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The Effect of Injurious Compression on the Elastic, Hyper-elastic and Visco-elastic Properties of Porcine Peripheral Nerves.

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Abstract

The aim of this study was to characterise the viscoelastic and hyper-elastic properties of the ulnar nerve before and after compression has been induced, in order to aid the understanding of how the mechanical properties of nerves are altered during nerve compression, a contributing factor to cubital tunnel syndrome. Ulnar nerves were dissected from porcine legs and tensile tested to 10% strain. The Young's modulus and Yeoh hyper-elastic model were used to evaluate the materials elastic and hyper-elastic properties respectively. Dynamic mechanical analysis (DMA) was used to evaluate the viscoelastic properties over a range of frequencies between 0.5 Hz and 38 Hz. The nerves were then compressed to 40% for 60 seconds and the same tests were carried out after compression. The nerves were stiffer after compression, the mean Young's modulus before was 0.181 MPa and increased to 0.601 MPa after compression. The mean shear modulus calculated from the Yeoh hyper-elastic model was also higher after compression increasing from 5 kPa to 7 kPa. After compression, these properties had significantly increased (p <0.05). The DMA results showed that the nerves exhibit frequency dependent viscoelastic behaviour across all tested frequencies. The median values of storage modulus before compression ranged between 0.605 to 0.757 MPa across the frequencies and after compression between 1.161 MPa to 1.381 MPa. There was a larger range of median values for loss modulus, before compression, median values ranged between 0.073 MPa to 0.216 MPa and after compression from 0.165 MPa to 0.410 MPa. There was a significant increase in both storage and loss modulus after compression (p <0.05). The mechanical properties of the nerve change following compression, however the response to decompression in vivo requires further evaluation to determine whether the observed changes persist, which may have implications for clinical recovery after surgical decompression in entrapment neuropathy.

Keywords: Cubital Tunnel Syndrome, Dynamics Mechanical Analysis, Elasticity, Hyper-elastic, Material Properties, Ulnar Nerve, Viscoelastic

1. Introduction

The ulnar nerve is a major peripheral nerve of the upper limb and one of the most frequently injured nerves of the upper limb (1-3). It can become trapped or impinged, which leads to compression or irritation of the nerve. The most common place for compression of the ulnar nerve is in the cubital tunnel, on the posteromedial aspect of the elbow, resulting in cubital tunnel syndrome (CTS). The ulnar nerve is especially vulnerable to compression between the superficial Osborne's fascia and the underlying posterior aspect of the medial epicondyle of the elbow. The tunnel is constrained with little soft tissue cover (4). While pain may sometimes occur, common symptoms of CTS include hypoaesthesia, paraesthesia and loss of motor function in the ulnar digital flexors and the ulnar-innervated intrinsic muscles of the hand. Compression was originally regarded as the principal mechanism of nerve damage in peripheral neuropathy (2). However, it has recently been accepted that this mechanism is multifunctional with compression, entrapment, excursion restraint and strain all hypothesised as contributing to CTS (5).

Peripheral nerves under tension exhibit non-linear stress-strain behaviour (6-8). Nerves have been characterised as both hyper-elastic (6,9-11) and visco-elastic (6,12-14). One study determined values for the shear modulus for an Ogden hyper-elastic model *in vivo* as 54 kPa, but with a lower *in vitro* value of 12 kPa (6) consistent with median nerve shear moduli in literature of 12.9 kPa (9). Visco-elasticity is a combination of elastic and viscous properties (15). Visco-elastic studies have primarily focused on creep and stress relaxation testing (12,13). However, a recent study has used Dynamic Mechanical Analysis (DMA) to assess the dynamic visco-elasticity of ulnar nerves (14). The advantage of DMA is that visco-elastic behaviour can be characterized at physiologically relevant loading rates and frequencies.

