Looking into Carpal Tunnel Syndrome

Prediction and Improvement of Clinical Outcome



V.J.M.M. Festen-Schrier

LOOKING INTO CARPAL TUNNEL SYNDROME: PREDICTION AND IMPROVEMENT OF CLINICAL OUTCOME

Een kijkje in carpaletunnelsyndroom: voorspellen en verbeteren van klinische uitkomst

Verena Johanna Martina Maria Festen-Schrier

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Table of contents

CHAPTER 1.	General introduction	7
PART I		**31**
CHAPTER 2.	The biomechanics of subsynovial connective tissue in health and its role in carpal tunnel syndrome	33
CHAPTER 3.	Subsynovial connective tissue development in the rabbit carpal tunnel	51
PART II		**69**
CHAPTER 4.	Reliability of ultrasound speckle tracking with singular value decomposition for quantifying displacement in the carpal tunnel	71
CHAPTER 5.	Relative motion of the connective tissue in carpal tunnel syndrome: the relation with disease severity and clinical outcome	89
CHAPTER 6.	Median nerve transverse mobility and outcome after carpal tunnel release	109
CHAPTER 7.	Shear wave elastography of the median nerve: a mechanical study	133
PART III		**155**
CHAPTER 8.	Ultrasound-guided hydrodissection with corticosteroid injection in the treatment of carpal tunnel syndrome: a pilot study	157
CHAPTER 9.	An incisionless ultrasound-guided carpal tunnel release technique	177
CHAPTER 10.	Minimal clinically important difference is lower for carpal tunnel syndrome patients undergoing injection versus surgery – letter to the editor	193
CHAPTER 11.	Better patient-reported experiences with health care are associated with improved clinical outcome after carpal tunnel release surgery	199
CHAPTER 12.	General discussion	215
CHAPTER 13.	Summary	231
CHAPTER 14.	Nederlandse samenvatting	237
APPENDICES		243

1

CHAPTER 1.

General introduction

Epidemiology

Carpal tunnel syndrome (CTS) is a compression neuropathy of the median nerve at the level of the carpal tunnel (Fig. 1, next page) and is the most common surgically treated hand condition. There is no clearly defined "gold standard" for the establishment of the diagnosis[1], which is reflected in the range of definitions and subsequent variations in reported incidence numbers. However, population-based prevalence numbers range between 1-5%, with an estimated 3:1 ratio of women versus men and a peak incidence between 40-50 years of age[2-4]. The lifetime risk of developing CTS has been reported to be as high as 10%[5]. A population-based study done in Olmsted County, Minnesota showed a continuous increase in the rate of CTS diagnosis in the decades between 1980 and 2005 and attributed that to improved methods of diagnosis as well as a surge in general public awareness of the condition[3]. With an associated annual cost of over $2 billion[6] and a median of 30 days of work missed[7], CTS represents a relevant socio-economic burden.

Clinical presentation

Although there is a wide variation in reported symptoms, the classical presentation consists of tingling (paresthesia), numbness (hypoesthesia) in the distribution area of the median nerve, often with nocturnal exacerbation. Patients will describe the inability to perform tasks that require fine motor skills involving the median nerve-innervated thenar muscles, or due to hand numbness, and will frequently complain of, dropping things, and will note symptomatic relief that comes with shaking of the hand. The timeline fluctuates with sensory symptoms presenting with an episodic character and exacerbation after a period of provocative hand usage, but it can transit into a progressive worsening of symptoms[8]. Just over half of cases have a bilateral presentation, and most patients present first with their dominant hand[9]. Finally, there are indications that in a relevant proportion of patients (~23%) symptoms resolve without active interference[10], which complicates treatment choice. With not all patients seeking treatment, there is a reservoir of patients with CTS symptoms who do not come in contact with the health care system and thus cause an underestimation of CTS prevalence in the general population. Conversely, if left untreated, some cases progress to a severe disease stage with full sensory loss, severe thenar atrophy and even skin ulcerations in the insensate fingertips. Prompt surgical treatment is then indicated, but outcomes are less predictable than in patients with more moderate presentation[1].

