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Effect of Collagen Nerve Wrapping in a Rabbit Peripheral Neuropathy Model

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Background: Collagen nerve wraps (CNWs) theoretically allow for improved nerve gliding and decreased perineural scarring, and create a secluded environment to allow for nerve myelination and axonal healing. The goal of this study was to investigate the effect of CNWs on nerve gliding as assessed by pull-out strength and nerve changes in a rabbit model of peripheral neuropathy.

Methods: Ten New Zealand rabbits were included. Sham surgery (control) was performed on left hindlimbs. To simulate compressive neuropathy, right sciatic nerves were freed of the mesoneurium, and the epineurium was sutured to the wound bed. Five rabbits were euthanized at 6 weeks [scarred nerve (SN); n = 5]. Neurolysis with CNW was performed in the remaining rabbits at 6 weeks (CNW; n = 5), which were euthanized at 22 weeks. Outcomes included peak pull-out force and histopathological markers of nerve recovery (axonal and Schwann cell counts).

Results: The CNW group demonstrated significantly higher pull-out forces compared with the CNW sham control group (median: 4.40N versus 0.37N, $P = 0.043$) and a trend toward greater peak pull-out forces compared with the SN group (median: 4.40N versus 2.01N, $P = 0.076$). The CNW group had a significantly higher median Schwann cell density compared with the CNW control group (CNW: 1.30×10^{-3} cells/μm^2 versus CNW control: 7.781×10^{-4} cells/μm^2, $P = 0.0431$) and SN group (CNW: 1.30×10^{-3} cells/μm^2 versus SN: 7.31×10^{-4} cells/μm^2, $P = 0.009$). No significant difference in axonal density was observed between groups.

Conclusion: Our findings suggest using a CNW does not improve nerve gliding, but may instead play a role in recruiting and/or supporting Schwann cells and their proliferation. *(Plast Reconstr Surg Glob Open 2021;9:e3919; doi: 10.1097/GOX.0000000000003919; Published online 11 November 2021.)*

INTRODUCTION

Nerve wrapping is a surgical treatment option that can be used in peripheral nerve injury repair or neuropathy treatment to prevent neural scarring, adhesion formation, and allow for adequate nerve gliding, which, when lost, can result in a traction nueropathy.[1–3] Clinically, nerve wrapping has been described with successful outcomes in multiple settings of chronic or recurrent nerve compression, such as carpal tunnel syndrome,[4–6] Wartenberg syndrome,[7] and cubital tunnel syndrome.[6,8,9] A variety of nerve wraps can be utilized, ranging from venous wraps, collagen nerve wraps (CNWs), and novel nerve wraps (eg, polyglycolic acid).

Prior animal model studies have investigated the biomechanical properties and promotion of nerve healing by nerve wraps to support the positive outcomes reported in the clinical literature. These investigations have been focused on the role of nerve wraps in treating peripheral nerve injury in animal models of neurotmesis,[10] where the peripheral nerve is completely transected. Despite clinical outcome studies reporting on nerve wraps, and specifically CNWs[6] for the use of chronic peripheral nerve neuropathy, prior animal studies have been limited to investigating the effects of CNW in animal models of peripheral nerve repair.[11–15] Specifically, these studies have focused on the histological outcomes of CNWs. To our knowledge, no

prior study has investigated the biomechanical properties of CNW in a peripheral nerve neuropathy animal model.

The purpose of this study was to (1) describe a novel rabbit model of peripheral nerve neuropathy with adhesions, (2) investigate the effect of CNW used to treat the neuropathy on nerve gliding as assessed by pull-out strength, and (3) investigate the neural changes (eg, axonal density and Schwann cell proliferation) after using a CNW. Based on clinical experience, we hypothesized that, contrary to popular belief, using a CNW would not restore nerve gliding properties to that of the native state, and instead remain fixed to scar tissue, resulting in a higher pull-out strength than the control state.

METHODS

Ten adult female New Zealand white rabbits (Charles River Laboratories International, Wilmington, Mass.) all weighing 3–4 kg were included in this study. A rabbit model was selected over other small mammals (eg, rats and mice) due to their greater similarity to humans in regard to neural tissue and healing properties compared with smaller mammals.[16,17] Animal care complied with the guidelines of the authors' institution, the National Institutes of Health on the care and use of laboratory animals, and USDA guidelines. All animals were housed and provided food and water ad libitum. All facets of this study were performed following Institutional Animal Care and Use Committee approval at SUNY Downstate Medical Center.

To investigate nerve gliding in the setting of peripheral neuropathy with perineural adhesions, a New Zealand White rabbit sciatic nerve model was developed through surgical scarring of the sciatic nerve. An overview of the protocol is presented in Figure 1. All rabbit surgeries were performed under the supervision and care of the veterinary staff of SUNY Downstate Medical Center Division of Comparative Medicine using standardized protocols. All rabbits received appropriate perioperative monitoring and supportive therapy.