Changes in nerve biomechanics after a crush injury have been documented in mice (16). The results concluded that nerve stiffness changes with time following crush injury. Mechanical testing showed damaged nerves had approximately the same strength as the normal nerves, but they exhibited approximately 6% greater stiffness. In patients, median nerve elasticity (referring to strain) was altered during carpal tunnel syndrome; carpal tunnel syndrome is a similar condition to CTS (17). However,

studies on the median nerve in carpal tunnel syndrome have conflicting conclusions, some studies reporting reduced strain in patients with carpal tunnel syndrome (18,19) and others reporting increased strain in damaged nerves (20); measured using elastography based techniques. The study concluding increased strain in median nerves for patients with carpal tunnel syndrome was only conducted on pregnant women, which might explain differing results as compared to other studies. Increased strain in the nerves of pregnant women could be explained by studies which conclude that connective tissues within pregnant women become more extensible during pregnancy (21,22).

It is currently unclear how the mechanics of the nerve is altered following an injurious compression, including elastic, hyper-elastic and visco-elastic behaviour. However, there are potential applications of such knowledge to techniques for performing nerve repair (23). For example, to better qualify limitations of using healthy nerves when assessing surgical techniques or for developing more realistic models (24). Additionally, there are potential applications and implications in the use strain as a measurement of success following CTS (25).

The aim of this study is to characterise the material properties of porcine peripheral nerves before and after injurious compression. *Ex vivo* porcine nerves have been characterised in terms of elastic and hyper-elastic behaviour from data obtained through quasi-static materials' testing. Subsequently, visco-elastic properties have been characterised using DMA over a frequency-sweep. Materials characterisation has taken place before and after the use of a repeatable technique to induce an acute compressive injury.

2. Methodology

2.1 Specimens and Dissection

Two female porcine hind limbs, both under twelve months of age were obtained (Dissect Supplies UK, Kings Heath, Birmingham, UK), from which 14 upper limb nerve tissue samples were excised. To dissect the nerves, the brachial plexus was identified at the top of the porcine shoulder and was followed distally. Smaller nerves close to the surface of the limb were dissected initially, these contained several branches (Figures 1a and 1b) and were therefore not used for testing. The nerves typically followed the same path as arteries which were followed during dissection, leading to larger major nerves found deeper within the limb (Figures 1c and 1d). The nerves were excised by dissecting them from surrounding tissue and fascia, with excess tissue removed. On removal from the limb, the nerves were approximately 280 mm in length. These nerves were then divided into test samples approximately 40 mm in length. In total, 14 test samples were obtained.

Test samples were dissected to 40 mm, to include a 20 mm gauge length (L) for material's testing and characterisation (Section 2.4), with 10 mm used for clamping both ends of the sample onto the test machine (6,14). Measurements were made using a manual Vernier calliper to ensure each sample was of equal length. The cross-sectional area of each sample was calculated as an ellipse (14). The elliptical cross-sectional area (A) equation is given in Equation 1, where a is the major radius and b is the minor radius. Once dissected, the samples were wrapped in Ringer's solution soaked tissue paper, placed in heat sealed plastic bags and all frozen at -40℃ until testing was carried out.

$$A = \pi ab \qquad \textbf{Equation 1}$$

2.2 Mechanical Testing

2.2.1 Testing procedure

Test samples were thawed for testing. Subsequently, each sample was soaked in Ringers solution 15 minutes prior to testing. Additionally, test samples were hydrated during testing by wrapping a thin layer of tissue paper soaked in Ringers solution around the sample; a method often used for keeping biological tissues hydrated (24,26-29).

All mechanical testing was performed using a BOSE ElectroForce 3200 materials testing machine (Bose Corporation, ElectroForce Systems Group, Minnesota, USA; now TA Instruments), controlled using WinTest 4.1 software. Mechanical testing in order of testing included: Tensile tests (Section 2.2.2) and DMA (Section 2.2.3) for materials characterisation as well as compression tests to mimic injury (Section 2.2.4). Test samples were characterised for elastic, hyper-elastic, and visco-elastic properties before and after acute injurious compression (Section 2.3).

