# Viscoelastic & Mechanical Properties of the Carpal Tunnel Complex and Overlying Soft Tissue Layers



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#### Abstract

**Background:** Carpal Tunnel Syndrome (CTS) is a form of compression neuropathy of the median nerve as it passes through the carpal tunnel of the wrist. CTS is characterised by pain and paresthesia of the palm of the hand and the lateral three and a half fingers, muscular atrophy of the thenar and hypothenar muscles, leading to decreased grip and pinch strength, and decreased hand function. Severe and chronic cases of CTS often require open or endoscopic release of the transverse carpal ligament (TCL) for relief of carpal tunnel pressure. Techniques to stretch the TCL have been proposed but further understanding of the viscoelastic properties of the carpal tunnel complex is required.

**Methods:** Six embalmed cadaveric hands  $(82 \pm 6.29 \text{ yrs})$  were tested in confined compression using a 10 mm diameter indenter at different levels of dissection (Intact, Skin and Adipose removed, thenar or hypothenar removed, and TCL exposed). Mean peak load, Load relaxation and Stiffness were recorded and compared between dissection levels using paired and unpaired t-tests.

**Results:** 'Skin removed' condition relaxed significantly quicker than 'Intact'. No other significant differences were found for load relaxation. Peak loads and stiffness generally increased with removal of each tissue layer. Significant differences were found for 'Intact' vs. 'TCL exposed' and the removal of the thenar muscle group resulted in greater peak loads and stiffness compared to removal of the hypothenar muscle group.

**Conclusion:** The skin and adipose layer ('Intact' condition) of the hand had the slowest relaxation times; while the muscular layer (Thenar muscle group) showed the fastest load relaxations, and greatest load absorption. Peak loads and stiffness generally increases for each layer of tissue removed. Testing at the 'TCL exposed' level with the test protocol used here does not isolate the TCL adequately to be able to attribute any mechanical or viscoelastic characteristics.

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#### **1** Introduction

Carpal Tunnel Syndrome is a form of compression neuropathy of the median nerve as it passes through the carpal tunnel of the wrist. It is a painful and debilitating condition affecting approximately 4 to 12 people per 10000 of the UK population each year and is more prevalent in females than in males, with females accounting for approximately two thirds of total CTS cases, and more common in the over 40 age groups (NHS, 2010). CTS is characterised by pain, tingling, numbness and burning sensation of the lateral three and a half fingers, and palm of the hand; muscular atrophy of the thenar and hypothenar muscles, leading to decreased grip and pinch strength, and decreased hand function (Mathura *et al*, 2004; Gellman *et al*, 1989).

There are several possible causes of CTS including: tendonitis, tenosynovitis, bursitis, lesions and inflammation of the transverse carpal ligament. The effect of these is an increase of pressure within the carpal tunnel; compressing and impinging the median nerve.

Various conservative treatments may be prescribed to address the underlying cause of the condition, such as physical therapy, rest, nocturnal wrist splinting, nonsteroidal anti-inflammatory drugs and corticosteroid injections. Surgical intervention to relieve carpal tunnel pressure is favoured where conservative methods have been unsuccessful or symptoms have persisted for six months or more. There are, predominantly, two surgical methods that are used to accomplish this: open-carpal release and endoscopic release, both with the end goal of increasing the available space within the carpal tunnel.

These methods involve complete transaction of the transverse carpal ligament. Open release surgery is the more traditional method and involves a 2 inch incision being made longitudinally over the palm and wrist with complete transaction of the palmar aponeurosis, flexor retinaculum and the transverse carpal ligament. The endoscopic release involves one to two incisions being made: one at the proximal wrist crease and another at the mid palm level. An endoscopic camera and an open top tube are inserted under the TCL, and a retrograde knife is used to transect the TCL only (Aroori and Spence, 2008).

Due to the invasive nature of these procedures complications are a risk and can include infection, damage to the median nerve, soft tissue fibrosis, stiffness and pain at the scar, loss of grip and pinch strength (Mathura *et al*, 2004), and wrist biomechanical abnormality (Kiritsis and Kline, 1995).

The risk of these complications has prompted the design of an alternative method for decompressing the carpal tunnel that avoids transection of the TCL so that wrist biomechanics and integrity are not unduly compromised. Berger (1993), proposed using a balloon catheter device inserted into the carpal tunnel and serially inflating and deflating the device to expand the carpal tunnel by stretching the TCL. Little research has been conducted into the efficacy of such a procedure and in particular the ability of the TCL to permanently elongate. Studies by Sucher (2005), Li *et al* (2009, 2011) and Holmes *et al* (2011) have investigated the mechanical properties of the TCL but do not investigate the viscoelastic properties of the TCL. Furthermore, little research has been conducted (Zheng *et al*, 2006) into the mechanical and viscoelastic properties of the Soft tissue layers superficial to the TCL.

#### **1.1 Research Objectives**

The purpose of this study is therefore; to determine the mechanical and viscoelastic properties of the transverse carpal ligament and superficial soft tissue layers in the direction perpendicular to the palmar surface.

#### **1.2 Background Information**

#### **1.2.1 Basic Anatomy of the Carpal Tunnel Complex**

The carpal tunnel is a narrow passageway at the base of the hand comprised of a concave arch formed by the carpal bones to the posterior and sides with the TCL forming the roof on the palmer aspect. The carpal tunnel provides a conduit for the flexor tendons of the forearm and the median nerve. Annotated drawings of the wrist can be found in figures 1 and 2 at the end of this chapter.

#### **1.2.2 Skeletal Structure and Joints**

The hand and wrist is comprised of 27 bones: 8 carpals, 5 metacarpals and 14 phalanges. At the wrist the 8 bones of the carpus are divided into a proximal row and distal row (4 in each). The proximal row consists of the scaphoid, lunate and triquetral; with the pisiform sitting anteriorly to triquetral. The proximal ends of scaphoid and lunate articulate with the distal ends of the radius and ulnar to form the radiocarpal joint; while the distal end articulates with the proximal border of the distal carpal row. The distal row of the carpus is comprised of the trapezium, trapezoid, capitate and hamate and they articulate distally with the 5 metacarpals at the carpometacarpal joints (CMC). The trapezium articulates distally with the base of the first metacarpal; trapezoid with the second metacarpal; capitate with third and fourth metacarpals, and hamate articulates distally with fifth metacarpal. The 5 metacarpals each go on to articulate with the proximal phalanges of the fingers.

The carpus also forms a palmarly concave arch, called the carpal tunnel; walled on the ulnar side by the pisiform and hook of hamate, and on the radial side by the scaphoid and trapezium. The intercarpal joints are supported by strong intercarpal ligaments which provide stability and restricted intercarpal motion.

The transverse carpal ligament spans the 'roof' of the carpal tunnel; inserting radialy into the tubercle of scaphoid and the ridge of the trapezium; and ulnarly into the hook of hamate, the pisiform and pisohamate ligament.

#### **1.2.3 Muscles and Tendons**

**Extrinsic:** The carpal tunnel provides a conduit for the passage of 9 flexor tendons (within the common flexor synovial sheath) and the median nerve from the forearm to the hand. The flexor tendons consist of: 4 tendons of flexor digitorum profundus, 4 tendons of flexor digitorum superficialis (encased in common ulnar sheath) and flexor pollicis longus (in radial sheath).

The tendon of flexor carpi ulnaris inserts into the base of the  $5^{\text{th}}$  metacarpal via the pisiform and the tendon of flexor carpi radialis passes through a fibro-osseus tunnel, on the radial aspect within the carpal tunnel, to insert into the base of the  $2^{\text{nd}}$  metacarpal.

**Intrinsic:** There are four groups of intrinsic hand muscles: the thenar and hypothenar muscles, the lumbrical muscles and the interossei muscles. The thenar group consists of abductor pollicis brevis, flexor pollicis brevis, oppenens pollicis and adductor pollicis. The thenar group all have their origin on the radial aspect of the TCL and insert into the proximal phalanx of the thumb. The thenar muscles, with the exception of adductor pollicis, are innervated by the recurrent branch of the median nerve as it exits the carpal tunnel. The thenar muscles provide adduction, abduction, flexion and opposition of the thumb.

The hypothenar group consists of: abductor digiti minimi, flexor digiti minimi and opponens digiti minimi. Their origin is on the ulnar aspect of the TCL and insert into the base of the 5<sup>th</sup> proximal phalanx. All the muscles of the hypothenar group are innervated by the ulnar nerve and operate to perform flexion, abduction and opposition of the 5<sup>th</sup> metacarpal and phalange.

The lumbrical muscles are located deep in the palm of the hand. They originate from the tendons of flexor digitorum profundus, distal to the carpal tunnel, and insert into the radial aspect of the digital extensor tendons. The  $2^{nd}$  and  $3^{rd}$  lumbricals are innervated by the median nerve while the  $4^{th}$  and  $5^{th}$  are innervated by the ulnar nerve.

The interossei muscle consist of 3 palmar and 4 dorsal muscles. They originate from the two borders of the metacarpals between which they sit and insert into the proximal phalanges of the  $2^{nd}$  to  $4^{th}$  digitis. They act to adduct and abduct the fingers and are all innervated by the ulnar nerve.

Superficial to the TCL and palm of the hand lays the palmar aponeurosis and its tendon, Palmaris longus. Palmaris longus is absent in approximately 15% of the population, but where present, it inserts into the palmar fascia of the hand before dividing into slips attaching to the palmar skin of each finger ( $2^{nd}$  to  $5^{th}$ ).

#### 1.2.4 Skin and Nerves

Two main nerves pass into the hand; the ulnar and median nerves. The ulnar nerve passes into the hand external to the TCL via the ulnar canal. The median nerve passes into the hand through the carpal tunnel to provide motor function and sensation for the radial aspect of the palmar hand. The palmar skin is thick with strong attachments to the underlying palmer fascia. The palmar creases of the hand indicate the strongest attachments of the skin to the underlying tissue. A layer of subcutaneous adipose can also be found across much of the palmar surface to provide shock absorption. (Agur and Dalley, 2009; Yu *et al*, 2004).