All rabbits underwent an index procedure in which a sham surgery was performed in the left hindlimbs (control group, n = 10). In this sham surgery, the rabbits were anesthetized with general anesthesia under the care of the veterinary care staff. Proper aseptic technique was maintained for the patient, surgeon, instruments, and materials. After induction and intubation, the rabbit was positioned prone and prepared and draped in a sterile fashion. The sciatic nerve was then exposed via a muscle-splitting approach to the posterolateral thigh (Fig. 2). After the nerve was directly visualized, surgical wounds were closed in a layered fashion.
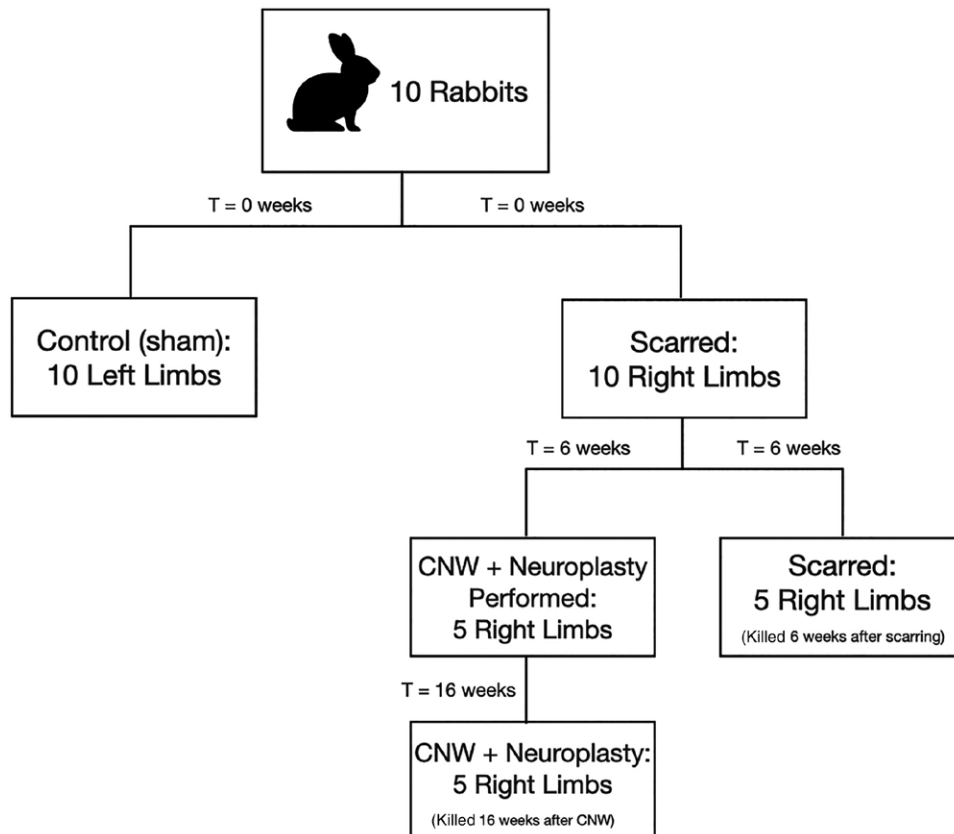
Fig. 1. Flow chart of surgical methods. Each rabbit underwent an initial surgery (at 0 weeks), a sham surgery to the left hindlimb (n = 10), and a nerve scarring procedure to the right hindlimb (n = 10). At 6 weeks after the nerve scarring procedure, half of the rabbits were killed (n = 5) and the other half underwent a CNW procedure with neuroplasty (n = 5). The CNW group was killed 16 weeks after the CNW procedure (22 weeks after the scarring procedure).
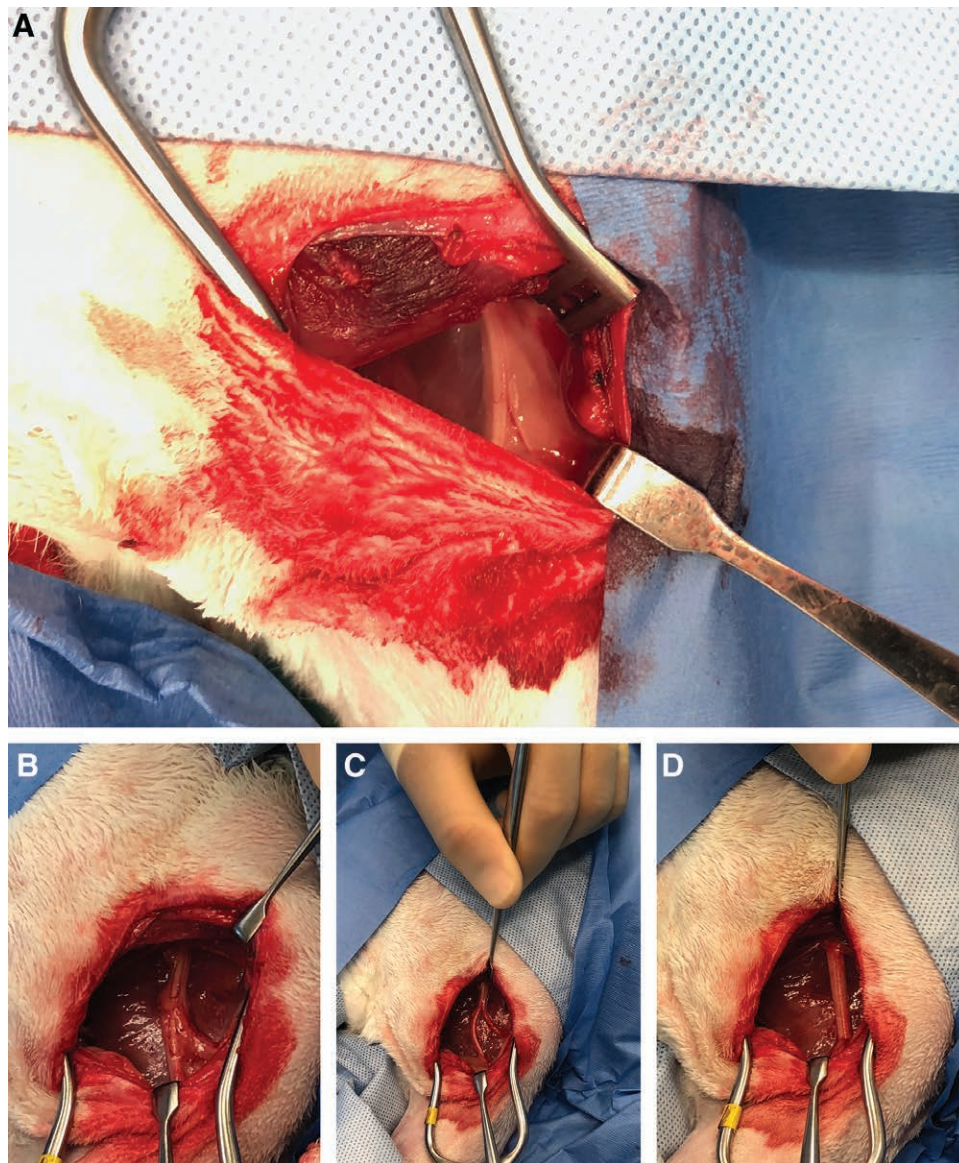
Fig. 2. Pictures of the (A) native sciatic nerve, (B) scarred sciatic nerve, (C) sciatic nerve after neurolysis, and (D) sciatic nerve after CNW.