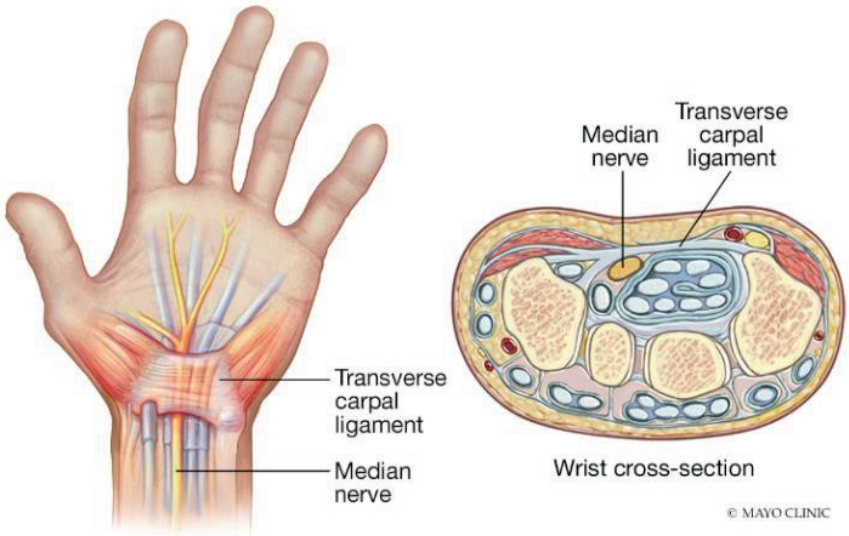


Figure 1: CTS is caused by focal compression of the median nerve at the level of the carpal tunnel.

Aims for this thesis

CTS seems like a relatively straightforward health care problem with clinicians having an abundance of diagnostic techniques and treatment options at their disposal. Yet, a relevant portion of patients have poor treatment outcomes while experts in the field disagree about the role of treatment options including steroid injections, and the merit of various surgical approaches.

This thesis sets out to investigate several CTS care elements, with an emphasis on the novel applications of ultrasound and a pathophysiological model inclusive of the nerve and the surrounding tissue. To structure the dissertation, we set out three aims, which correspond to a classical clinical timeline working from diagnosis to treatment.

Aim 1: *Assess the role of connective tissue in carpal tunnel syndrome as part of the pathophysiological mechanism.*

Aim 2: *Assess the potential for (dynamic) ultrasound to predict patient response to treatment in the clinical assessment of a CTS patient. What can be measured and what should be measured?*

Aim 3: A*ssess ways to improve outcome during and after treatment by (1) describing novel therapeutic approaches that utilize ultrasound and (2) determining factors associated with treatment success defined by patient-reported outcomes.*

The following sections of the introduction provide the reader with the status quo in terms of CTS pathophysiology and ultrasound usage as a starting point for the subsequent chapters.

CTS pathophysiology and the role of connective tissue

CTS is caused by compression of the median nerve at the level of the carpal tunnel, as has been well established in studies showing that patients with CTS have higher intra-carpal tunnel pressure and that elevating carpal tunnel pressure can induce typical symptoms of CTS in normal subjects[11,12]. The carpal tunnel is a confined space, with limited potential for expansion due to the presence of rigid carpal bones dorsally and laterally and the transverse carpal ligament palmary. Within the carpal tunnel are the eight digital flexor digitorum tendons, the flexor pollicis longus, and the median nerve. Therefore, any condition that causes an increase in volume will result in increased pressure on these structures.

Although systemic diseases as diabetes, rheumatoid arthritis, connective tissue disorders, and hypothyroidism[13], as well as post-traumatic causes and pregnancy[14], have been

associated with CTS, most cases remain idiopathic. Pathological analyses of idiopathic CTS patient samples have shown edematous changes in the endo- and perineurium of the median nerve as well as vascular proliferation, intra-neural fibrosis, and nerve damage consisting of decreased myelin volume and fiber degeneration[15-18]. Not just the nerve itself shows biological changes, the surrounding tissue is affected as well. This tissue is known as the subsynovial connective tissue (SSCT; also referred to as tenosynovium), which in adults shows a characteristic architecture of multiple horizontal sheets (Fig. 2) interconnected with collagen fibrils[19]. This layout allows for sequential recruitment of layers during tendon excursion thereby preventing excessive stretching and shearing of the nerve. Whether the multilayered organization arises spontaneously during development or is due to functional adaptation to mechanical loading is currently unknown.