Figure 1: Transverse Carpal Ligament

Key:

- 1. 1<sup>st</sup> metacarpal
- 2. 2<sup>nd</sup> metacarpal
- 3. 3<sup>rd</sup> metacarpal
- 4. 4<sup>th</sup> metacarpal
- 5. 5<sup>th</sup> metacarpal

- H. Hook of Hamate
- C. Capitate
- Tm. Trapezium
- P. Pisiform
- Tq. Triquetrium
- L. Lunate
- S. Scaphoid
- U. Ulna
- R. Radius



Figure 2: Anatomy of the wrist at the level of distal carpal tunnel

#### 2 **Review of the Literature**

#### 2.1 Introduction

The mechanical and viscoelastic properties of human soft tissue has been investigated extensively and there are numerous studies into carpal tunnel syndrome; aetiology, methods of treatment, their efficacy, carpal tunnel mechanics and many more. Berger (1993) proposed a method of elongating the TCL, as opposed to transection, as a treatment method to relieve pressure within the carpal tunnel and to avoid many of the post-operative complications associated with open and endoscopic release surgery. Some research has been conducted into the validity of this idea, and one key factor requiring investigation is the viscoelastic properties of the TCL. This review of the literature, therefore, aims to explore the research previously conducted on wrist biomechanics, soft tissue mechanics and ligament viscoelasticity; to give the reader a full understanding of the current field of knowledge of this subject and to highlight where more work is required.

#### 2.2 Method

Online internet searches were performed using PubMed central, ISI Web of Knowledge and Google Scholar for studies published in English between 1980 and 2011. The following keywords were used in the search, either alone or in any combination: Transverse carpal ligament, viscoelastic, carpal tunnel syndrome, percutaneous carpal tunnel plasty, mechanical properties, stiffness, stress/loadrelaxation, and confined compression. 34 journal articles have been reviewed in this paper.

#### 2.3 Discussion

#### 2.3.1 Anatomy of the Human Wrist

There has often been some ambiguity with regard to the use of the terms *flexor retinaculum* and the *transverse carpal ligament* to describe the structure that forms the roof of the carpal tunnel; uniting the four marginal bones of the carpus and serving as an insertion for the thenar and hypothenar muscle groups. Previously, these terms have both been utilised to describe the same structure even though their fundamental definitions differ. A 'retinaculum' is a structure that retains an organ or other tissue in place, while a 'ligament' is defined as a band of fibrous tissue that connects bone to bone (or cartilage) and supports joints (Medical dictionary, online, 2011)

Grant's Atlas of Anatomy (11<sup>th</sup> edition, Agur and Dalley, 2009) does not make any distinction between the flexor retinaculum and the transverse carpal ligament indicating they are one and the same structure while Middleton *et al* (1987) refers to this structure only as the flexor retinaculum. Cobb *et al* (1993) investigated 26 cadaveric arms via histological and radiographical methods and proposes there to be three distinct but contiguous segments to the flexor retinaculum: a proximal deep investing fascia of the forearm; the transverse carpal ligament spanning the carpal arch; and a distal portion of aponeurosis between the thenar and hypothenar muscle groups.

Stecco *et al* (2010) also performed a cadaveric study to more accurately differentiate the flexor retinaculum and the transverse carpal ligament. In their study, 30 unembalmed hands were examined by dissection and histological, and immunohistochemical staining. They found there to be two distinct structures on the volar aspect of the wrist: the antebrachial fascia, being composed of three layers of undulating collagen fibre bundles with different fibre orientations between layers, many nerve fibres and Pacini and Ruffini corpuscles. Deep to this fibrous layer another fibrous structure was identified; comprised of thicker fibre bundles with few nerve fibres, and spanning the carpal arch between the pisiform and hook of hamate on the ulnar side to the tubercle of scaphoid and ridge of trapezium on the radial side. The researchers concluded that the superficial tissue is in continuity with the antebrachial fascia and acts as its reinforcement while the deeper structure is characteristic of ligamentous tissue and connects the four marginal bones of the carpus together. They propose that the term transverse carpal ligament be used to describe the lamina of tissue connecting the pisiform and hook of hamate to the trapezium and scaphoid, while the term flexor retinaculum should be abandoned as it does not relate to a specific structure.

#### **2.3.2** Role of the Transverse Carpal Ligament

Some of the previous ambiguity in the terminology of the structure covering the carpal tunnel can be attributed to the role the TCL in wrist and hand biomechanics. The TCL has two main roles in wrist function: carpal arch stability and retention of the extrinsic digital flexor tendons within the carpal tunnel for effective wrist biomechanics.

The TCL is important in the stability of the carpus. A study by Xiu et al (2010) investigated the structural mechanics of the carpal arch and the TCL. They applied paired forces to the attachment sites of the TCL (two forces applied to the pisiform and hook of hamate on the ulnar side and two forces applied to the scaphoid and trapezium on the radial side). Carpal tunnel and carpal arch deformation was compared with the TCL intact and transected; with both inwardly and outwardly applied loads. They report that under 10N of inwardly applied load with the TCL intact, carpal arch width decreased by 10.8% at the distal portion and by 37.5% at the proximal portion. With the TCL transacted the same test resulted in a decrease of 10.6% and 37.9% distally and proximally respectively. When an outwardly applied load of 10N was placed with the TCL intact, carpal arch width increased by 3.7% distally and 18.8% proximally. While following TCL transection increases of 9.6% distally and 33.9% proximally were reported. The authors concluded that the TCL plays an important role in the stabilisation of the carpal tunnel under outwardly applied loads and that the proximal portion of the carpal arch is more compliant than the distal portion.

The findings reported by Xiu *et al* (2010) support those of a similar study by Tengrootenhuysen *et al* (2009) who found that gradual sectioning of the TCL resulted in significant increases in carpal arch width while under load. However, the authors of this study stated that the carpal arch retains a good level of stability after transection of the TCL.

Grip and pinch strength can also be affected following transection of the TCL. A study by Gellman *et al* (1988) evaluated the length of time required following carpal tunnel release surgery for grip and pinch strength to return to preoperative levels. They found that grip strength returned to preoperative levels by 3 months post-operation and that by 3 weeks and 6 weeks grip strength was 28% and 73% respectively. Pinch strength was found to return sooner with 74% and 96% recovery by weeks 3 and 6 respectively and 108% by 3 months.

Grip strength was also analysed by Mathura *et al* (2004). In this study, 30 patients with carpal tunnel syndrome were tested for power grip and key pinch strength with the wrist extended and flexed, and before and after carpal tunnel release surgery. There were no reported differences in key pinch strength; however there was a significant decrease in grip strength following carpal tunnel release in both wrist extension and flexion with the greatest decrease evident during wrist flexion. The authors attribute the loss of grip strength during wrist flexion to excursion of the flexor tendons out of the carpal tunnel.

The excursion or 'bow-stringing' of the flexor tendons against the palmar surface of the hand during wrist flexion has been documented in some earlier studies (Kline and Moore, 1992; Kiritsis and Kline, 1995). Kline and Moore (1992) examined four fresh-frozen cadaver hands for flexor tendon excursion during extension and flexion of the wrist before and after TCL transection. Following transection of the TCL, tendon excursion palmarly during wrist flexion was found to increase by 25% and 20% for the tendons of digitorum profundus and digitorum superficialis respectively. This equated to a 'bow-stringing' displacement of  $5.4 \pm 1.2$  mm and  $5.5 \pm 1.3$  mm respectively. The authors propose that the TCL acts to maintain the flexor tendons close to the centre of rotation of the radiocarpal, intercarpal, and carpometacarpal joints; creating an effective pulley system. Transection of the TCL disrupts this flexor pulley system and palmarly directed excursion of the tendons during flexion

limits the available motion at other joints where these tendons act. The authors also state that this may contribute to the loss of grip strength experienced following carpal tunnel release surgery.

## **2.3.3** Mechanical and Viscoelastic Properties of Different Tissue Layers

If ligament elongation is chosen over transection as a treatment method, it would be necessary and prudent to consider not only the viscoelastic properties of the TCL but also those of the tissue layers and structures superficial to the TCL. Anterior to the TCL, the flexor retinaculum and the attaching fibres of the thenar and hypothenar muscles can be found. Above this layer, in the majority of individuals, lays the palmar aponeurosis and its tendon, Palmaris longus. Anteriorly to this lies the subcutaneous adipose layer followed by the dermal and epidermal layers of the skin.

Various studies have been conducted to investigate the mechanical properties of the skin layers (with various results reported). An older study by Agache *et al* (1980) looked at the mechanical properties of the dermis in vivo using a rotating disc (enclosed in a guard ring) to apply a fixed amount of torque to the skin. A torque of 28.6 Nmm was applied to the dorsal skin of the forearm of 138 volunteers, aged 3-89 years. The researchers report Young's Modulii of 0.42 and 0.85 Mpa for young and older age groups respectively (<30 yrs: >30 yrs). The researchers also report that elasticity of the skin remains stable until the age of 30 years where it drops by approximately 50%, while Young's Modulus follows an inverse pattern whereby it increases by approximately 50% around the age of 30 years.

Smalls *et al* (2006) reports some similar findings in their study. They looked at the effect of dermal thickness, body site and tissue composition on the biomechanical properties of healthy female volunteers using a negative pressure (suction) method. The researchers report significant correlations for skin thickness to stiffness, energy absorption and biological elasticity (ratio of elastic recovery to elastic deformation). They also report differences between body sites. Stiffness was found to be significantly higher at the calf compared to the thigh and shoulder; and significantly

higher at all sites on the dominant side compared to the non-dominant side. Age was found to be a factor. Significant decreases in elasticity were found for increases in age; however, no increase in stiffness was found at any site with age; a finding that contradicts those of Agache *et al* (1980) but may be due to the different methodologies employed between the two studies.

A more recent study by Chrichton et al (2011) employed yet another technique to measure the viscoelastic properties of the skin. The researchers here used Atomic Force Microscopy (AFM) indentation on mouse skin layers using two different sized spherical nano-indenters. They compared a 6.62 µm indenter to a 1.9 µm indenter on the elastic modulii and stress relaxation of the stratum corneum (SC), viable epidermis (VE) and the dermis of mouse ear skin. They report significantly greater stiffness values for the dermis (7.33 - 13.48 MPa) compared to the SC (0.75 - 1.62 MPa)MPa) and VE (0.49 – 1.51 MPa. The lower – larger values represent the larger – smaller indenter results respectively). They also found the VE to show the greatest amount of relaxation; almost fully relaxing within 10 seconds. The SC and dermis in comparison, both relaxed initially before reaching a plateau at approximately 40% relaxation. The researchers postulate that the higher stiffness values for the dermis compared to the other layers may be due to the higher collagen and elastin content of the dermis preventing rapid expulsion of fluid. The shorter term viscoelasticity of the SC layer is thought to be due to the hydration gradient found between the superficial surface (15%) and the SC/VE boundary layer (70%). The author proposes that the dryer, rigid surface of the SC allows stress distribution of the indenters and that the higher fluid content and lower cellular matrix density of the VE allows faster fluid expulsion compared to the other layers.