During the same procedure, all rabbits (n = 10) also underwent a sciatic scarring procedure on the right hindlimb in a similar manner as previously described; however, an additional injury to the meosneurium was performed.[18,19] While the rabbits remained under general anesthesia, the sciatic nerve was exposed using the same approach as on the contralateral control side. After exposing the sciatic nerve using 3.5× magnification, a 10 mm portion of the proximal sciatic nerve (before branching) was freed of the mesoneurium. The mesonerium forms a sheath around the epineurium and allows for nerve gliding, protects the nerve from trauma, and contains a significant amount of segmental vasculature to the nerve. Thus, disruption of the mesoneurium could not only inhibit nerve gliding but also result in adhesion formation. Moreover, removing part of the mesonerium exposes the nerve directly to possible trauma. Therefore,

we hypothesized that this procedure would also induce a peripheral neuropathy of the proximal sciatic nerve and this was tested using the subsequently described histopathology protocol. In addition, 10 mm of the surrounding tissue bed was scarred using bipolar electrocautery to further promote adhesion formation. The nerve epineurium was then affixed to the cauterized nerve bed with 8-0 nylon suture, and wounds were closed in a layered fashion (Fig. 2).

The rabbits were then maintained for 6 weeks, during which no rabbit experienced any perioperative or postoperative complication. At 6 weeks following the scarring procedure, five rabbits [scarred nerve (SN) group, n = 5] were euthanized via an American Veterinary Medical Association acceptable method, as this time point has reliably demonstrated high rates of adhesion formation in a variety of animal models, including rabbits.[18–22]

The other five rabbits (CNW group, n = 5) underwent neuroplasty (removal of the epineural adhesions) and CNW application at 6 weeks after the index scarring procedure to the right hindlimb (Fig. 2). CNWs were selected instead of other nerve wrapping alternatives due to their availability and available clinical literature. The rabbits were anesthetized with general anesthesia. The nerve was mobilized in the mid-thigh over an area 15 mm proximal from the splitting of the sciatic nerve. After neuroplasty, the nerve was wrapped in a type 1 collagen nerve wrap (NeuroMend, Collagen Matrix, Oakland, NJ) to isolate the nerve from the scarred tissue bed, and the wound was closed in layered fashion. No perioperative or postoperative complications were observed after the CNW surgery. These five rabbits were euthanized at 16 weeks after CNW (22 weeks after the nerve scarring procedure) using the same protocol described previously. This time point, 16 weeks after CNW, was based on a prior study by Bulstra et al,[16] which was selected because it had the most similar methodology as the present study and was performed in rabbits. Bulstra et al investigated recovery at 0, 1, 2, 4, 8 12, and 16 weeks after segmental peroneal nerve reconstruction. The authors found that on all outcome measures, the highest recovery rate was observed at 16 weeks. Specifically, using a validated ultrasound measurement, at 16 weeks the cross-sectional area of the TA had achieved 91% of the preoperative size. In addition, the isometric tetanic force measurement as described by Guisti et al[23] showed 104% recovery at 16 weeks. Furthermore, the N-ratio, which was defined as the total myelinated fiber area divided by the total tissue cable area as assessed with slides of the peroneal nerve stained with toluidine blue, demonstrated 83% recovery at 16 weeks. Based on these findings, 16 weeks was selected as the appropriate motor recovery period after CNW and neuroplasty.

Peak Pull-out Force Biomechanical Testing

Biomechanical pull-out force testing, as described by Crosio et al[24] and Tos et al[25] was performed in all hindlimbs following rabbit death. The sciatic nerves were exposed and dissected both proximally and distally, with care given to not violate the surrounding tissue bed. The distal healthy nerve ends were transected and proximally, the sciatic nerve was transected at the sciatic notch. The rabbits were affixed to wooden platforms. The distal nerve segments were sutured and interconnected to a 10N Instron static load cell (Instron 5566, Norwood, Mass.). A pulley was used to align the course of the sciatic nerve with the pull direction (Fig. 3). Using custom machinery and a uniaxial biomechanical servohydraulic testing frame, the nerves were pulled at a rate of 29 mm/min with 0.005 N pre-loading based on pilot testing from the distal transected healthy end. Load to failure testing when then performed to measure the peak force required to pull the nerve segment out of the tissue bed in the control, SN, and CNW groups. Lower pull-out force correlates with improved nerve gliding.