2.2.2 Quasi-static testing

Tensile ramp tests were carried out for elastic and hyper-elastic material characterisation, each test was performed ten times on each sample of which the final 4 tests were used for analysis. Test samples were gripped by two clamps, with the lower clamp fixed to a 225 N load cell and the upper clamp fixed into an actuator (Figure 2). A preload of 0.05 N was applied before quasi-static testing began, consistent with previous studies (9). Ramp tests were run at 0.05 mm/s (6,14) and to a displacement of 2 mm, i.e. 10% strain, for consistency with previous studies (6) and to avoid permanent damage (7). Raw data in the form of Force (F) and displacement (Δl) were obtained from which stress (σ) and strain (ε) were calculated (Equation 2 and Equation 3, respectively) from the sample's gauge length (L) and cross-sectional area (A); see section 2.1.

$$\sigma = \frac{F}{A} \qquad \textbf{Equation 2}$$

$$\varepsilon = \frac{\Delta l}{L} \qquad \textbf{Equation 3}$$

2.2.3 Dynamic Mechanical Analysis (DMA)

DMA was used to characterise dynamic viscoelasticity over a frequency sweep. Sinusoidal displacement loading was induced between 2-8% strain, over a frequency range of 0.5 – 38 Hz (Table 1). The range of strain is below that at which damage is expected to occur in the nerve (6) and within a linear data range. The frequency sweep was selected to characterise test samples from typical everyday activities

(0.5 Hz) up to frequencies (38 Hz) which correspond to expected peak nerve strain rates for nerves under fast movement or exercise (8,27,30). The focus being on activities associated with the median nerve within the carpal tunnel (31). A preconditioning loading cycle was used (24) to produce repeatable material properties (32-34). Preconditioning was run at 1 Hz for 20 cycles.

2.2.4 Compression Testing

After tensile ramp testing and DMA, a compression ramp test was carried out on each sample. The compression test was designed to mimic the pressure the ulnar nerve experiences within the body for a patient with CTS. A compression plate was attached to the actuator of the material's testing machine, with the nerve sample supported on a second plate attached to the load cell (Figure 3). The compression plate had a diameter of 30 mm, ensuring the 20 mm test area of the sample was completely covered during the compression test ensuring uniform compression. The compression test was carried out in displacement control at a rate of 0.05 mm/s (35).

Test samples were compressed to a strain of 40% (Equation 3) and held for 60 s. Injurious compression of 40% strain was chosen following preliminary testing where a range of 30-70% compression was evaluated (Table 2); matching the compression reported clinically for CTS (36). An injurious acute compression of 40% was the lowest strain which led to repeatable data, in addition to being comparable to previous studies focused on the compression of peripheral nerves (9). Following injurious compression, leading dimensions of test samples were re-measured. The average cross-sectional area decreased after compression from 59.96 mm^2 to 53.31 mm^2 (Table 3). Quasi-static and DMA tests were repeated as described in Sections 2.2.2 and 2.2.3.

2.3 Material Characterisation

2.3.1 Elastic Characterisation

The Young's modulus (E) was evaluated before and after compression at 8% strain (Equation 4). Additionally, both pre- and post-transitional moduli were calculated (28).

$$E = \frac{\sigma}{\epsilon} \qquad \textbf{Equation 4}$$

2.3.2 Hyper-elastic Characterisation

Hyper-elasticity was evaluated using the material evaluation tool within the finite element analysis software ABAQUS (ABAQUS 2017, Dassault Systems, Providence, RI, USA). The stress-strain data calculated from the tensile ramp testing was inputted into ABAQUS, and characterised with hyper-elastic models (37). Neo-Hookean, Yeoh and Ogden models were compared, with the Yeoh model (Equation 5) providing the best fit for the data and remaining stable for the full data-sets evaluated (Figure 4).

$$U = \sum_{i+j=1}^{N} C_{ij} (\overline{I_1} - 3)^i (\overline{I_2} - 3)^j + \sum_{i=1}^{N} \frac{1}{D_i} (\overline{J_{e1}} - 1)^{2i} \qquad \textbf{Equation 5}$$

$$\mu_0 = 2 (C_{10} + C_{01}) \qquad \textbf{Equation 6}$$

$$K_0 = \frac{2}{D_1} \qquad \textbf{Equation 7}$$

Where U is the strain energy potential, described in terms of a strain energy potential, $\overline{J_{e1}}$, which describes the elastic volume strain with $\overline{I_1}$ and $\overline{I_2}$ being the first and second strain invariants, respectively (37). C_{10} describes the shear behaviour of the material (Equation 6) where μ_0 is the shear modulus and D_i introduces compressibility, from which bulk modulus (K_0) can be calculated (Equation 7).