Lying deep to the dermis is the hypodermis, also known as the subcutaneous tissue, is a layer consisting of loose connective tissue housing lobules of adipocytes and other cells such as fibroblasts and macrophages. The hypodermis is found at all sites of the body and the subcutaneous adipose found within the hypodermis primarily acts as a layer of thermal insulation and energy storage. There are sites of the body that require fat pads for purposes other than previously mentioned. The adipose tissue found in the calcaneal and palmar fat pads are responsible for shock absorption and load bearing. This results in a different chemical composition of the adipose at these sites compared to the rest of the body's subcutaneous adipose. The fat pads of the hands and feet have a larger ratio of unsaturated to saturated fatty acids (Geerligs *et al*, 2008).

Geerligs *et al* (2008) investigated the linear viscoelastic properties of porcine subcutaneous adipose using rotational rheometer methods. They reported a shear modulus of 7.5 kPa at 10 rad/s and 37°C. They also looked at the effects of snap freezing and report that the mechanical properties of the tissue did not change even though some cell damage was evident under histological examination. The researchers also reported that the storage and loss modulus at up to 0.1% strain showed strain rate and temperature dependency. A more relevant study by Miller-Young et al (2002) performed unconfined compression tests on human calcaneal fat pad samples to determine its material properties. Cylindrical sections  $\sim 10$  mm thick and 8 mm in diameter were tested with instantaneous compression to 40% strain. The researchers report peak stresses of 21.3 kPa relaxing to 5.4 kPa after 60s. A strain rate dependence on peak stress was also found here with peak stresses of 33.7 and 25.2 kPa being recorded for displacement rates of 350 mm/s and 175 mm/s respectively. The author explains that the structure of the fat pad; with its high ratio of unsaturated fatty acids, may explain the rapid stress relaxation. They state that due to unsaturated fatty acids being highly branched they are able to move quite freely, and combined with a reduced intercompartment fluid flow, results in a less viscous material. These finding may be relevant since the palmar fat pad is similar in its composition to the calcaneal fat pad.

The layer of soft tissue anterior to the TCL is comprised of myoctes (muscle fibres) of the thenar and hypothenar muscles and their tendinous fibres as they converge with the TCL and each other. Skeletal muscles are generally connected to bones on either side of one or more articular joints to provide movement and postural support. They are connected either directly or indirectly to the bone via continuation of the collagenous fibres of the muscles endomysium, perimysium and epimysium with those of the periosteum of the bone. An indirect attachment implies there is a gap between the muscle fibres and the bone that is bridged by the convergence of the

muscular fascias into tendinous fascicles which then continue into the periosteum. The origin of the thenar and hypothenar muscles is considered to be the carpal bones via the TCL (Agur and Dalley, 2009). However, Tanabe and Okutsu (1997) found that the originating fibres of the thenar and hypothenar muscles attach to each other; forming a layer of transverse fibres separate and slightly distal to the TCL. Whether these fibres originate from the TCL or not, they are still an important component in wrist and hand biomechanics, and also in the mechanics of carpal tunnel syndrome. For effective relief of CTS, decompression of the carpal tunnel is required and this is usually accomplished through transection of the TCL. Tanabe and Okutsu (1997) examined 20 cadaveric hands and found that the distance between the cut ends of the TCL increased from 1.5 mm to 6.6 mm when the transverse fibres between the thenar and hypothenar muscles play a role in carpal tunnel mechanics and may add to the structural integrity provided by the TCL.

The point of attachment of the tendinous fibres of these two muscle groups along the TCL may differ among individuals, along with the extent to which the muscle fibres traverse the TCL (Dr Q Fogg, personal communication, 2<sup>nd</sup> June 2011). Since the thenar and hypothenar muscles also contribute to load bearing and shock absorption over the TCL, their mechanical and viscoelastic properties should also be considered.

Van Loocke *et al* (2008) investigated the viscoelastic properties of passive skeletal muscle. They performed uniaxial unconfined compression and hold tests on fresh porcine skeletal muscle at different fibre orientations (along fibres, cross-fibre and 45° fibre angle) and deformation rates. Both the rate of deformation and the fibre orientation during ramped compression were found to be factors in the stiffening effect of the muscle. Greater stiffness was observed cross-fibre at low strain rates (<5% s<sup>-1</sup>), while above this strain rate, stiffness was greatest when the fibre orientation was parallel to the direction of compression. Stress relaxation was greatest for the parallel fibre direction (60% relaxation) compared to cross-fibre compression (45% relaxation) but in all cases, 80% of the stress relaxation occurred within the first 100 s. The rate of relaxation was shown to only be affected by the amount of compression and not the rate of compression. No significant differences

were reported for relaxation rate due to compression rate but a drop from 30% to 10% deformation resulted in a 20% increase in relaxation rate. The difference in viscoelastic behaviour due to the fibre orientation, therefore, results in the skeletal muscle being anisotropic.

The stress relaxation reported by Van Loocke *et al* (2008) is slightly less than that reported in an earlier study by Silver-Thorn (1999) as noted by the authors of the more recent study. Silver-Thorn (1999) reported *in-vivo* force relaxations of 55-95% on the soft tissue bulk of the calf of trans-tibial amputees and non-amputees, with equilibrium being reached within 60 s, quicker than that reported by Van Loocke *et al* (2008). However, there are major methodological differences between the two studies that may account for the difference.

#### 2.3.4 Viscoelastic Properties of the Transverse Carpal Ligament

Ligaments are a viscoelastic material, in that their behaviour is time-dependent. These responses include stress relaxation and creep. When a ligament is held at a constant strain the stress experienced by the ligament decreases with time (stress relaxation), and conversely, when a ligament is held with a constant level of stress the ligament elongates with time (creep). Further to these definitions, the stress relaxation of ligaments is said to be strain dependent. Duenweld et al (2009) found that the rate of relaxation in tendons is dependent on the level of strain and that as strain increases; the rate of relaxation also increases. This contradicts the finding of Van Loocke et al (2008) which reports faster relaxation rates in muscle with lower levels of strain. An explanation to this discrepancy may be found in the structural and compositional differences of muscle and tendinous tissue. Bonifasi-Lista et al (2005) found that stress relaxation progressed faster at lower strain levels in human medial collateral ligaments, and that energy dissipation was greatest at high strain levels. The author attributes this to the uncrimping of the collagen fibres during lowlevel strains; the solid constituents of the MCL are less involved at low strains and that the ground substance matrix (glycosaminoglycans (GAG), proteoglycans (PG) and water) is more viscous in character and bears the majority of the load.

Provenzano *et al* (2001) also found relaxation rate to decrease with increasing strain in the MCLs of rats. The researcher here posited that at higher strains, a greater amount of fluid is expelled from the tissue making the tissue more elastic. Therefore, water content affects relaxation with higher water content in ligaments resulting in greater levels of load relaxation (Chimich *et al*, 1992).

There is an abundance of research conducted into the viscoelastic properties of ligaments, especially those of the knee, however, ligament function varies by location as does its composition and structure, as Frank (2004) suggests. Specific research into the viscoelastic properties of the TCL is much scarcer. Studies have been carried out on the extensibility of the TCL and/or the expansion of the carpal tunnel.

Sucher et al (2005) investigated transverse carpal arch (TCA) width under static and dynamic loading. The static loading comprised of 10 N loads being applied to pins inserted in the four (TCL attaching) carpal bones and the TCA width was measured over 3 hours, after which the load was removed and the TCA allowed to recover. Measurements were also taken during the 2 hour recovery. Dynamic loading consisted of a series of osteopathic manipulations being applied to the hands prior to static loading as above. Their results showed that under static loading, the TCL elongated by approximately 3.7 mm (13%) and recovered to an elongated length of 2.6 mm. The dynamic loading condition resulted in an even greater elongation after the static load, as would be expected. The research also found that female hands were more compliant than those of males; while a study by Li (2005) found the carpal tunnels of females to be less compliant than those of males. In the latter investigation, manual indentation of the TCL region, perpendicular to the palmar surface of 12 male and 12 female volunteers was performed. With load increases from 0.25 kg to 2.00 kg, indentation depths of  $1.82 \pm 0.30$  mm and  $1.38 \pm 0.25$  mm were recorded for males and females respectively. This equated to an effective compliance of  $0.101 \pm 0.018$  mm/N for males and 0.075 mm/N for females (24.5% less). The researcher posited that this may be a factor for the greater prevalence of CTS in females. This may suggest that the carpal tunnels in females are less compliant due to their smaller size compared to males. Bower (2006) in an MRI

investigation into CT dimensions, found males had significantly larger carpal tunnels and carpal tunnel contents than females, but there was no difference in 'CT': 'CT content' ratio between males and females. The research also found that the crosssectional area of the carpal tunnel was smallest at the distal end (level of hook of hamate) especially during wrist flexion.

A smaller volume of the carpal tunnel may result in greater carpal tunnel pressures associated with CTS, with an average of 36 mmHg being reported by Uchiyama *et al* (2010). An increase in carpal tunnel pressure can be the result of numerous causes; mostly inflammatory pathologies of various structures associated with the carpal tunnel complex. Another cause that may be a factor in repetitive stress related CTS is the incursion of the lumbrical muscles into the carpal tunnel during finger flexion. Cobb *et al* (1993) measured the extent to which the lumbrical muscles were pulled into the carpal tunnel on five cadaveric hands. The lumbrical muscles have their origin on the tendons of flexor digitorum profundus in the palm of the hand. The researcher found that the origins of the lumbrical muscles translated from 7.8 mm distal to the TCL, at full finger extension, to 30 mm within the tunnel at 100% finger flexion.

A method of elongating the TCL, as opposed to transection, was developed by Berger (1993) using an inflatable balloon catheter that could be inserted under the TCL and then inflated to palmarly stretch the TCL. The device also incorporated a nerve shield to lessen the pressure exerted on the median nerve during the procedure. To validate the efficacy of this procedure, proper understanding of the mechanics of the carpal arch and TCL are needed.

Zheng *et al* (2006) used a Tissue Ultrasound Palpation Sensor (TUPS) to measure the thickness and elasticity of the tissue in the carpal tunnel area of five male volunteers. They report average tissue thicknesses overlying the TCL of  $7.98 \pm 1.05$  mm and a tunnel depth of  $9.59 \pm 1.12$  mm (with 5 N loads and a 9 mm diameter indenter). The stiffness of the soft tissue layer above the TCL was reported as  $6.72 \pm 2.10$  N/mm and  $15.63 \pm 8.42$  N/mm for the tunnel contents. The author also posited that the stiffness may be affected by the boundary conditions: a small tunnel would provide

less available space to accommodate deformation of the tunnel contents; resulting in greater apparent stiffness.