Histopathology

After biomechanical testing, explanted nerves were immersed in 4% paraformaldehyde while held

Fig. 3. Experimental setup for assessing pull out strength.

in extension for 3 minutes. The specimens were then embedded. A series of transverse sections (thickness: 7–8 μm) were created using a microtome and positioned on slides. Slides were then Nissl stained and deparaffinated, rehydrated with decreasing ethanol passages, and VEGF stained for high quality imaging of axons, as previously described by Sondell et al[26] This approach, using VEGF, allowed us to visually identify the circumference of an axon while the Nissl staining (a nucleic acid staining) stained nuclei and rough endoplasmic reticulum. Because axons themselves, unlike neuronal cell bodies, do not contain any ER or nuclei, positive Nissl staining within the circumference of the axon must be indicative of a myelin cell. In this case, because the samples were taken from the peripheral nervous system, these cells were Schwann cells. Cells beyond the circumference of the axon, as indicated with the VEGF staining, were not included in analysis, as the origin of these cells is unknown and likely heterogeneous (eg, inflammatory cells).

To quantify the axons and Schwann cells, all slide images were opened in their original format in QuPath (v0.1.2)[27] Four fields were then systematically selected from each slide and exported from QuPath into FIJI/ImageJ (v1.52)[28] All images were then batch segmented in FIJI/ImageJ. VEGF images were first segmented in ImageJ using the mean algorithm for axon counts. Axon lumens were then counted using a size filter of 15–250 μm^2 and a circularity filter of 0.25–1. Nissl images were segmented

using the Rényi entropy algorithm for Schwann cell counts[29] Schwann cell nuclei were then size filtered at 5–30 µm² and a circularity filter of 0.1–1. Particles recorded during segmentation were counted along with their respective field areas. The batch function in ImageJ was then used to automate image filtering. Data collected from batch segmentation were exported to Python (v3.6, Python Software Foundation, Wilmington, Del.) to calculate the axon density and Schwann cell density per slide.

Statistical Analysis

Data were aggregated and analyzed in Microsoft Excel (Microsoft, Redmond, Wash.). Statistical analyses were performed in STATA (v13, STATAcorp, College Town, Tex.). To assess for normal distribution of the data, a Shapiro-Wilk test was used. The values for peak pull-out force and Schwann cell density demonstrated a nonnormal distribution ($P > 0.05$) and are therefore presented with medians. For statistical comparisons, nonparametric tests were used. To compare the control to each of the treatment groups, a Wilcoxon sign-rank test was used, whereas a Mann–Whitney U test was used to compare the two treatment arms (scarred versus CNW). In addition, to compare the Schwann density at each location a group (SN or CNW), a Friedman test was used. Axonal densities demonstrated a normal distribution ($P > 0.05$); a paired *t*-test was used to compare the control groups with the treatment groups, whereas the treatment groups were compared with an unpaired *t*-test. To compare across multiple groups of paired or unpaired data, a one-way repeated measures ANOVA or one-way ANOVA was used, respectively. Significance was set at a *P* value less than 0.05.

RESULTS

Peak Pull-out

In the SN group (2.51 ± 1.90N, median: 2.01N), a significantly larger force was required to free the sciatic nerve compared with the SN control group (0.50 ± 0.40N, median: 0.55N $P = 0.043$) (Fig. 4). Similarly, the CNW group (4.31 ± 1.98N, median: 4.40N) required a significantly larger force to free the nerve from the surrounding tissue bed compared with the CNW control group (0.73 ± 0.51N, median: 0.37N, $P = 0.043$). When comparing the CNW group with the SN group, the CNW group trended to require a greater peak pull-out force to free the nerve ($P = 0.076$).

Histopathology

The CNW group had a significantly higher median Schwann cell density compared with the CNW control group (CNW: 1.30×10^{-3} cells/µm² versus CNW control: 7.781×10^{-4} cells/µm², $P = 0.0431$) (Fig. 5). There was no significant difference between the SN and SN control group ($P = 0.225$).

When investigating Schwann cell density by location, there was no significant difference between the median Schwann cell density in the segment proximal to the scar in the CNW versus SN groups (CNW: 9.13×10^{-4} cells/µm² versus SN: 9.09×0^{-4} cells/µm², $P = 0.465$) (Fig. 6). The CNW group had a higher density of Schwann cells in the region distal to the scar compared with the SN group; however, this difference was not significant (CNW: 1.28×10^{-3} cells/µm² versus SN: 6.67×10^{-4} cells/µm², $P = 0.117$). The CNW group, however, did have a significantly higher Schwann cell density compared with the SN group at the scar (CNW: 1.30×10^{-3} cells/µm² versus SN: 7.31×10^{-4} cells/µm², $P = 0.009$). No significant differences in density were noted within the SN group. Within the CNW group, the proximal segment demonstrated a significantly lower Schwann cell density compared with the scarred segment ($P = 0.031$), and a trend toward a lower density compared with the distal segment ($P = 0.069$).

Axonal density was then compared between groups. No significant differences were observed between the SN and SN control groups ($P = 0.200$) nor the CNW and CNW control groups ($P = 0.444$) (Fig. 7).
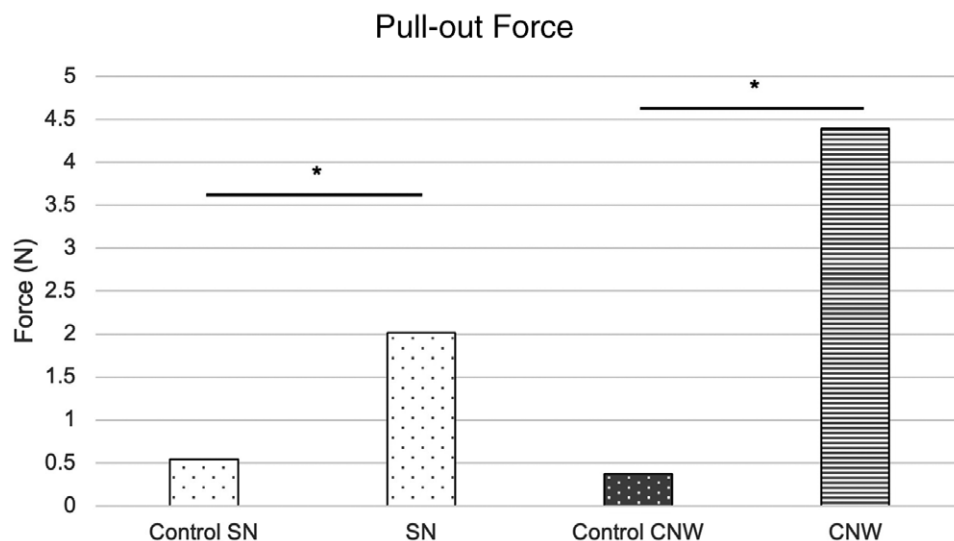