2.3.3 Visco-elastic Characterisation

The WinTest software used for DMA calculates the phase angle (δ) between the force and the displacement (d) from the load cell and the displacement transducer. From this, the storage (E') and loss (E'') modulus are calculated, where E' characterises the material's ability to store energy and E'' the material's ability to dissipate energy (38). Further details on DMA are available elsewhere (39). Briefly, a fast Fourier transform is used to determine the magnitude of the force (F^*) and the displacement (d^*) from which the dynamics stiffness (k^*) is calculated (Equation 8).

$$k^* = \frac{F^*}{d^*} \qquad \textbf{Equation 8}$$

An elliptical shape factor (S_c) shown in Equation 9 is then used to calculate E' (Equation 10) and E'' (Equation 11). Where *h* is the length of the specimen and *a* and *b* are the parameters defined for Equation 1.

$$S_c = \frac{\pi}{h} ab \qquad \textbf{Equation 9}$$

$$E' = \frac{k^* \cos \delta}{S_c} \qquad \textbf{Equation 10}$$

$$E'' = \frac{k^* \sin \delta}{S_c} \qquad \textbf{Equation 11}$$

$E'$and E'' are related to the complex modulus (E^*) and δ according to Equations 12 and 13.

$$E^* = \sqrt{E'^2 + E''^2} \qquad \textbf{Equation 12}$$

$$\delta = tan^{-1}\left(\frac{E''}{E'}\right) \qquad \textbf{Equation 13}$$

To evaluate the data across the frequency range, regression analysis was performed for both E' and E'' A logarithmic was used to empirically evaluate the relationship between E'' and the test frequency. E' was not found to vary with frequency, *f*, and so it was evaluated as a constant.

$$E'' = Aln(f) + B \qquad \textbf{Equation 14}$$

2.4 Statistical Analysis

Statistical analysis was performed using Sigma Plot 13.0 (SYSTAT, San Jose, CA, USA). Data obtained following elastic and hyper-elastic characterisation, before and after injurious compression, were assessed using Paired T-tests normally distributed data-sets. If a data-set was not normally distributed, then a Wilcoxon signed rank test was performed. Normality was assessed using a Shapiro-Wilk test ($p < 0.05$).

A Friedman repeated measure analysis of variance (ANOVA) on ranks was performed on the data before and after compression across the range of frequencies used for DMA for both the storage and loss moduli. If the Friedman test showed a significant difference between the groups ($p < 0.05$), a Tukey multiple comparison test was used to determine the differences between the before and after groups. Tests were carried out at 95% confidence levels to test the variance of the results and assess whether material properties before and after compression were significantly different ($p < 0.05$).

3 Results

3.1 Quasi-static testing

Stress-strain curves of the tensile tests are provided in Figure 5. At 8% strain, the stress induced in the healthy nerves was 14.5 kPa, this increased significantly to 48.1 kPa following injurious compression ($p < 0.05$). There was variability across the nerve samples tested (Figures 5c and 5d), but all test samples experienced increased stress after compression. There was an increase in the mean Young's modulus at 8% strain ($p < 0.05$) following compression (Figure 6). Likewise, there was a significant increase in both the pre- and post-transitional moduli following compression ($p < 0.05$; Table 4).

Material properties taken from the Yeoh hyper-elastic model are provided in Table 5. The mean values for parameters C_{10}, C_{20} and C_{30} all increased significantly ($p < 0.05$) after compression. Values for C_{01}, C_{02} and C_{03} were zero for all test samples before and after compression. The shear modulus increased significantly following compression ($p < 0.05$; Table 6).