A study by Li et al (2009) looked at the ability of the TCL to stretch from a palmarly applied load from within the carpal tunnel. The hands of ten fresh frozen cadaveric specimens were tested in this investigation. The dissected hands were secured and a lever inserted into the excavated carpal tunnel. Loads of 10 N to 200 N were applied to stretch the TCL palmarly. The researchers found that length of the TCL did not change during loading but formed an arch, pulling in the side walls of the carpal tunnel. The researchers report that under a 10 N load the carpal tunnel area (CTA) increased by  $33.3 \pm 5.6 \text{ mm}^2$  from a resting area of  $148.4 \pm 36.8 \text{ mm}^2$  (22.4% increase), and by  $48.7 \pm 11.4 \text{ mm}^2$  under 200 N of load (32.8% increase). The author concludes that since there was no elongation of the TCL, the increase in CTA was due to changes in shape of the tunnel; carpal arch width decreased and tunnel depth increased. A more recent Study by Li et al (2011) incorporated a similar method to increase CTA but utilising a balloon device inflated in the carpal tunnel with precise amount of pressure. In this investigation they found that the cross-section area at the hook of hamate level increased by 9.2% with 100 mmHg of pressure and by 14.8% with 200 mmHg. Again, the author notes that the circularity of the carpal tunnel increases during the procedure rather than any actual elongation of the TCL.

Elongation of the TCL may still be possible though. A study by Holmes *et al* (2011) investigated the effects of wrist posture and indenter size on the mechanical properties of ten fresh frozen, exposed TCL (*ex vivo*) and the carpal tunnel (carpal tunnel contents intact) complexes. Here, perpendicular indentation of the TCL was performed using four different sized indenters (5, 10, 20 and 35 mm diameters) and in three different wrist postures (neutral, 30° flexion and 30° extension) to measure the resulting displacements from a 50 N load applied at a rate of 5 N/s. The researchers report that the flexed wrist posture resulted in significantly greater stiffness ( $40.0 \pm 3.3$  N/mm) than neutral ( $35.9 \pm 3.5$  N/mm) and extended ( $34.9 \pm 2.8$  N/mm) positions. Stiffness was found to be significantly greater for the 10 mm and 20 mm indenters compared to the 5 mm indenter, while the 35 mm indenter produced significantly less stiffness than the 20 mm indenter only. Also of interest, and in

contrast to the findings reported by Li *et al* (2009 and 2011), was that there was no significant decrease in carpal arch width reported between peak loading ( $28.7 \pm 0.4$  mm) and at rest ( $29.3 \pm 0.5$  mm). However, the different methodological approaches to testing the carpal tunnel complex between these two studies may account for the difference. Holmes *et al* (2011) postulates that the ratio of indenter size to ligament width may affect the stiffness. Since the radial and ulnar portions of the TCL have been found to be thicker at the radial and ulnar edges (Pacek *et al*, 2010), the wider indenters (apart from the 35 mm indenter) will encroach on these thicker portions of the TCL to a greater degree than a narrow indenter.

While the study by Holmes et al (2011) does not address the viscoelastic properties of the TCL, the results are of interest. A similar short study by Chaise et al (2003) does investigate the viscoelastic properties of the TCL, but only from one fresh frozen hand (81 yr old male). Indentation was performed using a 10 mm indenter at three levels of dissection: on the palmar surface, on the exposed TCL with tunnel contents intact, and on the exposed TCL with the tunnel contents removed. Cyclic preconditioning (0-6 mm at 2 Hz for 5 min) was applied before each of 3 trials of ramp and hold displacement (8 mm, held for 30 min) and for 3 trials of cyclic load relaxation (0-8 mm at 2 Hz for 10 min) at each dissection level. A 1 hour recovery period between trials was included. The peak load recorded for the palmar surface, TCL-exposed and TCL-only conditions were 27.1, 80.5 and 21.7 N respectively. The stiffness of the TCL –exposed condition (46.0 N/mm) was reported to be approximately three times greater than the other two conditions (palmar surface: 15.6 N/mm, and TCL-only: 10.1 N/mm) for the ramp and relaxation test. The present study employed a very similar methodology to that used by Holmes et al (2011). Indentation was performed with a 10 mm indenter, in a neutral wrist posture only, indentation was controlled by displacement (6 mm for preconditioning and 8 mm for ramp and hold test) and the specimen was subjected to an extensive number of trials.

#### 2.4 Conclusion

Following a review of the literature, it can be seen there are many different wrist structures that need to be considered when treating CTS. The various testing protocols and methodologies used to investigate soft tissue mechanics across these studies impose limitations on the ability to directly compare the results.

When testing the mechanics of the TCL, few researchers employ the same methodology of testing. Tensile methods have been used by Li *et al* (2009, 2011) while indentation methods have been utilised by Holmes *et al* (2011) and Chaise *et al* (2003). Regardless of the test methodologies used, there is still a lack in research into the viscoelastic properties of the TCL with little consideration given to the mechanical and viscoelastic properties of the tissues superficial to the TCL.

#### **3** Methodology and Procedures

#### 3.1 Subjects

Six embalmed cadaveric hands, amputated at the mid forearm level, were tested in this investigation ( $82 \pm 6.29$  years of age). The six hands were from four individuals (two pairs and two individual hands), three male (four hands) and one female (both hands). All cadaveric specimen donors have no history of injury or pathology of the wrist. The specimens were supplied by the Laboratory of Human Anatomy, University of Glasgow.

#### 3.2 Design

The viscoelastic and mechanical properties of the intact TCL were tested in compression (indentation) using a materials testing machine.

Four indentation trials for each specimen at different levels of dissection were performed to compare the mechanical and viscoelastic properties between conditions and to compare the individual effect of the thenar and hypothenar muscle groups on the measured variables.

There are two independent variables: Group; with three levels (Thenar group, Hypothenar group and Combined); and Condition; with four levels (Intact, Skin removed, Hypothenar or Thenar removed, and TCL exposed). There are three dependant variables of interest: Peak Load (N), 80% Load Relaxation Time (s), and Stiffness (N/mm).

#### 3.3 Materials

Cadaveric specimens were transported in secure, leak proof, and appropriately labelled containers from the Laboratory of Human Anatomy, University of Glasgow to the Level II containment suit of the Bioengineering department, University of Strathclyde, Glasgow.

Anthropometric measurements, such as: wrist width, wrist depth, palm width and palm depth (at the carpus and at the distal metacarpus), and TCL width, length and thickness were recorded for all specimens using digital precision callipers (Mitutoyo Digimatic).

Dissection was performed using standard dissection tools: forceps, scalpel, and scissors. The researchers donned appropriate protective laboratory coats and blue nitrite gloves when handling and testing the cadaveric specimens.

Indentation was performed using a custom made 10 mm diameter aluminium indenter (fig. 7, Appendix A). Indentation was performed using a BOSE ElectroForce 3200 materials testing machine with a BOSE 450.00 N dry load cell (model: 10-32 THD) fitted in series between the indenter and the upper actuator of the machine. The testing machine was operated by personal computer (PC) running Wintest 4.1 Digital Control System software package.

The cadaveric hands were secured to a custom built aluminium plate that was fixed to the base of the testing machine (fig. 8, Appendix A). The specimens were secured in place via three rectangular aluminium bars that could be tightened down via two 8 mm diameter threaded bolts and wing nuts per bar.

#### 3.4 Procedure

Six specimens were each tested in four indentation trials:

- 1. Intact hand
- Skin removed: epidermis, dermis, subcutaneous adipose and palmar aponeurosis removed
- 3. Removal of one muscle group (thenar or hypothenar)
- 4. TCL exposed (removal of remaining muscle group to fully expose the TCL, with carpal tunnel contents intact)

One specimen was tested in a fifth trial following removal of the carpal tunnel contents (TCL Only) to give a single comparison of the effect of the tunnel contents on stiffness and load relaxation.

#### **3.4.1 Specimen Preparation and Dissection**

Prior to testing, anthropometric details were recorded for each specimen and the centre of the TCL was located and marked for accurate indenter placement. Measurements were taken for wrist width, wrist thickness, palm width (carpal level), distal palm width, and palm thickness at the mid TCL level.

The centre of the TCL on the intact hand was determined through palpation and marking of the pisiform and hook of hamate ulnarly and the tubercle of scaphoid and ridge of the trapezium radialy. Two transverse lines were drawn: one distal connecting the hook of hamate to the first carpometacarpal joint, and one proximal connecting the pisiform to the tubercle of scaphoid. A longitudinal line was then drawn following the radial side of the fourth phalange and intersecting both transverse lines. The centre of the TCL was identified as a point on the longitudinal line 10 mm proximally from the distal transverse line (Appendix B).

Following each trial the specimens were dissected in preparation for the next trial. After trial one, all specimen were carefully dissected to remove the skin, subcutaneous adipose, the palmar aponeurosis and palmaris longus tendon, and palmaris brevis muscle. The centre of the TCL was then relocated and marked using the above procedure.

Upon completion of trial two (skin removed) three specimens were dissected to remove the thenar muscles (leaving the hypothenar muscles intact) and three were dissected to remove the hypothenar muscles (leaving the thenar muscles intact).

Trial three was then performed with three specimens in each group (thenar removed, and hypothenar removed).

Following the third trial the remaining thenar or hypothenar muscle group was removed along with any remaining flexor retinaculum to fully expose the transverse carpal ligament. The contents of the carpal tunnel were left intact along with the tendons for flexor carpi ulnaris and flexor carpi radialis.

Sample specimen dissection images can be found in Appendix C.

#### 3.4.2 Specimen Mounting

Following preparation, the specimens were secured to a flat aluminium platform attached to the lower crosshead of the testing machine. The specimens were secured to the platform, supinely and in a neutral wrist posture, by three aluminium bars: one distal across the heads of the  $2^{nd} - 5^{th}$  metacarpals and the  $1^{st}$  distal interphalangeal joint (thumb); and two proximal, across the distal forearm. The specimens were fully secured once centrally located below the indenter. The lower crosshead platform was then raised to position the specimen in as close proximity as possible, without contact, to the indenter.

#### **3.4.3 Test Protocol**

The test protocol was developed using the Wintest 4.1 software package and one pilot cadaveric hand specimen.

The actuator of the BOSE testing machine has an operational displacement range of  $\pm 6.50$  mm (13.00 mm total range). Safety displacement limits were set to  $\pm 5.50$  mm and load safety limits were set to  $\pm 400.00$  N in order to protect the load cell from damage. A peak displacement of 8.00 mm was originally chosen, in line with the protocol described in a similar study by Chaise *et al* (2003), for the test protocol; however, this displacement could not be achieved without failing the 400.00 N safety

limit of the load cell. A peak displacement of 4.50 mm was found to be the largest safe displacement that left a suitable margin for specimen peak load variability.