Fig. 4. Peak pull-out force for the SN, CNW, and their control groups (presented as medians, *$P < 0.05$).
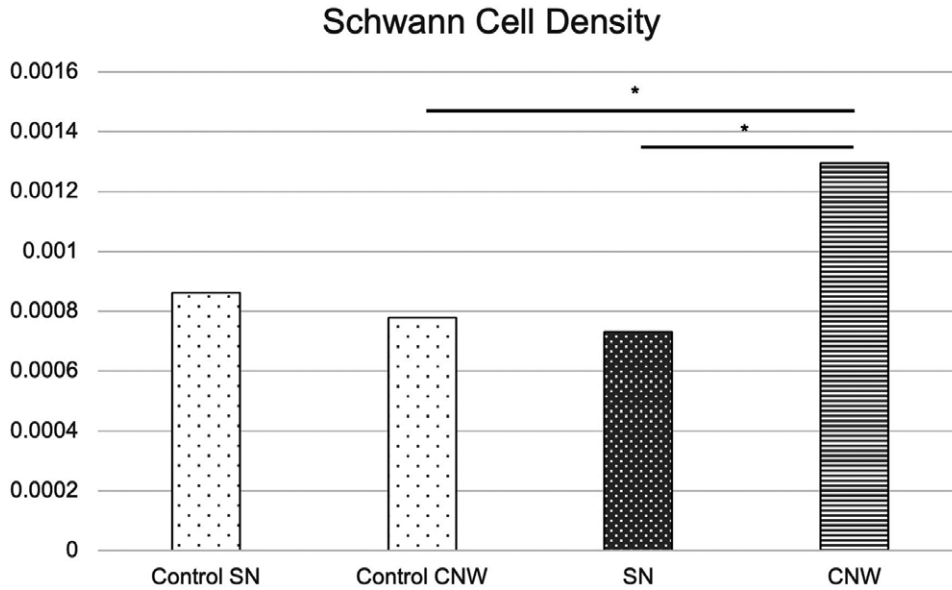
Schwann Cell Density

Fig. 5. Schwann cell density (cells/µm^2) for the SN, CNW, and their control groups at the scarred area (presented as medians, *$P < 0.05$).

There were no significant differences in density between the SN and CNW groups at the proximal, scarred, or distal segment (Fig. 8). In the CNW group there were no significant differences in density between segment locations. In contrast, the SN group demonstrated a significantly higher axonal density in the proximal segment compared with the distal one (proximal: $4.56 \times 10^{-3} \pm 3.3 \times 10^{-3}$ cells/µm^2, distal: $2.69 \times 10^{-3} \pm 3.02 \times 10^{-3}$ cells/µm^2 $P = 0.020$).

DISCUSSION

The primary goal of this study was to describe a rabbit model of peripheral neuropathy with adhesions and to investigate whether treatment with a CNW would significantly improve nerve gliding. Our results demonstrate that both scarring the nerve and using CNWs significantly increased peak pull-out pressure compared with the contralateral control. The CNW group, however, trended toward having a significantly greater peak pull-out force compared with the SN group. In addition, the CNW group had significantly higher Schwann cell densities compared with their matched controls and the SN group. Moreover, the proximal aspect of the nerve in the CNW group demonstrated a significantly lower Schwann cell density compared with the scarred area and a trend
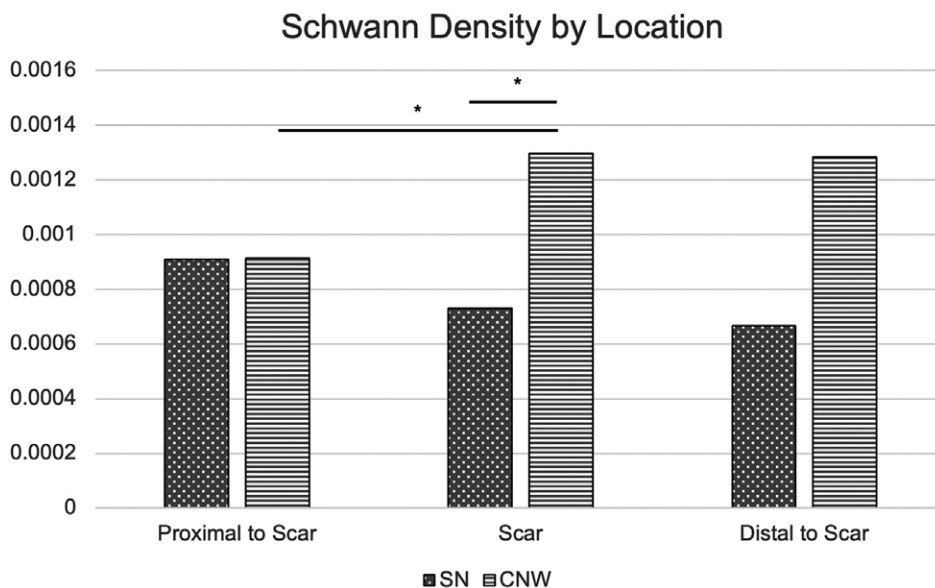
Schwann Density by Location