3.2 Visco-elasticity

All test samples displayed visco-elastic behaviour across the frequency-sweep, with the values for E'' being consistently lower than those for E'. Figure 7 shows the viscoelastic response of the nerves before and after compression, with both E' and E'' increasing significantly ($p < 0.05$) after compression.

The median values of E' before compression ranged between 0.61 MPa to 0.76 MPa across the frequencies tested. Following compression this range increased to 1.16 to 1.38 MPa. Overall, E' increased from 0.81 MPa before compression, up to 1.44 MPa after compression. This is an overall increase of 77%.

The median values for E'' before compression were between 0.073 MPa to 0.22 MPa increasing after compression from 0.17 MPa to 0.41 MPa. After compression all but one sample (sample 7) followed the logarithmic fit ($p < 0.05$; Table 7). Both constants *A* and *B* (Equation 14) increased after compression ($p < 0.05$), demonstrating an increased offset and increased gradient of E'' with frequency.

4. Discussion

To the authors' knowledge, this is the first study to characterise the mechanical behaviour of nerves before and after acute injurious compression in terms of elastic, hyper-elastic and viscoelastic properties. It has been found that following injurious compression nerves demonstrate: an increase in Young's moduli at 8% strain; an increase in pre- and post-transitional moduli; increase in parameters which define hyper-elasticity (using a Yeoh model); and increased storage and loss moduli (describing its viscoelastic behaviour). Ultimately, these results demonstrate an increase in stress induced within a nerve, per given strain, following acute injurious compression.

Nerves have been shown to display different mechanical properties following injurious compression. This is clinically relevant because there could be the potential to measure strain in nerves before and after CTS has been performed (40). Our current study has highlighted the difference in the mechanical properties of healthy and injured nerves, this has implications for using nerve strain as a surrogate for stress, and thereby tension in nerves during clinical assessment (e.g. reducing strain across a repair region to enhance axon regeneration). For instance, the crushing of porcine nerves led to an increase in the average mean tension per strain from 13.6 N to 26.5 N. Hence, the tension through an otherwise healthy nerve would be much lower than that through a nerve injured following compression, at a given strain. It also implies inherent limitations in using healthy nerves during *in vitro* studies for assessing corrective surgery, as their mechanical response to loading will differ to that of an already injured nerve tissue. An open question which remains during long-term damage is: how do healing and scar formation within a nerve (e.g. paraneurial tissues) alter the stress-strain relationship?

As highlighted above, an implication, from a clinical perspective, is that tension as measured in a healthy nerve would differ, per strain, as compared to a nerve which has undergone injury; hence, evaluation of surgical techniques aimed at reduced nerve tension may need to account for how tension may differ according to any change to the material properties of the nerve. It is unclear, however, how acute compressive injury might alter the electrophysiological function of the nerve. One hypothesis, requiring further investigation, might be that the travel of an action potential through the axon is impaired, at strains at which axons become damaged. Additionally, it would be useful to assess which

structures (e.g. epineurium, perineurium, endoneurium) become damaged during compressive injury and determine whether they correspond to the changes in material properties identified in this current study.

The Young's modulus measures the resistance of a material to recoverable deformation under a load. After the nerves were compressed, they all displayed an increased Young's modulus at 8% strain. Hence, after injurious compression nerves experienced changes in length under greater tension due to their increased material stiffness. This finding is in line with other studies on peripheral nerves where an increase in stiffness after compression or within patients with carpal tunnel syndrome was observed (19,21,22). From the hyper-elastic Yeoh model, the shear modulus was found to increase after compression of the nerve. This is representative of a more rigid material, with a larger force required to produce a change in shape (38). The Yeoh model assumes the nerves to be non-compressible, reflected by the D_1 values equalling zero, an assumption was in line with other studies (39). The shear modulus increase was consistent with the increase in Young′s modulus following injurious compression, implying more force was required to both lengthen and deform the nerves.