#### Stage 1: Preconditioning cycle

The indenter was lowered to 0.50 N preload contact with the specimen. Current displacement was recorded and a peak displacement -4.50 mm from this position was calculated and entered into the software package. Ten sine wave preconditioning cycles of 4.50 mm displacement at a rate of 2 Hz were performed. A half sine block was added to ensure the indenter finished the tenth cycle at 0.00 mm relative displacement.

Stage 2: Recovery

A short recovery period of 10 s was then afforded the specimen following the precondition cycle.

Stage 3: Displacement Ramp and Hold

Following the 10 s recovery period, a ramped displacement of -4.50 mm at 2.00 mm/s was performed and held at constant displacement (constant strain) for 1800 s (30 minutes).

A 1 hour recovery period followed completion of the test; the specimen was removed from the platform and dissected in preparation the next trial. During the recovery period hydration of the specimens was maintained using a 0.9% phosphate buffered saline solution (1 tablet per 200 ml water) sprayed onto the specimens following dissection.

#### **3.5 Data Analysis**

Time (s), load (N) and displacement (mm) data was sampled at a frequency of 200 Hz and 1 Hz for the ramped displacement and load relaxation (hold) portions of the test respectively. A higher sampling frequency for the ramped displacement was

chosen to more accurately calculate stiffness of the tissue. Stiffness was calculated as the slope of the linear portion of the stress/strain graph and a minimum linear regression ( $R^2$  –value) was set at  $R^2 = 0.95$  for all trials (Holmes *et al*, 2011).

#### 3.5.1 Calculation of Ligament Strain

Ligament elongation and strain was calculated using a method developed by Holmes *et al* (2011). Figure 1, below illustrates a simple 2D model of a cross section of the mid TCL-carpal arch; used to calculate the final ligament length.



Figure 3: Simple 2D model of final ligament width: final ligament length is calculated from Pythagoras Theorum. Where:  $W_{CI}$  = half ligament width – half indenter width;  $I_D$  = Indentation depth; and  $I_W$  = Indenter width.

Proximal, mid, and distal ligament width were recorded for all specimens. The mean of these three widths was used for this calculation. Ligament width during indentation can be calculated as

$$L_w = (2 \times T) + I_w \tag{1}$$

Where  $L_w$  = instantaneous ligament width, T = length of TCL on either side of the Indenter, and  $I_w$  = indenter width.

#### **3.6** Statistical Analysis

Paired t-tests were used to compare the difference in means between conditions (dissection level); within subjects, and unpaired t-tests were used to compare the difference in mean scores, between subjects, for comparison between the

'Hypothenar Removed' and 'Thenar Removed' conditions.

For all comparisons, alpha ( $\alpha$ ) was set at 0.05.

Validity for performing these parametric statistical tests are inferred from tests performed for assumption of parametric use.

Measures of central tendency are reported via means and standard deviations (M  $\pm$  SD) and the statistical results of the t-tests are reported with regards to the means, standard error, t-value (with degrees of freedom), significance value (*p*), and the power of the significance (r-value) of the two conditions being compared.
#### 4 **Results**

Tests were completed for the assumption of the use of a parametric inferential test. The three measured dependent variables (Peak Load (N), Stiffness (N/mm), and 80% Relaxation Time (s)) are all ratio level data.

Specimens were chosen at random from a selection of cadaveric hands available at the Laboratory of Human Anatomy, University of Glasgow, Scotland. All data sets (except for Relaxation Time: Thenar Group: Intact\* and TCL Exposed\*\*) are normally distributed (Table 3). Tests of normality were performed using Anderson-Darling test for normality since degrees of freedom is less than 50 (df = 2 to 5). All graphical representations for normal distribution of the data can be found in Appendix D.

Table 1: Anderson-Darling test for Normality (* unable to perform normality due to degrees of
freedom = 1, ** contains an outlier)

		Intact Skin		Thenar	Hypothenar	TCL
		Intact	Removed	Removed	Removed	Exposed
	Hypothenar	P =	P = 0.239		P = 0.096	P = 0.535
	Group	0.294	I = 0.257		1 = 0.090	1 = 0.555
Load	Thenar	P =	P = 0.510	P = 0.220		P = 0.080
Load	Group	0.146	I = 0.319	I = 0.229		I = 0.000
	Combined	P =	P = 0.142			P = 0.690
	Comonica	0.167	I = 0.142			I = 0.000
	Hypothenar	P =			D 0.506	D 0.040
	Group	0.454  P = 0.565			P = 0.526	P = 0.248
Stiffness	Thenar	P =	D = 0.142	D = 0.191		D = 0.570
	Group	0.079	P = 0.142	P = 0.181		P = 0.370
	Combined	P =	P = 0.250			P = 0.200
	Comonieu	0.246	I = 0.239			I = 0.209
	Hypothenar	P =	P = 0.304		P = 0.202	P = 0.503
Relaxation Time	Group	0.527	I = 0.304		I = 0.292	I = 0.303
	Thenar	*	P = 0.630	P = 0.087		P = 0.118
	Group		I = 0.030	I = 0.007		I = 0.118
	Combined	P =	P = 0.270			P = < 0.005
	Comonieu	0.072	1 - 0.279			**

Homogeneity of variance was tested for using Bartlett's Test. Equal Variance can be assumed for all data (graphs for homogeneity of variance with Bonferroni 95% Confidence Intervals can be found in Appendix E).

#### Table 2: Bartlett's Test for equal variance

	Test Statistic	Sig.
Peak Load	18.04	P = 0.054
Stiffness	8.12	P = 0.617
Relaxation Time	12.35	P = 0.262

## 4.1 Load Relaxation

Peak load corresponds to the load recorded at the end of the displacement ramp, or beginning (0 s) of the load relaxation. The mean time (seconds) for the load to drop to 80% of peak load for each group's condition is also reported (\*\* Includes possible outlier, excluding =  $210 \pm 99$  s).

Table 3: Mean  $\pm$  SD Peak Loads and Load relaxation results for the three test groups across all conditions

								80%
		Peak					Min	Load
		Load	1 s	10 s	100 s	1000 s	Load	Relaxat
		(N)					( <b>1800 s</b> )	-ion
								Time
	Integ	76.11 ±	$60.08 \pm$	$36.59 \pm$	$19.03 \pm$	$10.20 \pm$	$8.98 \pm$	293 ±
dn	Intact	40.21	31.90	18.83	9.99	5.86	5.36	171
Gro	Skin	115.48	93.93 ±	52.58 ±	$24.64 \pm$	13.44 ±	12.11 ±	146 ±
ar (	Removed	$\pm 75.39$	67.60	36.43	14.43	6.88	6.06	54
hen	Hypothenar	115.71	$94.98 \pm$	54.13 ±	$24.70 \pm$	13.09 ±	11.78 ±	123 ±
poti	Removed	$\pm 64.9$	60.83	35.64	16.28	8.66	7.79	60
Hyl	TCL	172.51	148.21	85.52 ±	39.01 ±	19.05 ±	16.71 ±	147 ±
	Exposed	$\pm 75.39$	$\pm 71.35$	37.58	16.70	8.63	7.55	19
	Intert	59.93 ±	43.53 ±	29.30 ±	17.11 ±	9.33 ±	8.01 ±	422 + 5
	Intact	52.74	34.62	21.94	11.67	5.97	5.14	$422 \pm 3$
dr	Skin	128.10	109.72	$65.60 \pm$	$31.62 \pm$	15.54 ±	13.50 ±	223 ±
iroi	Removed	$\pm 67.40$	$\pm 64.51$	39.48	18.49	8.83	7.71	115
ır (	Thonar	228.15	209.73	122/13	50.00 +	20 11 +	25.64 +	206 +
nen	Removed	±	±	+ 69 66	37.07 -	19.00	16 56	125
L		101.18	113.42	± 07.00	57.20	17.00	10.50	125
	TCL	186.77	173.88	115.16	$63.13 \pm$	32.12 ±	$27.75 \pm$	715 ±
	Exposed	$\pm 47.32$	$\pm 50.30$	$\pm 35.75$	23.87	14.30	12.72	670
	<b>.</b>	$68.02 \pm$	51.81 ±	32.95 ±	$18.07 \pm$	9.77 ±	$8.50 \pm$	344 ±
dno	Intact	42.87	31.12	18.72	9.77	5.31	4.73	140
l Gr	Skin	121.79	101.83	59.09 +	28.13 +	14.49 +	12.81 +	154 ±
nec	Removed	± 64.33	± 59.73	34.71	15.32	7.17	6.25	91
iqm								
Cot	TCL	179.64	161.05	100.34	51.07 ±	25.59 ±	22.23 ±	440 ±
	Exposed	56.84	± 56.98	± 36.60	22.67	12.76	11.14	587**
	TCL Only	19.80	17.44	13.82	9.9	7.02	6.52	67.07%

Normalised load relaxations with time for the three groups are illustrated in figures 2 to 4, below. The rapid load relaxation within the first 100 seconds is evident across all groups followed by a more gradual relaxation to 1000 s.



Figure 4: Normalised mean load relaxation graph for the Hypothenar Group; illustrating the similar viscoelastic properties at the different levels of dissection.



Figure 5: Normalised mean load relaxation graph for the Thenar Group.



Figure 6: Normalised mean load relaxation for the Combined Group.

Paired t-tests were performed to compare the difference in means between the different levels of dissection within each group. All six specimens were tested at the 'Intact', 'Skin Removed' and 'TCL Exposed' levels. Following indentation at the 'Skin removed' level, three specimens underwent dissection to remove the Hypothenar muscle group first (leaving the Thenar muscle group intact) while the other three specimens had the Thenar muscle group removed first (leaving the Hypothenar muscle group intact).

#### **4.1.1 Combined Group:**

The time taken to reach 80% load relaxation was significantly longer for the 'Intact' hands (M = 344, SE = 63) compared to the 'Skin Removed' condition (M = 154, SE = 26), t(9) = 3.19, p = 0.033, r = .73. There were no other significant differences found for relaxation time for any group though some differences were observed. The 'TCL Exposed' condition took slightly longer to relax (M=440, SE=263) than the 'Intact' condition (M=344, SE=63) and the 'Skin Removed' condition (M=154, SE=26).

#### 4.1.2 Hypothenar Group:

'Intact' (M=293, SE=99) relaxation time was longer than 'Hypothenar Removed' (M=123, SE=34.8), t(4)=1.38, p=0.303, r=.57. No difference in relaxation time was noted between 'Skin Removed' and 'Hypothenar Removed', and between 'Hypothenar Removed' and 'TCL Exposed'.

#### 4.1.3 Thenar Group:

Two large but non-significant differences were noted in the Thenar Group. 'Intact' (M=422, SE=3.5) relaxation time was longer than 'Thenar Removed' (M=206,

SE=72) and 'TCL Exposed' (M=715, SE=387) relaxation time was greater than 'Thenar Removed' (M=206, SE=72).