Fig. 6. Schwann densities (cells/µm^2) for the SN group and CNW group proximal to the scar, at the scar, and distal to the scar (presented as medians, *$P < 0.05$).
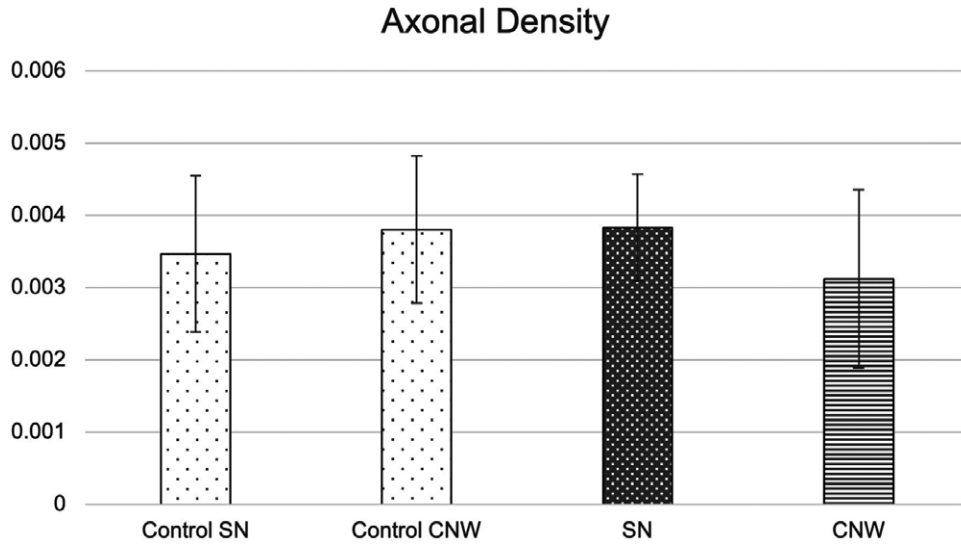
Axonal Density

Fig. 7. Axonal density (cells/μm²) for the SN, CNW, and their control groups at the scarred area.

toward lower density compared with the distal aspect of the nerve. Together, these results suggest that using a CNW does not improve nerve gliding but may instead play a role in recruiting and/or supporting Schwan cells and their proliferation.

Prior studies have put forth nerve adhesion models; however, few rabbit models have been reported, with the literature instead focusing on rat models.[19,20,24] Abe et al, for example, created perineural adhesions surrounding rabbit sciatic nerves by suturing the nerve adventitial layer to the nerve bed.[19] The authors observed fibrosis in the sciatic nerves and significant decreases in amplitude in compound muscle action potentials. This study, however, did not investigate the biomechanical properties of neural adhesions. In the present study, we observed that in our similar rabbit model, the scarred nerves required

significantly higher peak pull-out forces to break perineural adhesions compared with their matched controls, adding to the prior findings of Abe et al.

Furthermore, we also found that the CNW group needed a significantly higher peak pull-out force compared with the CNW control group and a trend toward the CNW group having significantly higher peak pull-out forces compared with the SN group. These findings suggest that CNWs do not restore nerve gliding to the native state and that, instead, they actually increase perineural adhesions. One of the motivations behind this study was that we observed the counter-intuitive finding that CNWs appeared to be scarred into local tissue during revision nerve repair surgeries. This clinical hypothesis has also been suggested in the literature. A case series by Liodaki et al, for example, investigated the histology of NeuraGen
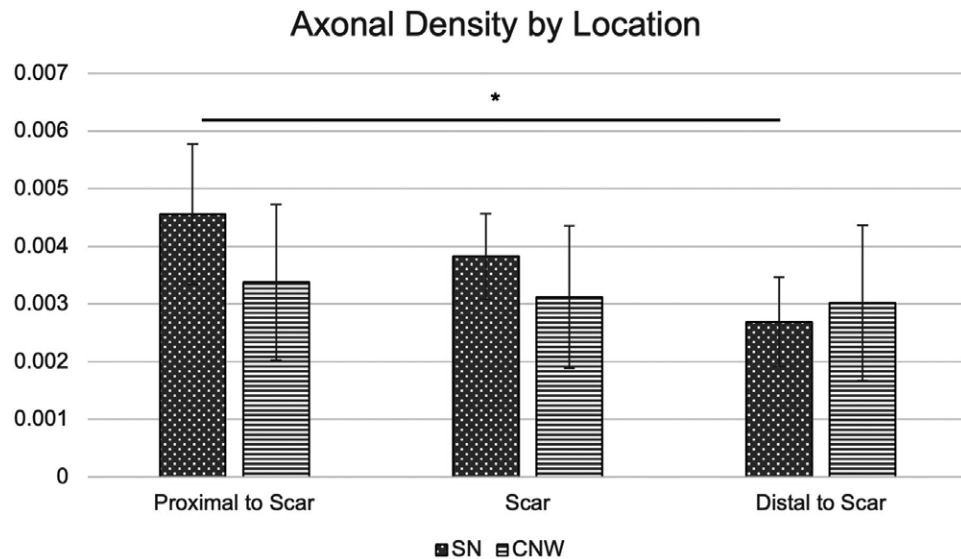
Axonal Density by Location

Fig. 8. Axonal densities (cells/μm²) for the SN group and CNW group proximal to the scar, at the scar, and distal to the scar (*$P < 0.05$).

nerve graft explants in patients who failed CNWs and were undergoing revision nerve autografts.[30] In three of the four cases, the authors observed significant areas of perineural fibrotic scar, but the authors were unable to discern the boundaries of the CNW versus the nerve bed, suggesting complete CNW resorption. In contrast, other rat studies have suggested that CNWs decrease perineural scars compared with primary suture repair.[12,31] However, these discrepancies may be due to differences in healing and adhesion formation between rats, rabbits, and humans or because these studies only histologically examined micron-level adhesions. Our findings support these clinical observations that CNWs increase adhesions, which may be in part due to the intrinsic collagen composition of the CNW or to an increase in local tissue damage when a CNW is placed.