Within the context of CTS, non-inflammatory fibrotic thickening of the SSCT has been described[20-23], but inflammatory cells are usually absent[17,18] indicating that synovitis alone is not the root cause. The combination of the SSCT fibrosis and increase carpal tunnel pressure is thought to be the basis of the intra-operative characteristic "hourglass" shape of the nerve within the carpal tunnel[24]. In addition, increased pressure in the carpal tunnel blocks the continuous flow of axoplasma, causing an increase in the nerve diameter at the carpal tunnel inlet, which is a useful feature for diagnostic evaluation[25].

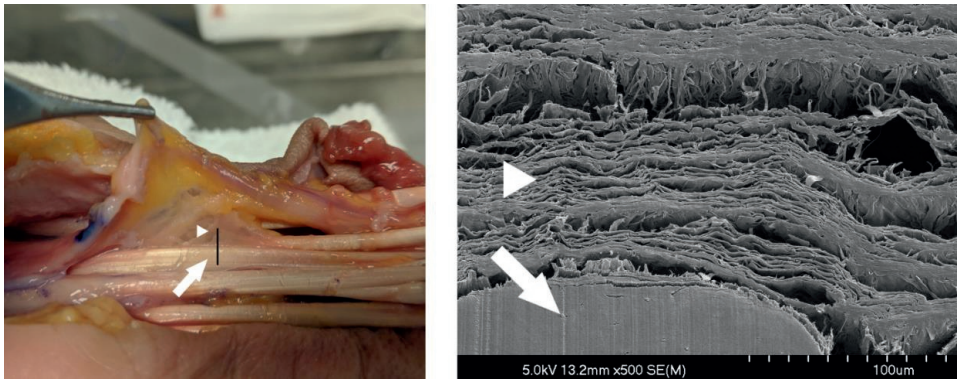


Figure 2: Macroscopic (left) and transverse scanning electron microscopic view, 500x magnified, taken at the location of the black line. Distinguishable is the flexor tendon (arrows) and the overlying SSCT (arrowheads).

Clinical evaluation of a CTS patient

The standard clinical work-up of CTS consists of the patient's history, physical exam, and in some cases, diagnostic tests. Sensitivity of the fingers can be tested using a two-point discrimination test, and provocative moves associated with median nerve neuropathy include Phalen's test, Tinel's sign, and the carpal tunnel compression test, but these all have been reported to be prone to low sensitivity and specificity[26-28] (Table 1). Specifically, in advanced CTS, these tests are associated with worse reliability[29]. As described above, the median nerve undergoes several pathophysiological changes

that affect both function as well as morphology. These changes can be measured with electrodiagnostic studies and ultrasonography (US), which can thus help support clinical decision making.

Table 1: Reported Accuracy Scores of Common Tests Used in CTS Evaluation.

Test	Sensitivity	Specificity
Phalen[30-33]	0.46-0.80	0.51-0.91
Tinel[30,32-34]	0.28-0.73	0.44-0.95
Carpal tunnel compression[30,31,35]	0.04-0.79	0.25-0.96

Evaluation tools

Nerve conduction studies

Clinical practice concerning the use of electrodiagnostic studies, including nerve conduction studies (NCS) and electromyographic (EMG) assessments, differs per country and institution. An NCS performed across the wrist can function as a proxy for nerve damage and can assist in establishing disease severity by combining median nerve sensory and motor latency and amplitude results. These tests are also useful to identify potential alternatives to the diagnosis of CTS, such as the presence of radiculopathy or polyneuropathy. A systematic review on NCS diagnostic value in CTS from 2002 reported sensitivity ranges between 56-85% and specificity 94-99%[36]. The 2016 guidelines of the American Academy of Orthopaedic Surgeons (AAOS) state that there is "*…moderate evidence that electrodiagnostic studies could aid the diagnosis of CTS…*"[1], leaving room for the interpretation in how to best use it clinically. Expert opinions vary between 'obtaining NCS for all cases' to 'no added value in the diagnostic work-up of CTS patients'[37,38]. Most commonly, practice lies somewhere in the middle with only obtaining NCS tests in case of non-classic CTS or if invasive treatment is considered. Currently, the Dutch guideline does not recommend the use of electrodiagnostic tests to diagnose CTS, provided that the patient has a classic CTS presentation[39]. In all other cases, due to its non-invasive nature, the guideline recommends using ultrasound as the first choice for additional diagnostic evaluation.