## 4.1.4 Between Group:

The 'Hypothenar Removed' condition (M=123 s, SE=35) showed faster load relaxation than the 'Thenar Removed' condition (M=206 s, SE=72), t(4)=-1.04, p=0.409, r=0.46, though not significantly slower.

Table 4: t-test results for Relaxation Time (s) (M = Mean, SE = Standard Error) (\* = Significant difference).

			М	SE	М	SE				
	Condition 1	Condition 2	(Condition	(Condition	(Condition	(Condition	t(df)	t-	<i>P</i> -	r
			1)	1)	2)	2)	•(•)	Value	Value	
q	Intact	Skin Removed	344	63	154	26	9	3.19	0.033*	0.73
ombine Group	Intact	TCL Exposed	344	63	440	263	9	-0.39	0.719	0.13
C	Skin Removed	TCL Exposed	154	26	440	263	10	-1.20	0.283	0.35
Hypothenar Group	Intact	Hypothenar Removed	293	99	123	35	4	1.38	0.303	0.57
	Skin Removed	Hypothenar Removed	146	31	123	35	4	0.70	0.556	0.33
	Hypothenar Removed	TCL Exposed	123	35	147	11	4	-0.94	0.446	0.43
dno	Intact	Thenar Removed	422	4	206	72	3	1.70	0.339	0.70
nar Gr	Skin Removed	Thenar Removed	223	66	206	72	4	0.18	0.874	0.09
The	Thenar Removed	TCL Exposed	206	72	715	387	4	-1.61	0.248	0.63
	Between Group Comparison									
	Hypothenar Removed	Thenar Removed	123	35	206	72	4	-1.04	0.409	0.46

#### 4.2 Peak Load

#### 4.2.1 Combined Group:

The peak load following 4.5 mm indentation was recorded and compared between the dissection levels (conditions) for the three groups. The peak load on the 'Intact hands' (M=68.0 N, SE=17.5) was significantly lower than that experienced for the 'TCL Exposed' condition (M=179.6 N, SE=23.2), t(10)=-3.78, p=0.013, r=.77, and 'Intact' was also lower, but not significantly, than the 'Skin Removed' condition (M=121.8 N, SE=26.3), t(10)=-2.49, p=0.055, r=.62.

#### 4.2.2 Hypothenar Group:

Within the Hypothenar group the 'TCL Exposed' condition recorded the highest mean load (M=172.5  $\pm$  75.39 N, SE=43.5) though this load was not significantly greater than the 'Hypothenar Removed' condition (M=115.7 N, SE=37.5), t(4)=-3.05, *p*=0.093, r=.84.

No mean difference in load was noted between 'Skin Removed' (M=115.5 N, SE=43.5) condition and the 'Hypothenar Removed' condition (M=115.7 N, SE=37.5), t(4)=-0.03, p=0.980, r=.01.

#### 4.2.3 Thenar Group:

The 'Thenar Removed' condition (M=228.2 N, SE=58.4) recorded a significantly greater peak load than both the 'Intact' condition (M=59.9 N, SE=30.4), t(4)=-5.99, p=0.027, r=.95, and the 'Skin Removed' condition (M=128.1 N, SE=38.9), t(4)=-4.67, p=0.043, r=0.92. The 'Thenar Removed condition also recorded a higher mean peak load than the 'TCL Exposed' condition (M=186.8 N, SE=27.3), t(4)=0.73, p=0.540, r=0.34 though this difference is not significant.

## 4.2.4 Between group:

The 'Hypothenar Removed' condition (M=115.7 N, SE=37.5) reported a lower peak load than the 'Thenar Removed' condition (M=228.2 N, SE=58.4), t(4)=-1.62, p=0.204, r=0.63, though not significantly lower.

_										
	Condition 1	Condition 2	M (Condition 1)	SE (Condition 1)	M (Condition 2)	SE (Condition 2)	t(df)	<i>t</i> - Value	<i>P</i> - Value	r
ed	Intact	Skin Removed	68.0	17.5	121.8	26.3	10	-2.49	0.055	0.62
ombine Group	Intact	TCL Exposed	68.0	17.5	179.6	23.2	10	-3.78	0.013*	0.77
C	Skin Removed	TCL Exposed	121.8	26.3	179.6	23.5	10	-2.54	0.052	0.63
lar	Intact	Hypothenar Removed	76.1	23.2	115.7	37.5	4	-1.09	0.390	0.48
pothen Group	Skin Removed	Hypothenar Removed	115.5	43.5	115.7	37.5	4	-0.03	0.980	0.01
Hy	Hypothenar Removed	TCL Exposed	115.7	37.5	172.5	43.5	4	-3.05	0.093	0.84
dno	Intact	Thenar Removed	59.9	30.4	228.2	58.4	4	-5.99	0.027*	0.95
nar Gr	Skin Removed	Thenar Removed	128.1	38.9	228.2	58.4	4	-4.67	0.043*	0.92
The	Thenar Removed	TCL Exposed	228.2	58.4	186.8	27.3	4	0.73	0.540	0.34
	Between Group Comparison									
	Hypothenar Removed	Thenar Removed	115.7	37.5	228.2	58.4	4	-1.62	0.204	0.63

Table 5: Paired t-test results for Peak Load (\* Significant difference)



Figure 7: Mean Peak loads and standard error for all dissection levels and groups. Illustrates the general increase in peak loads with each level of dissection.

Figure 5 above illustrates the increase in load peak load recorded with individual standard errors. Peak load can be seen to increase after each dissection with the exception of the 'Hypothenar Removed' level of dissection, which does not show any increase in peak load recorded over the 'Skin Removed' condition.

#### 4.3 Stiffness

#### 4.3.1 Combined Group:

Within the combined group, the 'TCL Exposed' condition (M=27.05 N/mm, SE=4.94) recorded the greatest stiffness; being significantly higher than the 'Intact' condition (M=8.50 N/mm, SE=2.52), t(10)=-3.12, p=0.026, r=.70, and greater but not significantly, than the 'Skin Removed' condition (M=16.90 N/mm, SE=3.50), t(10)=-2.25, p=0.074, r=.58.

#### 4.3.2 Hypothenar Group:

No significant difference were observed within the Hypothenar group, though 'TCL Exposed' showed slightly greater stiffness (M=20.96 N/mm, SE=2.94) than the 'Hypothenar Removed' condition (M=14.19 N/mm, SE=3.01), t(4)=-4.12, p=0.054, r=.90. No difference in stiffness was noted between 'Skin Removed' (M=13.24 N/mm, SE=3.68) and the 'Hypothenar Removed' condition t(4)=-0.85, p=0.485, r=0.39.

#### 4.3.3 Thenar Group:

A large but not significant difference was found between the stiffness of the 'Intact' condition (M=7.17 N/mm, SE=2.9) and that of the 'Thenar Removed' condition (M=30.73 N/mm, SE=9.92), t(4)=-3.35, p=0.079, r=.86. The stiffness for the 'Skin Removed' condition (M=20.57 N/mm, SE=5.85) was approximately 33% lower than that for the 'Thenar Removed' condition (M=30.73 N/mm, SE=9.92), t=(4)=-2.26, p=0.152, r=.75, and 'Thenar Removed' was only slightly less stiff than 'TCL Exposed' (M=33.14 N/mm, SE=8.75), t(4)=-0.18, p=0.874, r=.09.

### 4.3.4 Between Groups:

Stiffness was greater for the 'Thenar Removed' condition (M=30.7 N/mm, SE=9.9) compared to the 'Hypothenar Removed' condition (M=14.19 N/mm, SE=3.0), t(4)=1.60, p=0.251, r=0.62, though the difference is not significant.

	Condition 1	Condition 2	M (Condition 1)	SE (Condition 1)	M (Condition 2)	SE (Condition 2)	t(df)	<i>t</i> - Value	<i>P-</i> Value	r
q	Intact	Skin Removed	8.50	2.52	16.90	3.50	10	-2.54	0.052	0.63
ombine Group	Intact	TCL Exposed	8.50	2.52	27.05	4.94	10	-3.12	0.026*	0.70
C	Skin Removed	TCL Exposed	16.90	3.50	27.05	4.94	10	-2.25	0.074	0.58
Hypothenar Group	Intact	Hypothenar Removed	9.82	4.66	14.19	3.01	4	-0.85	0.483	0.39
	Skin Removed	Hypothenar Removed	13.24	3.68	14.19	3.01	4	-0.85	0.484	0.39
	Hypothenar Removed	TCL Exposed	14.19	3.01	20.96	2.94	4	-4.12	0.054	0.90
dno	Intact	Thenar Removed	7.17	2.90	30.73	9.92	4	-3.35	0.079	0.86
nar Gr	Skin Removed	Thenar Removed	20.57	5.85	30.73	9.92	4	-2.26	0.152	0.75
The	Thenar Removed	TCL Exposed	30.73	9.92	33.14	8.75	4	-0.18	0.874	0.09
	Between Group Comparison									
	Hypothenar Removed	Thenar Removed	14.19	3.0	30.7	9.9	2	-1.60	0.251	0.62

Table 6: Paired t-test results and significance\* values for stiffness



Figure 8: Mean stiffness with standard error across all dissection levels an groups. This graph illustrates the general increase in recorded stiffness at each subsequent level of dissection.

As with peak load, the stiffness can be seen to increase with each level of additional dissection, again with the exception of the removal of the hypothenar muscles which showed no increase in stiffness compared to the 'Skin Removed' level of dissection.

Anthropometric measurements for all specimens can be found in Appendix F.

#### 5 Discussion

The purpose of this investigation was to examine the viscoelastic and mechanical properties of the transverse carpal ligament and to quantitatively describe the contribution of the soft tissue layers superficial to the TCL.

Peak load differences were large among the three conditions of the combined group with subsequent levels of dissection resulting in an increase in peak loads ( $68.02 \pm 42.87$ ,  $121.79 \pm 64.33$  and  $179.64 \pm 56.84$  N for Intact, Skin removed, and TCL exposed conditions respectively) however the only significant difference found was between 'Intact' and 'TCL Exposed' conditions (t(10)=-3.78, *p*=0.013, r=.77. The general increase in peak loads observed gives an indication of the load absorption capability of each layer of tissue. The 'Intact' hand reported a mean of 111.62 N less than 'TCL exposed' while the 'Skin removed' condition resulted in 57.85 N less than 'TCL exposed' and 53.77 N more than 'Intact'. From this we can say that the skin and adipose layer provided an average of 53.77 N of load absorption and the combined hypothenar/thenar layer provided an average of 57.85 N of load absorption.