We also observed that the CNW group had a significantly higher Schwann cell density at the scar site compared with the control group and SN group. Furthermore, there was a significantly lower density of Schwann cells proximal to the scar site. Together these results suggest that CNWs play a role in the proliferation and/or recruitment of Schwann cells. The clinical relevance and implications of this finding needs to be further examined, as Schwann cell senescence is well known to affect outcomes of nerve repair and regeneration.[32] In contrast to the Schwann cell findings, there were no significant differences in axonal densities between groups. Only a significantly higher axonal density was observed in the proximal segment of the SN group compared with the distal region. This distal axonal injury is likely due to downstream effects of the scarring procedure, which the CNW did not rescue. In contrast to prior studies, our study did not observe that CNWs influence axonal density.[33–35] This may be because our study concluded at 16 weeks after CNW application, which may not have been a long enough follow-up period to appreciate these changes. It may also be because our study, unlike the prior studies that used a neurotmesis model, used a neuropathy model. Chronic neuropathy first results in myelin sheath degradation and delayed nerve conduction, with axonal loss occurring as a late finding. Thus, in a neuropathy model, CNWs may only affect nerve myelination (as assessed with Schwann cell histopathology) and not axonal density.

Limitations

This study is subject to various limitations. The first limitation is the use of an animal model. This approach has inherent limitations, and it is unclear how directly translatable our findings are to human patients. However, we chose to use a rabbit as our animal model instead of smaller mammals such as mice or rats, as these smaller mammals have neurobiological regenerative and immunobiological mechanisms that are not directly relatable to humans.[16,17] A second limitation is the small sample size of this study. An a priori analysis was not performed before the initiation of this study. This may have limited our ability to detect a significant finding between the SN and CNW group for peak pull-out force, where instead a trend was observed. An additional limitation is the difference in duration between the nerve scarring procedure and euthanasia for the SN (a total of 6 weeks) and CNW (a total of 22 weeks) groups. Furthermore, during this duration, the CNW group underwent an additional surgery of neuroplasty and implantation of the CNW, whereas the SN group did not undergo a second sham surgery. This difference may play a role in peak pull-out forces because the additional surgery may have increased the risk of further adhesion development. In addition, this study examined differences in histopathology between the groups and along the nerve, but we did not perform any electrophysiology or axon propagation analysis. Finally, comparison of Schwann cell counts can be difficult to interpret in the setting of the different time-periods of nerve harvest. This could have changed with additional time and may have affected pull-out strength and cell counts.

CONCLUSIONS

CNWs resulted in significantly higher peak pull-out forces compared with the CNW control group and a trend toward greater pull-out forces compared with the SN group. CNWs, however, displayed significantly higher Schwann cell densities compared with the control and SN groups; no difference in axonal density was observed between the four groups. Our findings suggest using a CNW does not improve nerve gliding but may instead play a role in recruiting and/or supporting Schwann cells and their proliferation.

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REFERENCES

1. Weber RA, Breidenbach WC, Brown RE, et al. A randomized prospective study of polyglycolic acid conduits for digital nerve reconstruction in humans. *Plast Reconstr Surg*. 2000;106:1036–45; discussion 1046.
2. Zhu X, Wei H, Zhu H. Nerve wrap after end-to-end and tension-free neurorrhaphy attenuates neuropathic pain: A prospective study based on cohorts of digit replantation. *Sci Rep*. 2018;8:620.
3. Mackinnon SE. Pathophysiology of nerve compression. *Hand Clin*. 2002;18:231–241.
4. Spielman AF, Sankaranarayanan S, Skowronski P, et al. Recurrent and persistent carpal tunnel syndrome: "Triple-therapy approach". *J Orthop*. 2020;22:431–435.
5. Kokkalis ST, Mavrogenis AF, Vottis C, et al. Median nerve biodegradable wrapping: Clinical outcome of 10 patients. *Acta Orthop Belg*. 2016;82:351–357.
6. Soltani AM, Allan BJ, Best MJ, et al. Revision decompression and collagen nerve wrap for recurrent and persistent compression neuropathies of the upper extremity. *Ann Plast Surg*. 2014;72:572–578.
7. Gaspar MP, Kane PM, Vosbikian MM, et al. Neurolysis with amniotic membrane nerve wrapping for treatment of secondary wartenberg syndrome: A preliminary report. *J Hand Surg Asian Pac Vol*. 2017;22:222–228.
8. Papatheodorou LK, Williams BG, Sotereanos DG. Preliminary results of recurrent cubital tunnel syndrome treated with neurolysis and porcine extracellular matrix nerve wrap. *J Hand Surg Am*. 2015;40:987–992.

9. Gaspar MP, Abdelfattah HM, Welch IW, et al. Recurrent cubital tunnel syndrome treated with revision neurolysis and amniotic membrane nerve wrapping. *J Shoulder Elbow Surg.* 2016;25:2057–2065.