Ultrasound in clinical practice

The use of ultrasound as a diagnostic image modality has increased in popularity over the last two decades, as evidenced by the abundance of research published on the topic. The low-cost, non-invasive nature and relative user-friendliness are all arguments for its use, along with the potential to assess for structural nerve and carpal tunnel abnormalities. In practice, the visualization of the nerve is not challenging since it has different visual characteristics from the surrounding tendons. In the transverse view, the nerve can be identified as the oval-shaped hypoechoic superficial structure with a hyperechoic delineation (epineurium) and a "honeycomb"-like appearance due to the perineurial separations between the nerve's fascicles (Fig. 3).

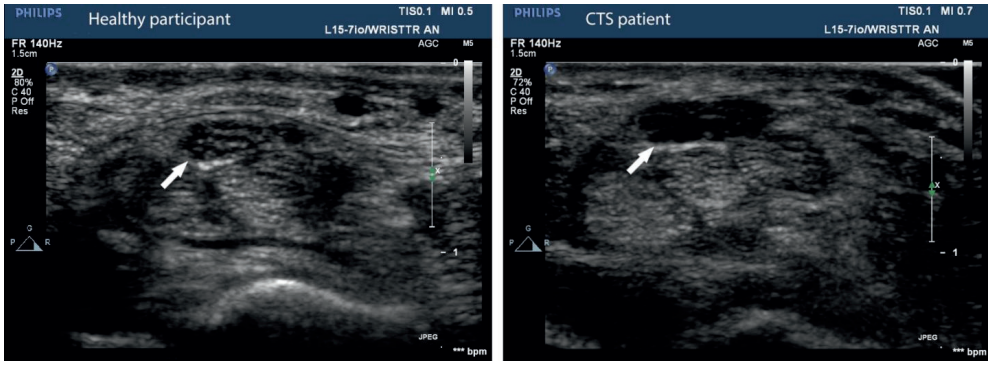


Figure 3: A typical transverse ultrasound image of the median nerve (white arrows) of a healthy participant (left) and a patient with CTS (right). Note the increase in size and less prominent appearance of the perineurium in the CTS patient. The radial side is on the left side of the screen.

For static Brightness-mode imaging of the nerve, it is well-established that there is a measurable increase in focal cross-sectional area of the nerve just proximal to the carpal tunnel in CTS patients, attributed to the earlier-mentioned edema and distal compression, but there is conflicting literature on how best to utilize that information. At present, no less than seven meta-analyses and systematic reviews have been published[25,40-45] on the assessment of the diagnostic potential of ultrasound in CTS, with conclusions varying between a negative recommendation for diagnostic purposes[40,43] to the suggestion for implementation as a first-line confirmatory test[41]. The most common threshold values for a positive diagnosis are between 9 and 10mm^2, and sensitivity and specificity rates are reported between 78-94% and 78-97%[40,41,44], respectively. However, most literature reviews include a remark on the limitations caused by variations in CTS definition and ultrasonographic measurement protocols. An expert panel convened by the American Association of Neuromuscular and Electrodiagnostic Medicine wrote an evidence-based guideline and concluded that there is Level A evidence that the median nerve cross-sectional area can be offered as an accurate diagnostic test for CTS with the accompanying recommendation that individual laboratories should establish scanning protocols and reference values prior to full clinical implementation. Additionally, this is confirmed in their closing remark that neuromuscular ultrasound evaluation allows for assessment of structural abnormalities which complements the functional information from NCS[25]. In contrast however, the most recent AAOS guideline, published in 2016, advices to "*not routinely use ultrasound for the diagnosis*"[1] based on twelve clinical studies that compared US to NCS, followed by a statement that a consensus on location and threshold values is required before ultrasound becomes effective as a diagnostic addendum (AAOS guideline; page 147).