Within the thenar group, the highest peak load recorded was by the 'Thenar removed' condition  $(228.15 \pm 101.18 \text{ N})$  which was significantly greater than both the 'Intact' and 'Skin removed' conditions, and higher (but not significantly) than the 'TCL exposed' condition.

The hypothenar group showed a different trend with the 'Hypothenar removed' condition reporting the same mean peak load as the 'Skin removed' condition  $(115.71 \pm 64.9 \text{ N} \text{ and } 115.48 \pm 75.39 \text{ N} \text{ respectively}).$ 

A large amount of load relaxation occurred across all conditions and groups, reaching 45-61% relaxation within the first 10 s. The time taken to reach 80% relaxation occurred for all conditions (except 'TCL exposed' of the thenar group) within 123 s to 440 s, with an average of 253 s.

The only significant difference to occur between conditions was among the combined group; 'Intact' (M = 344 s, SE = 63) took significantly longer than 'Skin removed' (M = 154 s, SE = 26) to reach 80% load relaxation. Another notable pattern across all groups was that the 'Intact' condition always took longer to reach

80% relaxation than any other condition. It should be noted that within the 'TCL exposed' condition of the thenar group, one specimen recorded an 80% relaxation time of 1486 s; exceptionally longer than any other specimen across all conditions. With the removal of this outlier the mean  $\pm$  SD 80% relaxation time is  $329 \pm 79$  s and  $210 \pm 99$  s for the 'TCL exposed' conditions of the thenar and combined groups respectively.

Across all groups, stiffness generally increased as tissue layers were dissected away. The only significant change however, was within the combined group for 'Intact' vs. 'TCL exposed' ( $8.50 \pm 6.18$  N/mm and  $27.05 \pm 12.11$  N/mm respectively).

Non-significant differences were found between 'hypothenar removed' and 'thenar' removed' for all of the dependant variables. Removal of the thenar muscles resulted in longer 80% relaxation times (67.5% longer), and greater peak loads (97.2% greater) and stiffness (116.6% greater) compared to the removal of the hypothenar muscles.

No statistical analysis was done for the 'TCL only' condition as only one specimen was tested in this condition.

#### 5.1 Peak Load and Load Relaxation

The peak loads recorded in this investigation are consistently greater than those reported by Chaise *et al* (2003). Test methodologies between the two studies are very similar and so some comparisons can be made. The mean peak loads recorded for the 'Intact' and TCL exposed' conditions in this study were  $68.02 \pm 42.87$  N and  $179.64 \pm 56.84$  N respectively, compared to 27.1 N and 80.5 N respectively reported by Chaise *et al* (2003). Though our recorded peak loads may be higher, the percentage increase in loads between the two conditions for both studies is very similar (62% and 66.3 % increases in peak load for the TCL exposed conditions of our study and that for Chaise *et al*, 2003, respectively). A possible explanation for the greater peak loads in our study may be our use of embalmed specimens as opposed to the fresh frozen specimen used by Chaise *et al* (2003). Formalin fixation has been reported in the literature to alter the mechanical properties of biological tissue through the formation of additional intramolecular and intermolecular cross-

links in proteins (Wilke *et al*, 1996), thus adding rigidity to the tissue. Wilke *et al* (1996) found formalin fixed bovine functional spine units to display an 80% reduction in range of motion (ROM) and greater stiffness compared to pre-fixation ROM (fresh). Viidik and Lewin (1966) found formaldehyde fixed rabbit knee ligaments demonstrated greater stiffness, lower failure loads and less elongation compared to a control group. Both authors conclude that fixed tissue mechanics are not entirely representative of those for fresh tissue. The choice to use embalmed specimens for this study is further discussed in the limitations of the study.

Load relaxation appears to be greater for our study compared to those reported by Chaise et al (2003). The mean time to reach 80% load relaxation for the 'Intact' condition in our study was  $344 \pm 140$  s and by 1000s the load had relaxed to only 14.4% of peak load. Chaise et al (2003) reports that the load on the palmar surface took 20-30 minutes (1200-1800 s) to reach an equilibrium load of 23.5%. For the 'TCL exposed' condition, relaxation was greater than the 'Intact' conditions for both our study and that of Chaise et al (2003). In our study, 80% relaxation was reached in  $210 \pm 99$  s and reached 85.8% by 1000 s; while Chaise *et al* (2003) reports 83.2% relaxation was reached between 20 and 30 minutes. The greater load relaxation reported in our study may be due to the lower strain level imposed. A displacement of 4.5 mm was used here compared to 8.0 mm used by Chaise et al (2003). These results support the findings of others (Van Loocke et al, 2008 and Bonifasi-Lista et al, 2005) in that the rate of relaxation is greater for smaller levels of strain. Some care must be taken though in making these comparisons as the results reported by Chaise et al (2003) are based on one fresh frozen specimen and this may not be entirely representative of the normal population.

#### 5.1.1 Tissue layer differences

As stated previously, the peak load increased with each dissection level and this may give an indication of the load absorption capability of each layer of tissue above the TCL. With regards to the results for the Combined Group, the 'intact' condition recorded a peak load of  $68.02 \pm 42.87$  N which is 53.77 N lower than 'skin removed'  $(121.79 \pm 64.33 \text{ N})$  and 111.62 N lower than 'TCL exposed'  $(179.64 \pm 56.84 \text{ N})$ , while 'skin removed' was 57.85 N lower than 'TCL exposed'. The layers of the skin and the subcutaneous adipose provided a large amount of load absorption (though not significant different) while the combined soft tissue layers provided a significantly large amount of load absorption ('Intact' vs. 'TCL exposed'). It is to be expected that as tissue thickness increases the load dissipation would also increase (Smalls et al, 2006). A large proportion of the load recorded on the 'Intact' hand can be possibly attributed to the layers of the dermis and epidermis. Miller-Young et al (2002) found peak stresses on human calcaneal fat pads of 21.3 kPa (0.0213  $N/mm^2$ ) when 10 mm thick by 8 mm diameters fat pad sections were compressed to 40% strain. In comparison, a peak stress of 866 kPa (0.866 N/mm<sup>2</sup>) was calculated for the 'Intact' condition in this study (based on a load of 68.02 N divided by indenter area of  $78.54 \text{ mm}^2$ ). The larger stresses recorded in our study may be attributed to the skin layer over the subcutaneous adipose tissue. Miller-Young et al (2002) found the calcaneal fat pad samples to stress relax very rapidly, attributing it to the high unsaturated fatty acid ratio of the fat pads found in load bearing areas.

The effect on recorded peak load due to removing of one of the muscle groups is of particular interest. Within the Hypothenar Group, there was no difference in peak loads between the 'Skin removed' ( $115.48 \pm 75.39$  N) and 'Hypothenar removed' ( $115.71 \pm 64.90$  N). While within the Thenar Group, the peak loads for 'Skin removed' ( $128.10 \pm 67.40$  N) was significantly less than 'Thenar removed' ( $228.15 \pm 101.18$  N). This indicating that the thenar muscle group's attachment onto the TCL absorbs far more load than the hypothenar attachment. Though not recorded objectively, visual inspection of the specimens during dissection did reveal that the thenar attachment on the TCL was visibly thicker and covered a greater area of the ligament compared to the hypothenar attachment. Unfortunately these parameters were not recorded and this is an obvious limitation to this study.

Another notable difference (though not significantly) was the longer 80% relaxation times for the 'Intact' conditions compared to all other conditions and across groups. For the Combined Group, the 'Intact' condition  $(344 \pm 140 \text{ s})$  took 123.3% longer to

reach 80% relaxation than the 'Skin removed' condition  $(154 \pm 91 \text{ s})$  and 63.8% longer than 'TCL exposed'  $(210 \pm 99 \text{ s} \text{ (outlier removed value)})$ . Within the Hypothenar Group, the 'Intact' condition  $(293 \pm 171 \text{ s})$  took 138.2% longer to reach 80% relaxation compared to 'Hypothenar removed'  $(123 \pm 60 \text{ s})$ , while in the Thenar Group, the 'Intact' condition  $(422 \pm 5 \text{ s})$  took approximately twice as long as the 'Thenar removed' condition  $(206 \pm 125 \text{ s})$ . A possible explanation for this may be the effect of the skin layer causing the longer relaxation times. Crichton *et al* (2011) found that the stratum corneum and dermal layer of mouse skin reached a plateau at about 40% relaxation. The author posited that this maybe due to the lower hydration level (~ 15%) and stiffer nature of the stratum corneum compared to the deeper layers. As mentioned earlier; the variability in specimen quality and use of embalmed specimens in this study should be considered when interpreting the results. There was a notable variation in the level of embalming fluid content among the specimens tested in this study; presenting difficulties in controlling for moisture across the specimens.

#### 5.2 Stiffness

The stiffness of the 'Intact' condition  $(8.5 \pm 6.2 \text{ N/mm})$  in the current study is similar to that reported in the literature. Chaise *et al* (2003) reports palmar surface stiffness values of 15.6, 8.2, and 6.8 N/mm for 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> trials respectively. The author discusses only the 2<sup>nd</sup> trial values as they are closer to those of the 3<sup>rd</sup> trial, surmising that the large decrease from the 1<sup>st</sup> trial scores are due to preconditioning. However, the author also reports that preconditioning cycles were included at the start of all trials. For this reason, the values from the first trial will also be considered here. Zheng *et al* (2006) reported stiffness values for tissue covering the TCL of 3.57 N/mm and 18.98 N/mm when indenting with 0-5 N and 5-20 N loads respectively, and reporting a mean value of  $6.72 \pm 2.10$  N/mm. These values are comparable to those in the current study despite lower indentation loads being applied with an ultrasound probe and to live human tissue also.

At the 'TCL exposed' level, a stiffness 3.2 times greater  $(27.05 \pm 12.11 \text{ N/mm})$  than at the 'Intact' level was recorded in the current study. Again, this is comparable to those reported elsewhere. Chaise *et al* (2003) reported a stiffness values 3 to 3.6 times greater for their TCL-exposed condition (29.8 to 46.0 N/mm). Zheng *et al* (2006) recorded stiffness values for the tissue layer between the TCL and floor of the carpal tunnel of up to 21.76 N/mm, while Holmes *et al* (2011) report a stiffness value of  $35.9 \pm 2.8$  N/mm for their TCL exposed condition. These values can be considered comparable if consideration is given to the range of the individual scores for the current study (17.11 to 47.23 N/mm). The relatively high variation in stiffness values among the specimens is most likely attributable to variability in interspecimen quality and condition, and to the relatively small sample size. (n=6).

#### **5.2.1** Tissue Layer Differences

Across all groups stiffness generally increased as tissue layers were dissected away. The differences though are not significant with the exception of 'Intact' vs. 'TCL exposed' for the Combined Group. A similar pattern was found for stiffness values between the layers as was seen for the peak load values.

