–1.44)</td>
<td>1.01 ± 0.05 (0.95–1.09)</td>
<td>0.001</td>
</tr>
<tr>
<td>Postcom 4 wks</td>
<td>1.31 ± 0.24 (1.11–1.69)</td>
<td>1.29 ± 0.10 (1.12–1.44)</td>
<td>1.31 ± 0.24 (1.00–1.69)</td>
<td>1.00 ± 0.03 (0.96–1.03)</td>
<td>0.008</td>
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<tr>
<td>Post-treat 2 wks</td>
<td>1.31 ± 0.15 (1.06–1.45)</td>
<td>1.22 ± 0.32 (0.96–1.90)</td>
<td>1.24 ± 0.15 (1.06–1.45)</td>
<td>0.99 ± 0.06 (0.91–1.08)</td>
<td>0.009</td>
</tr>
<tr>
<td>Post-treat 6 wks</td>
<td>1.22 ± 0.10 (1.11–1.43)</td>
<td>1.15 ± 0.20 (1.00–1.58)</td>
<td>1.13 ± 0.16 (1.11–1.43)</td>
<td>0.99 ± 0.04 (0.94–1.05)</td>
<td>0.018</td>
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<tr>
<td>P-value</td>
<td>0.000</td>
<td>0.011</td>
<td>0.000</td>
<td>ns</td>
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Dex = dexamethasone; SD = standard deviation; RC = range of change; Precom = precompression; Postcom = postcompression.
*relative values (mean ± SD) of latency with percentage of saline or different doses of dexamethasone or saline side of the same rat.
\(a-c,1–4\) different superscript letters and numbers indicate a significant difference across groups or time points (Kruskal-Wallis test).

### Table 4  Comparison of latency change of CMAP between Sham and Experimental groups

<table>
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<tr>
<th>Group</th>
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<th>3.2% Dex</th>
<th>Sham</th>
<th>P-value</th>
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</thead>
<tbody>
<tr>
<td>Time Point</td>
<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td>Mean ± SD* (RC)</td>
<td>P-value</td>
</tr>
<tr>
<td>Precom</td>
<td>1.02 ± 0.07 (0.90–1.12)</td>
<td>1.06 ± 0.11 (0.93–1.25)</td>
<td>1.03 ± 0.11 (0.93–1.18)</td>
<td>1.01 ± 0.07 (0.92–1.08)</td>
<td>ns</td>
</tr>
<tr>
<td>Postcom 2 wks</td>
<td>1.24 ± 0.13 (1.08–1.44)</td>
<td>1.26 ± 0.15 (0.96–1.35)</td>
<td>1.40 ± 0.12 (1.15–1.36)</td>
<td>1.06 ± 0.07 (0.95–1.17)</td>
<td>0.012</td>
</tr>
<tr>
<td>Postcom 4 wks</td>
<td>1.35 ± 0.17 (1.20–1.69)</td>
<td>1.32 ± 0.09 (1.16–1.39)</td>
<td>1.4 ± 0.13 (1.13–1.66)</td>
<td>1.09 ± 0.07 (1.04–1.18)</td>
<td>0.002</td>
</tr>
<tr>
<td>Post-treat 2 wks</td>
<td>1.27 ± 0.13 (1.06–1.33)</td>
<td>1.23 ± 0.24 (0.91–1.27)</td>
<td>1.21 ± 0.15 (0.92–1.36)</td>
<td>0.99 ± 0.09 (0.81–1.08)</td>
<td>0.023</td>
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<tr>
<td>Post-treat 6 wks</td>
<td>1.20 ± 0.11 (1.08–1.34)</td>
<td>1.14 ± 0.14 (1.00–1.43)</td>
<td>1.19 ± 0.13 (1.04–1.30)</td>
<td>0.98 ± 0.03 (0.96–1.04)</td>
<td>0.003</td>
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<td>P-value</td>
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<td>0.006</td>
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Dex = dexamethasone; SD = standard deviation; RC = range of change; Precom = precompression; Postcom = postcompression.
*relative values (mean ± SD) of latency with percentage of saline or different doses of dexamethasone or saline side of the same rat.
\(a-c,1–4\) different superscript letters and numbers indicate a significant difference across groups or time points (Kruskal-Wallis test).
Inflammation, attenuate inflammatory peptides, and reduce nerve ischemia [23,27]. In the early stage of compressive neuropathy, corticosteroids decrease the concentration of substance P, an important element in pain perception in rats with implanted sciatic nerve cuffs [23]. Corticosteroids are also well known to be membrane stabilizers. They act as a nociceptive C-fiber membrane stabilizer and inhibit ectopic neural discharge and decrease both thermal hyperalgesia and mechanical allodynia [28]. However, in middle-to-late stage experimental CNC, we found progressive connective tissue fibrosis and nerve fiber change, local demyelination and remyelination, concurrent apoptosis and proliferation of Schwann cells, and remodeling of Schwann cell myelin architecture [24,25,29]. Interestingly, a recent study [30] reported that endogenous glucocorticoids improved myelination through glucocorticoid receptor in Schwann cells in a rat peripheral nerve injury model. Endogenous neurosteroid protects sensory neurons of the dorsal root ganglion against sciatic-nerve-injury-induced apoptosis of dorsal-root-ganglion satellite-glial cells [31]. Local corticosteroids may also be involved in neurogenesis and neuroprotection through this mechanism in nerve compression injury. Our findings are comparable to the functional, electrophysiological, and histological changes of midstage CNC, and functional recovery corticosteroid-treated rats was significantly greater than in saline-treated rats, although some neural compromise still remained.