In addition to nerve size, an array of different US-based parameters have been investigated as diagnostic markers, including vascularity, nerve size ratio, nerve flattening, nerve elasticity, and transverse carpal ligament bowing. With the high frame-rates and proficient resolution provided by the majority of today's musculoskeletal US machines, new types of measurements of the carpal tunnel structures provide new opportunities. One of these relatively new techniques is shear wave elastography, which quantifies tissue elasticity by measuring propagation velocity of shear waves through targeted tissue. Several studies have already shown increased stiffness of the median nerve in patients with CTS[46-50]. Clinical studies have also shown altered mechanical characteristics of the flexor tendons and median nerve in CTS patients versus healthy controls[51] indicating that there is a relation between disease and movement patterns. This could be due to the increase in nerve size in combination with the non-inflammatory fibrosis in the connective tissue. Diminished excursion of the median nerve has been found both in the transverse[52-57] as well as the longitudinal direction[53,58-60] making these parameters interesting topics of investigation for both diagnostic and prognostic purposes. Longitudinal assessment of the nerve and flexor tendons can be done reliably with an algorithm that uses a normalized cross-correlation analysis to track kernels in consecutive frames within a given region of interest[59,61]. Within the kernels, groups of pixels referred to as 'speckles', reflect scattered ultrasound waves caused by tissue inhomogeneity. Since these structural variations differ per anatomical tissue, patterns of speckles can be identified and used to track the motion of a structure (Fig. 4). Although we know that speckle tracking can be done in larger structures, we do not yet know to what inter-rater variability errors this measure is subjected, particularly in structures such as the SSCT, which at 1-2 mm in thickness is close to the resolution of the typical 10-15 MHz US transducer. The relation between disease severity and measurement of pathophysiologic changes in SSCT is also a novel research question which we will try to answer.

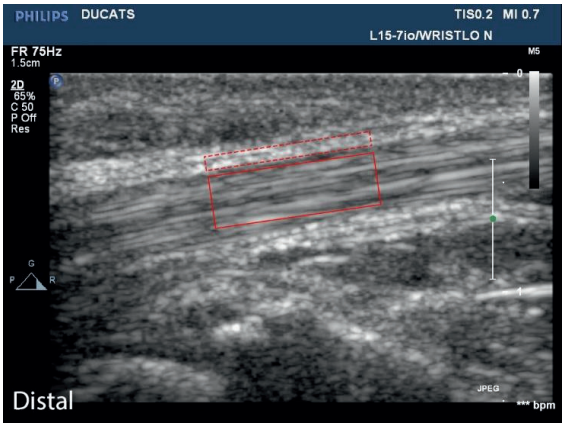


Figure 4: Longitudinal ultrasound of the carpal tunnel with artificial overlays of regions of interest indicating the position of the third superficial flexor tendon (uninterrupted line) and the overlying SSCT (interrupted line).

In the past 15 years, a series of studies on fibrosis in carpal tunnel connective tissue has been published at Mayo Clinic Rochester (Minnesota) and eventually led to a large scale prospective clinical study referred to as DUCATS (Dynamic Ultrasound assessment of CArpal Tunnel Syndrome). The main aims revolve around testing the prognostic potential of dynamic and static ultrasound-acquired parameters to identify those patients most or least likely to respond to treatment. In this thesis, we set out to see whether the results of this and concomitant studies would help to identify new patient-based prognostic measures by improving and incorporating novel image processing methods to quantify what we know happens biologically: decreased nerve elasticity, mobility, and fibrotic changes in the connective tissue.

Predicting CTS treatment outcome

So far, it has been difficult to determine which baseline factors best predict treatment outcome, both for injections and surgery. Especially due to the invasiveness of surgery and the reported long-term morbidity[62], adequate patient selection is essential. A review article on predicting surgical failure noted that a significant portion of articles showed poor methodological quality with many retrospective designs, variation in outcome assessment, small study groups and lack of follow-up[63]. Overall, generic patient-specific factors such as age, sex, or BMI are not closely correlated with outcomes. A recent prospective multicenter study showed that, in general, worse results can be expected for patients with a worse presentation[64]. A summary of reported baseline factors associated with outcome is reported in Table 2. The prognostic potential of nerve size determined by US has shown conflicting results on short-term outcome, and no relation with long-term outcome[65,66].

Table 2: Baseline factors associated with treatment outcome.