Within the Hypothenar Group, the removal of the hypothenar muscle group had no effect on the stiffness recorded ('Hypothenar removed':  $14.19 \pm 5.21$  N/mm; 'Skin removed':  $13.24 \pm 6.38$  N/mm), while in the Thenar Group, removal of the thenar muscle group resulted in a 49.4% increase in stiffness compared to 'Skin removed' ('Thenar removed:  $30.73 \pm 17.17$  N/mm; 'Skin removed':  $20.57 \pm 10.13$  N/mm). Again, the reason for this difference may be due to the extent to which the muscle fibres of the thenar eminence traverse the TCL and the thickness of this insertion layer. The viscoelastic properties of a layer of muscle fascicles above the TCL are likely to dissipate energy and result in a lower recorded value for stiffness. However, it is important to consider the orientation of the muscle fibres in relation to the compressive force. In the current study, indentation is perpendicular to the palmar surface. Therefore, the compression of the muscle fibres of the thenar muscle group will be in a cross-fibre direction and, as such, stiffness should be reported for cross-fibre direction and not axial tensile stiffness (Van Loocke et al 2008). Greater stiffness may be observed in a cross-fibre direction compared to with-fibre direction when strain rates are low ( $<5\%^{-2}$ ) as was the case in this study (average of 4.2% s<sup>-1</sup>).

An important consideration when analysing the results of the tissue properties on the exposed TCL is the effect of tissues in the carpal tunnel and carpal arch itself. Indentation of the exposed TCL when the tunnel contents are intact involves compression of the tunnel contents too, and their response may be affected by, what Zheng *et al* (2006) describes as the boundary conditions. To an extent the three bony walls of the carpal tunnel present a solid boundary that limits the lateral and dorsal expansion of the tunnel contents during indentation; with only the proximal and distal tunnel openings allowing some expansion of tissue. This 'confined compression' may subsequently have an effect on measured variables.

In this current investigation, one specimen was tested in a 5<sup>th</sup> trial following removal of the tunnel contents ('TCL only'). Peak load (19.80 N) and stiffness (3.14 N/mm) values were much lower than 'TCL exposed'. A similar finding was also reported by Chaise *et al* (2003) when they tested their fresh frozen specimen with the tunnel contents removed (peak load: 7.4 to 10.1 N; stiffness: 14.5 to 21.7 N/mm). At first thought it might be said that these values are solely attributable to the TCL; however, this still may be somewhat inaccurate. The carpal arches of the specimens tested in the current study were not immobilised in any way; and it is possible that indentation of the TCL may also deform the carpal arch (Li *et al*, 2009; Xiu *et al*, 2010) rather than isolate the deformation to the TCL. An argument against this notion is that Holmes *et al* (2011) found no significant difference in carpal arch width at rest (29.3  $\pm$  0.5 mm) and at peak load (28.7  $\pm$  0.5 mm). However, the carpal arch width reported at peak load in their study is with the carpal tunnel contents still intact and this many prevent any large increase in concavity of the carpal arch when loaded in this manner.

#### 6 Conclusion

The use of embalmed specimens for the determination of the mechanical properties of the soft tissue layers above the TCL is not recommended. Large variability in embalmed specimen quality can present erroneous results for peak loads and stiffness. However, the viscoelastic properties of embalmed specimens appear to be similar to those of fresh frozen tissue. The skin and adipose layer ('Intact' condition) showed slower load relaxations than any other tissue layer, while the muscular layer (the thenar muscle in particular) showed the fastest relaxation. The Thenar muscle group insertion on the TCL also provides more load absorption compared to that for the hypothenar muscle group. Peak loads and stiffness generally increased as each soft tissue layer was removed.

The test protocol used in this study presents limitations in attributing specific properties and characteristics to any one structure; and any characteristics determined (from the 'TCL exposed' level of testing) should be for the carpal tunnel complex as opposed to the TCL only.

#### 6.1 **De-limitations**

The decision to use embalmed specimens in this study was mostly one of convenience. The author recognises that the mechanical properties of embalmed tissue are not wholly representative of *in-vivo*, or indeed, fresh frozen human tissue. Embalmed specimens were readily available for testing via the Laboratory of Human Anatomy, University of Glasgow; and present less of a biological hazard. It was the original intention of the investigation to include at least two fresh frozen specimens to allow some comparisons to be made with the results of the embalmed specimens. However, fresh frozen specimens were not available to the researchers for testing within the time constraints of this study.

The age of the donors always requires consideration in such investigations since research has found that the stiffness of human soft tissue generally increases with age

(Agache *et al*, 1980); however, the age range of the specimen donors in this investigation is quite small (70 – 86 years of age, mean  $\pm$  SD: 82  $\pm$  6.29 yrs). The decision to test only one specimen for the 'TCL only' condition was due to the test protocol used in this investigation. To adequately test the TCL in isolation using the current indentation methodology would require immobilising the carpal arch and measuring changes in carpal arch width. Time constraints for this project prevented adequate pilot testing to develop a suitable protocol and would require that all conditions be tested with an immobilised carpal arch. The researcher preferred to test the specimens with the developed protocol used to better replicate *in-vivo* loading; as was used by Holmes *et al* (2011).

The inclusion of the 'Thenar removed' and 'Hypothenar removed' tests was chosen to attempt to determine their contribution to the mechanics of the carpal tunnel complex. This resulted in the requirement to split the specimens into separate groups so that adequate comparison of mean scores between conditions could be performed; and subsequently reduced the sample size to three specimens per group. The author acknowledges that as a result of the smaller sample size, the power of any statistical analysis performed is compromised.

### 6.2 Limitations

Throughout the investigation various factors presented themselves that may have had an effect on the outcomes of the study.

A one hour recovery period between trials was implemented to control for hysteresis effects among the specimens. However, due to complications out with the control of the investigator; one specimen was tested over two consecutive days. Resulting in a period of 17 hours recovery between 'Skin removed' and 'Hypothenar removed' trials. This did not appear to have an adverse effect on the data recorded following the extended recovery period as the results for peak load and stiffness for that specimen were close to the mean of the group.

The method used to secure the hand specimens to the platform may have an effect on the data obtained. Increased pressure by the restraining bars on the flexor tendons proximal and distal to the carpal tunnel may cause a 'bunching up' effect of the flexor tendons within the tunnel; furthermore, clamp pressures were not controlled for between trials.

It was the original intention of the author to compare the thickness of each layer of tissue to the results obtained at the respective levels of dissection. Unfortunately, an oversight by the author resulted in tissue layer thickness measurements not being fully recorded. Only 'Intact' palm thickness and 'TCL exposed' palm thickness were measured. As a result of this, and due to variability in specimen quality, it was decided not to perform correlation analysis between the outcome variables and anthropometric data. This is something that could be included in future investigations.

The test protocol developed for this investigation was inferred from those of similar studies (Chaise *et al*, 2003 and Holmes *et al*, 2011). However, as an aim of the investigation was to determine the mechanical and viscoelastic properties of the TCL and carpal tunnel, in order to further our understanding of the biomechanics of the wrist. The test protocol used does not adequately isolate the TCL and hence it is not possible to fully attribute any of the results obtained solely to the transverse carpal ligament.

### 6.3 Future Recommendation

Future research into the mechanical and viscoelastic properties of the TCL would need to better isolate the ligament, perhaps through dissection of the ligament and its four attaching carpal bones. The structure could then be mounted to fully immobilise the insertions of the TCL so that the ligament can be tested in isolation. A larger sample size of fresh cadaveric tissue is also recommended. Accurate, individual tissue layer properties should also be obtained through testing the tissues in isolation before comparing to those obtained here for validation.

Future studies into the biomechanics of the carpal arch should investigate the effects of the tunnel contents on the deformability of the carpal arch; with and without an intact TCL.

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## **Appendix A: Material Photos**



Figure 9: Custom made 10 mm diameter aluminium indenter



Figure 10: Custom made aluminium specimen platform. Dimensions: overall width: 250 mm; length: 400mm; depth: 15mm; bolt grooves: 25mm indent from side, 300mm length, 8mm width. Restraining bars (x3): width: 32mm; length: 233mm; depth: 10mm. Bolts: 8mm threaded bolts with wing nuts.

# **Appendix B: Indenter Loacation Guide**



Figure 11: Location of TCL centre of intact hand

# **Appendix C: Dissection level Images**



Figure 12: Intact Hand with TCL centre located



Figure 13: Skin, subcutaneous adipose and palmar aponeurosis removed



Figure 14: Thenar Muscle Group Removed



Figure 15: TCL Exposed

## **Appendix D: Anderson-Darling Normality Tests**

Normality graphs for Peak Load






















**Normality Graphs for Stiffness** 























## **Normality Graphs: Relaxation Time**





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## Appendix E Homogeneity of Variance graphs



















## Appendix G: Raw Anthropometric Measurements

Specimen No.	Thenar or Hypo- thenar Group	Wrist Width	Wrist Thickness	Palm Width with Thumb	Palm Width	Palm Thickness	Palm Depth TCL Exposed	Tissue Layer Thickness
1	Н	63.67	42.99	102.18	83.16	51.12	41.42	9.70
2	т	65.57	44.20	96.16	80.50	48.72	38.7	10.02
3	т	65.11	43.01	97.05	86.35	46.96	38.31	8.65
4	Н	62.45	41.00	84.95	77.72	40.07	35.32	4.75
5	т	60.54	36.99	75.67	76.76	37.35	31.09	6.26
6	Н	60.47	47.40	96.36	82.15	47.32	33.92	13.40
Mean		62.97	42.60	92.06	81.11	45.26	36.46	8.80
SD		2.20	3.46	9.82	3.57	5.35	3.73	3.04

Table 7: Individual spcimen anthropometric measurements for the hand and wrist.

 Table 8: Individual specimen antropometric measurements; TCL dimensions

Specimen No.	Thenar or Hypo- thenar Group	Distal Thickness	Proximal Thickness	Distal Width	Proximal Width	Mid Width	Ulnar Length	Radial Length	Mid Length
1	н	1.82	1.17	23.72	28.95	22.99	22.39	22.59	20.46
2	т	1.55	1.59	25.21	29.42	29.27	20.88	19.68	16.83
3	т	1.51	1.56	18.67	23.75	23.68	20.06	18.94	17.45
4	н	1.58	1.06	22.7	24.43	21.67	25.74	22.02	20.34
5	т	1.54	1.1	27.23	26.29	25.11	22.97	19	16.38
6	н	2.15	1.72	25.44	28.32	27.84	22.4	20.45	16.03
Mean		1.69	1.37	23.83	26.86	25.09	22.41	20.45	17.92
SD		0.25	0.29	2.97	2.41	2.94	1.96	1.55	1.98