The dynamic visco-elasticity of nerves, as measured in this study, are qualitatively in agreement with previous studies. For instance, the storage modulus was much greater than the loss modulus (14). However, there was a difference in the values measured. In our study otherwise healthy nerves had storage moduli in the range 0.6 to 0.8 MPa; rising to 1.4 MPa following injurious compression. This compares to storage moduli in the range of 10 MPa reported in one study in literature (14). Loss moduli between this current study and the study in literature (14) followed a similar logarithmic trend, however, the values in this current student were again lower than that reported previously. There are three main factors in the difference in values measured. Firstly, the only previous study to report dynamic viscoelastic properties of nerves did so on human nerves, as compared to porcine nerves in this study. Secondly, in the previous study nerves were embalmed, which was not the case for this current study. Finally, as ageing may affect the material properties of soft tissues (29), it is worth noting that the age of donors for the previous study with human samples ranged from 75-90 years of age. This compares to porcine samples in this current study aged below 12 months. Regardless of the difference in quantitative

values measured, the similarity in trends with frequency suggest that compressive injury would lead to an increase in storage and loss moduli of human nerves (as well as porcine nerves).

There are limitations for *in vitro* testing of nerves as biological materials may not display the same material properties outside the body as they would within the body. *In vivo* a stress of 58 kPa has been measured for nerves at 4.1% strain, however, this stress was over seven times greater than that measured *in vitro* at the same strain (8.2 kPa) (6). In our study, mean stress of 1.7 kPa was found at 4% strain. This highlights the potential difference in biomechanical properties of nerves *in situ* as compared to *ex vivo* where tensegrity of the tissue may be one factor the measured values, potentially altering the tissue pre-load. This difference between *in vitro* stress at 4% in this study with previous studies may be due to the different samples used, as the samples used in this project were porcine nerves rather than human ulnar nerves (6). Furthermore, there may be intrinsic differences between nerves in a human arm as opposed to a porcine limb. Although there are differences, porcine nerves offer a reasonable representation of human nerves *in vitro*, with other studies suggesting enough similarity between porcine and human nerves to propose the clinical use of porcine nerves as grafts following nerve injury (41). Furthermore, while there may be differences in material properties measured *in situ* and *ex vivo*, the trend of an increase in induced stress per given strain would still be expected in both cases.

The method of crush utilised in this study is a good model for acute injury but more limited in terms of longer-term changes which a nerve might experience during chronic injury. Such assessment would require further assessment of chronic compression. Our suggestion from this study is that it would be important to assess the dynamic visco-elastic and hyper-elastic (e.g. Yeoh model) properties of nerve injury following chronic compression. Developing chronic measurements of visco-elastic and hyper-elastic properties of compressed nerves will aid the further understanding of their behaviour when subjected to compression within the cubital tunnel. It may be the link between strain and tension which is of most clinical relevance.

CONCLUSION

This study has demonstrated that nerves exhibit frequency dependent visco-elastic properties and hyper-elastic properties before and after compression. Compressed nerves were found to be stiffer in terms of increased stress per strain using elastic and hyper-elastic models, as well as an increased ability to store and dissipate energy (as measured via storage and loss moduli). Hence, acute crush injuries alter the mechanical properties of nerve tissue.

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ABBREVIATION LIST (in order of appearance within text)

A	elliptical cross-sectional area
a	major radius of ellipse
b	minor radius of ellipse
F	Force
Δl	displacement
σ	stress
ε	strain
L	sample's gauge length
E	Young's modulus
U	strain energy potential
$\overline{J_{el}}$	elastic volume strain
$\overline{I_1}$	first strain invariant
$\overline{I_2}$	second strain invariant
C_{10}	describes the shear behaviour (see Equation 6)
μ_0	shear modulus
D_i	compressibility
$K_0)$	bulk modulus
δ	phase angle between force and displacement (during DMA tests)
d	displacement (from displacement transducer, during DMA tests).
E'	storage modulus
E''	loss modulus
F^*	magnitude of the force (following fast Fourier transform of load data from DMA tests)
d^*	magnitude of the displacement (following fast Fourier transform of displacement data from DMA tests)
k^*	dynamics stiffness
S_c	elliptical shape factor
f	frequency
h	length of the specimen (equivalent to L)
A	Empirically derived coefficient (see Equation 14)
B	Empirically derived coefficient (see Equation 14)

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TABLES

Table 1. DMA frequencies range tested for each sample.