A few studies [12,14,32] related to the effectiveness of different doses of corticosteroids used to treat CTS have reported diverse results. One comparison [14] of the effect of 25 mg and 100 mg of hydrocortisone, and another [32] of the effect of 15 mg and 35 mg of methylprednisolone showed no significant difference in favorable subjective responses about outcomes. In contrast, a randomized control trial [12] of 20, 40, and 60 mg of methylprednisolone showed a significant number of positive responses (i.e., free of major symptoms) in the 60-mg group. However, because of the design of this study, patients were not routinely followed up and not assessed by the primary outcome and the study has been criticized [7]. Because there is no consensus on the appropriate dose, careful delineation of the dose-dependent effects of corticosteroids on human peripheral nerves is critically important for a proper risk-benefit analysis of their local use. In this study, the improvements in electrophysiological, functional, and histological parameters in the low-dose and high-dose corticosteroid groups were not significantly different. Additionally, the low-dose group showed better longitudinal electrophysiological recovery in longitudinal compared with precompression data than did the high-dose group. We cannot explain this counterintuitive result. We hypothesized that high-dose corticosteroid therapy in CTS negatively affects circulation and nerve conduction.

Shishido et al. [34] found that even when it was not directly injected into a nerve, 30 min after a local application of dexamethasone (0.4%, 0.1 mL), the normal blood flow in rat sciatic nerves was reduced 14.4%.

<table>
<thead>
<tr>
<th>Table 5</th>
<th>Comparison of plantar test between Sham and Experimental groups</th>
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<tbody>
<tr>
<td>Group</td>
<td>Saline (0.8% Dex)</td>
</tr>
<tr>
<td></td>
<td>Mean ± SD* (RC)</td>
</tr>
<tr>
<td>Time Point</td>
<td>Precom</td>
</tr>
<tr>
<td></td>
<td>12.38 ± 1.011 (11.2–14.3)</td>
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<tr>
<td></td>
<td>9.02 ± 1.755 (6.3–11.1)</td>
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<tr>
<td></td>
<td>8.82 ± 1.305 (8.3–9.9)</td>
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<tr>
<td></td>
<td>8.27 ± 1.245 (6.2–9.7)</td>
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<tr>
<td></td>
<td>8.10 ± 1.665 (5.2–9.8)</td>
</tr>
<tr>
<td>P-value</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Dex = dexamethasone; SD = standard deviation; RC = range of change; Precom = precompression; Postcom = postcompression.

*Relative values (mean ± SD) of latency with percentage of saline or different doses of dexamethasone or saline side of the same rat.

**Different superscript letters and numbers indicate a significant difference across groups or time points (Kruskal-Wallis test).
recent study [26] also showed that topical perineural Dex dose-dependently adversely affected neural conduction. This impaired electrophysiological response might be associated with neural dysfunction. Moreover, many studies [8,35] provide evidence that corticosteroid injections cause musculoskeletal structural or functional damage; therefore, all these potential effects should be considered during the treatment.

**Limitations of this Study**

One limitation of our study is the lack of a long-term end-point that might have given us the opportunity to draw another and stronger conclusion about the long-term therapeutic effects of corticosteroids on the damage caused by CNC. However, because evidence from clinical practice indicates that there are no consistent

**Figure 4** Pathomorphological changes from the A-Group Control (saline); B-Group 0.8 % Dex ; C-Group 3.2% Dex ; D-Group Sham. (arrowhead- small diameter MBP cells, arrow- demyelinated cells)

**Figure 5** Comparison of myelin mean diameter change in each group. Note the significant increment in the Group 0.8% Dex and 3.2% Dex. (*, *** $P < 0.05$).
long-term results of corticosteroid therapy [11], the clinical applicability of this type of research is undoubtedly limited. A second limitation is the rat sciatic nerve silastic-tubing CNC model may not be applicable to human CTS because the mechanism of human entrapment neuropathy is not similar to direct compression on the rat sciatic nerve. A third limitation is that a Dex-soaked gelform was directly applied to the compressed nerve. Although local injection into the silastic tube is also possible, we have used this gelform method for the past decade, and have obtained valid, reliable, and well-controlled results when working with small animals. Our results in the present investigation are compatible with prior findings [26,33]. Finally, a fourth limitation is that when we interpreted the histological results, we used only hematoxylin and eosin staining. That we used osmium tetroxide staining in lieu of more advanced histochemical and immunohistochemical techniques and electron microscopy to detect the changes.

In conclusion, neurological deficits were significantly more attenuated, neural conduction was significantly more improved, and mean myelin diameter was well preserved significantly more augmented in Dex-treated groups than in saline-treated groups, but the changes were not significant difference between the low-dose and high-dose Dex-treated groups. Additional studies are necessary for a better understanding of the mechanisms involved. Our most important finding is that Dex was effective at a low dose, that may preclude the need for using high dose treatments that may potentiate neurotoxicity without clinical benefit.

REFERENCES
16 Diao E, Shao F, Liebenberg E, Rempel D, Lotz JC. Carpal tunnel pressure alters median nerve function in a dose-dependent manner: A rabbit model for carpal tunnel syndrome. J Orthop Res 2005;23:218–23.