10. Lopez J, Xin K, Quan A, et al. Poly(ε-Caprolactone) nanofiber wrap improves nerve regeneration and functional outcomes after delayed nerve repair. *Plast Reconstr Surg.* 2019;144:48e–57e.

11. Kim PD, Hayes A, Amin F, et al. Collagen nerve protector in rat sciatic nerve repair: A morphometric and histological analysis. *Microsurgery.* 2010;30:392–396.

12. Lee JY, Parisi TJ, Friedrich PF, et al. Does the addition of a nerve wrap to a motor nerve repair affect motor outcomes? *Microsurgery.* 2014;34:562–567.

13. Pertici V, Laurin J, Marqueste T, et al. Comparison of a collagen membrane versus a fibrin sealant after a peroneal nerve section and repair: A functional and histological study. *Acta Neurochir (Wien).* 2014;156:1577–1590.

14. Saltzman EB, Villa JC, Doty SB, et al. A comparison between two collagen nerve conduits and nerve autograft: A rat model of motor nerve regeneration. *J Hand Surg Am.* 2019;44:700.e1–700.e9.

15. Jeon T, Vutescu ES, Saltzman EB, et al. Evaluation of two collagen conduits and autograft in rabbit sciatic nerve regeneration with quantitative magnetic resonance DTI, electrophysiology, and histology. *Eur Radiol Exp.* 2018;2:19.

16. Bulstra LF, Hundepool CA, Friedrich PF, et al. Motor nerve recovery in a rabbit model: Description and validation of a non-invasive ultrasound technique. *J Hand Surg Am.* 2016;41:27–33.

17. Calvert GT, Doolabh VB, Grand AG, et al. Rat-strain differences in recovery following peripheral-nerve allotransplantation. *J Reconstr Microsurg.* 2001;17:185–191.

18. Ikeda K, Yamauchi D, Osamura N, et al. Hyaluronic acid prevents peripheral nerve adhesion. *Br J Plast Surg.* 2003;56:342–347.

19. Abe Y, Doi K, Kawai S. An experimental model of peripheral nerve adhesion in rabbits. *Brit J Plast Surg.* 2005;58:533–540.

20. Smit X, van Neck JW, Afoke A, et al. Reduction of neural adhesions by biodegradable autocrosslinked hyaluronic acid gel after injury of peripheral nerves: An experimental study. *J Neurosurg.* 2004;101:648–652.

21. Kikuchi K, Setoyama K, Takada S, et al. E8002 inhibits peripheral nerve adhesion by enhancing fibrinolysis of l-ascorbic acid in a rat sciatic nerve model. *Int J Mol Sci.* 2020;21:E3972.

22. Foran IM, Hussey V, Patel RA, et al. Native paraneurial tissue and paraneurial adhesions alter nerve strain distribution in rat sciatic nerves. *J Hand Surg Eur Vol.* 2018;43:316–323.

23. Giusti G, Kremer T, Willems WF, et al. Description and validation of isometric tetanic muscle force test in rabbits. *Microsurgery.* 2012;32:35–42.

24. Crosio A, Valdatta L, Cherubino M, et al. A simple and reliable method to perform biomechanical evaluation of postoperative nerve adhesions. *J Neurosci Methods.* 2014;233:73–77.

25. Tos P, Crosio A, Pellegatta I, et al. Efficacy of anti-adhesion gel of carboxymethylcellulose with polyethylene oxide on peripheral nerve: Experimental results on a mouse model. *Muscle Nerve.* 2016;53:304–309.

26. Sondell M, Lundborg G, Kanje M. Vascular endothelial growth factor has neurotrophic activity and stimulates axonal outgrowth, enhancing cell survival and Schwann cell proliferation in the peripheral nervous system. *J Neurosci.* 1999;19:5731–5740.

27. Bankhead P, Loughrey MB, Fernández JA, et al. QuPath: Open source software for digital pathology image analysis. *Sci Rep.* 2017;7:16878.

28. Schindelin J, Arganda-Carreras I, Frise E, et al. Fiji: An open-source platform for biological-image analysis. *Nat Methods.* 2012;9:676–682.

29. Dhadialla PS, Ohiorhenuan IE, Cohen A, et al. Maximum-entropy network analysis reveals a role for tumor necrosis factor in peripheral nerve development and function. *Proc Natl Acad Sci U S A.* 2009;106:12494–12499.

30. Liodaki E, Bos I, Lohmeyer JA, et al. Removal of collagen nerve conduits (NeuraGen) after unsuccessful implantation: Focus on histological findings. *J Reconstr Microsurg.* 2013;29:517–522.

31. Hanwright PJ, Rath JB, von Guionneau N, et al. The effects of a porcine extracellular matrix nerve wrap as an adjunct to primary epineurial repair. *J Hand Surg Am.* 2021;46:813.e1–813.e8.

32. Saheb-Al-Zamani M, Yan Y, Farber SJ, et al. Limited regeneration in long acellular nerve allografts is associated with increased Schwann cell senescence. *Exp Neurol.* 2013;247:165–177.

33. Waitayawinyu T, Parisi DM, Miller B, et al. A comparison of polyglycolic acid versus type 1 collagen bioabsorbable nerve conduits in a rat model: An alternative to autografting. *J Hand Surg Am.* 2007;32:1521–1529.

34. Pertici V, Laurin J, Féron F, et al. Functional recovery after repair of peroneal nerve gap using different collagen conduits. *Acta Neurochir (Wien).* 2014;156:1029–1040.

35. Kemp SW, Syed S, Walsh W, et al. Collagen nerve conduits promote enhanced axonal regeneration, schwann cell association, and neovascularization compared to silicone conduits. *Tissue Eng Part A.* 2009;15:1975–1988.