Condition	Frequency (Hz)
1	1.00
2	0.50
3	5.00
4	12.00
5	15.00
6	20.00
7	25.00
8	38.00
9	1.00

Table 2. Preliminary compression testing values and hold time.

Sample	Compression (%)	Hold time (s)
1	30	30
2	40	30
3	70	30
4	30	60
5	40	60
6	70	60

Table 3: Measured nerve dimensions before and after compression.

Sample	Pre Compression			Post Compression		
	a (mm)	***b (mm)***	$A\ (mm^2)$	***a (mm)***	***b (mm)***	$A\ (mm^2)$
1	5.10	1.90	30.44	5.80	1.10	20.04
2	4.30	2.10	28.37	6.40	1.20	24.13
3	7.30	4.20	96.32	8.40	2.90	76.53
4	5.10	3.20	51.27	5.20	2.80	45.74
5	6.60	5.40	111.92	9.70	2.10	63.90
6	9.60	3.30	99.52	11.20	3.40	119.63
7	9.10	5.20	148.66	9.80	4.80	147.78
8	4.80	3.10	46.75	5.70	2.60	46.56
9	6.10	2.60	49.83	6.80	1.90	40.59
10	7.20	4.20	95.00	9.10	3.20	91.48
11	5.00	2.10	32.98	6.40	1.60	30.66
12	3.40	1.50	16.02	4.10	1.30	16.74
13	3.60	2.10	23.75	3.90	1.20	14.70
14	2.30	1.20	8.670	2.50	1.00	7.85
Mean	**5.68**	**3.01**	**59.96**	**6.78**	**2.22**	**53.31**
Median	**5.10**	**2.85**	**48.29**	**6.40**	**2.00**	**43.17**
ST Dev	**2.11**	**1.32**	**42.52**	**2.54**	**1.11**	**42.12**

Table 4. Pre- and post-transitional moduli before and after compression.

Sample	Pre-transitional Moduli (MPa)		Post-transitional Moduli (MPa)	
	Pre Compression	**Post Compression**	**Pre Compression**	**Post Compression**
1	0.31	0.48	1.56	2.22
2	0.14	0.44	1.27	1.85
3	0.09	0.16	0.65	0.87
4	0.14	0.33	0.99	1.26
5	0.05	0.23	0.36	1.19
6	0.04	0.07	0.13	0.21
7	0.02	0.05	0.20	0.34
8	0.23	0.37	1.39	1.69
9	0.15	0.30	0.92	0.99
10	0.07	0.37	0.58	1.09
11	0.18	0.33	0.86	1.02
12	0.52	1.38	3.15	4.93
13	0.22	0.42	1.69	3.77
14	0.53	1.68	4.78	8.98
Mean	**0.19**	**0.47**	**1.32**	**2.17**
Median	**0.14**	**0.35**	**0.96**	**1.22**
ST Dev	**0.16**	**0.47**	**1.26**	**2.35**

Table 5. Values for coefficients of the Yeoh hyper-elastic model before and after compression.

Sample	$C_{10} \times 10^{-3}$ (N/m^2)		C_{20} (N/m^2)		C_{30} (N/m^2)	
	Pre Compression	Post Compression	Pre Compression	Post Compression	Pre Compression	Post Compression
1	5.89	21.54	0.12	0.17	30.78	75.97
2	11.72	24.75	-0.85	1.11	45.94	25.76
3	1.62	4.37	0.06	0.01	18.73	41.85
4	10.36	15.27	-0.57	1.03	29.60	22.17
5	4.18	10.56	-0.23	-0.20	12.02	43.56
6	8.34	14.06	-0.22	0.05	7.65	5.57
7	3.22	4.23	-0.10	-0.16	6.19	12.03
8	9.09	12.54	-0.09	0.08	33.60	46.98
9	6.71	8.95	-0.16	1.04	24.17	22.36
10	2.58	5.09	-0.08	0.06	18.54	42.86
11	6.49	9.63	0.04	0.80	24.49	46.97
12	12.44	16.27	-0.62	4.48	97.42	75.93
13	13.60	18.68	-0.97	3.74	51.19	97.78
14	48.36	20.75	-3.56	5.20	170.00	196.95
Mean	**10.33**	**13.34**	**-0.52**	**1.24**	**40.74**	**54.05**
Median	**8.34**	**13.34**	**-0.19**	**0.49**	**27.04**	**43.21**
ST Dev	**11.57**	**6.60**	**0.94**	**1.83**	**43.82**	**48.61**

Table 6. Shear modulus before and after compression.

Sample	**Shear Modulus × 10^{-3} (N/m^2)**	
	Pre Compression	**Post Compression**
1	11.78	43.07
2	23.43	49.50
3	3.23	8.75
4	20.72	30.53
5	8.36	21.13
6	16.69	28.11
7	6.44	8.47
8	18.17	25.08
9	13.41	17.90
10	5.16	10.19
11	12.98	19.26
12	24.88	32.54
13	27.20	37.36
14	96.71	41.49
Mean	**20.65**	**26.67**
Median	**16.69**	**26.67**
ST Dev	**23.14**	**13.20**

Table 7. Logarithmic fitting values for *E''* before and after compression.

Sample	A (N/m^2)		B (N/m^2)		R^2	
	Pre Compression	Post Compression	Pre Compression	Post Compression	Pre Compression	Post Compression
1	0.05	0.08	0.15	0.20	0.84	0.81
2	0.04	0.06	0.08	0.10	0.88	0.88
3	0.03	0.03	0.07	0.06	0.85	0.86
4	0.06	0.09	0.15	0.12	0.82	0.81
5	0.03	0.03	0.08	0.07	0.95	0.85
6	0.01	0.01	0.04	0.04	0.92	0.89
7	0.01	0.00	0.02	0.04	0.93	0.12
8	0.09	0.06	0.04	0.17	0.86	0.78
9	0.03	0.01	0.07	0.03	0.89	0.93
10	0.03	0.05	0.07	0.25	0.84	0.67
11	0.04	0.12	0.11	0.27	0.85	0.84
12	0.12	0.23	0.27	0.59	0.84	0.75
13	0.03	0.04	0.07	0.84	0.92	0.80
14	0.05	0.06	0.15	0.39	0.87	0.76
Median	**0.03**	**0.05**	**0.08**	**0.15**	**0.86**	**0.81**
Mean	**0.04**	**0.06**	**0.10**	**0.22**	**0.88**	**0.77**
ST Dev	**0.03**	**0.06**	**0.06**	**0.24**	**0.04**	**0.20**

FIGURE CAPTIONS

Figure 1. Dissection and excision of peripheral nerves. (a) Small branched nerves near the surface of porcine leg. (b) Nerve and artery shown together following same path into centre of specimen. (c) and (d) Large main ulnar nerve of porcine.

Figure 2. Tensile testing set up with nerve test sample gripped in place.

Figure 3. Compression testing set up used to induce acute injury.

Figure 4. Experimental data from test sample 4, post-compression, fitted with four hyper-elastic material models.

Figure 5. A comparison of stress-strain for porcine peripheral nerves before and after injurious compression. (a) Stress-strain curves for all nerve samples before compression. (b) Stress-strain curves for all nerve samples after compression (c) Stress-strain curve before and after compression for test sample 9. (d) Stress-strain curve before and after compression for test sample 12.

Figure 6. Young's modulus for all test samples before and after injurious compression.

Figure 7. Frequency-dependent visco-elasticity of peripheral porcine nerves. (a) Frequency-dependent storage modulus before and after compression (median ± 95% confidence intervals). (b) Frequency-dependent loss modulus before and after compression (median ± 95% confidence intervals).