Improved…	
…surgical outcomes	**…injection outcomes**
• Lower symptom scores, • Less overall comorbidities, • Lower anxiety scores[64] • Absence of upper extremity comorbidities[67] • Female sex, • Good response to injection, • Moderate-severe NCS result[68,69] • Absence of abductor pollicis brevis atrophy[70] • Absence of legal case involving worker's compensation[71]	• Low NCS severity, • Higher injectate volume[72] • Lower baseline symptom scores[64]

Treatment and outcome measures

Based on the clinical context of the patient, there are several therapeutic pathways at the patient's and physician's disposal. Mild symptoms without clear evidence for neurogenic injury can often be treated conservatively with (nocturnal) splinting and activity modification[73,74]. With more severe symptoms, more invasive treatment is recommended in the form of a corticosteroid injection or carpal tunnel decompression (release) surgery.

Corticosteroid injection

There is clear evidence that one or multiple corticosteroid injections can relieve symptoms, but only for a short period(<1 month)[75]. The rate of treatment failure is considerable, with literature suggesting that only 1-3 out of 10 patients do *not* require additional intervention[76-78]. This indicates that there are opportunities for improvement, including the identification of patients likely to respond. In terms of improving the treatment itself, one of the complicating factors is that the method of action of the corticosteroid is unclear; the presence of a placebo effect cannot be ruled out[79]. Ultrasound guidance during injection allows for real-time visualization during the procedure, supporting standardized placement and spread of the injectate as well as prevention of iatrogenic nerve injury. US-guided procedures are associated with better clinical outcomes and lower secondary treatment rates[80,81]. In addition, the increased precision of the injection could allow for targeted disruption of connective tissue around the nerve using a method called "hydrodissection". Type and volume of corticosteroid treatment vary widely, but higher volumes are associated with decreased odds of reinjection[72], better clinical outcome[79,82], and increased nerve gliding[83]. However, so far, studies are lab-based, retrospective in design, or without clinically-relevant controls. This is why we set out to design a pilot study to look at patient satisfaction with higher volumes as well as acquire preliminary results on the additional benefit of hydrodissection above and beyond regular corticosteroid injection.

Carpal tunnel release surgery

Surgical intervention for CTS refers to the division of the transverse carpal ligament, thereby decompressing the tunnel, removing a prominent etiological factor. In the United States, about 70% of CTS patients receive surgery as their primary treatment[84]. The surgical costs alone add up to an estimated total cost of $2 billion on a yearly base[6]. After carpal tunnel release, the decrease in carpal tunnel pressure is immediately measurable[11], after which both symptom relief[85] and functional recovery of the nerve[86] progress over an extended period. Although there seems to be a learning curve at the resident level, significantly affecting outcome[87], annual volume at the consultant level appears less relevant[88]. The two most commonly-used techniques include the open and the endoscopic approach; the open approach involves an incision made over the proximal palm (with a possible extension over the distal forearm) after which the palmar fascia is visualized and the transverse carpal ligament can be divided under direct visualization.

The endoscopic approach utilizes an endoscope and specialized blades or cutting devices via one or two smaller incisions. Overall, in terms of symptom relief, there seems to be no superiority for either approach, although the endoscopic technique is associated with less post-surgical pain and faster return to work[89-92]. At the same time, possibly due to the decrease in visibility, there is a higher risk for nerve injury and incomplete ligament release compared to the open approach[93]. Patient satisfaction rates are higher when using the endoscopic approach[94]. Irrespective of approach, surgery is argued to be the only definitive solution for CTS and has a high overall success rate (75%[62]). However, as noted earlier, due to presence of variation in the definition of CTS and type of outcome, the reported success range is considerably wide (27-100%). About 8% of patients reporting that they are worse after the intervention[62]. Recently, ultrasound during surgery has become topic of investigation, possibly combining the visibility familiar to an open approach with the minimally invasive nature of the endoscopic technique. In order to divide the ligament, an abrasive thread can be used; the role of ultrasound in this novel technique will be a topic of investigation in this thesis.

Measuring outcome

In quality-of-healthcare evaluation, it is common to use quantifiable measures that relate directly to the treatment and the patient's symptom relief (e.g., time to surgery, complication rate, patient satisfaction). For assessment of carpal tunnel syndrome symptoms and interference in function, an often-used tool is the Boston Carpal Tunnel Questionnaire[95]. Since its introduction in 1993, it has been thoroughly tested for reliability, validity, and responsiveness[96] and is available in multiple languages. Patients can complete this form both for clinical as well as research evaluation. The outcome is then often defined as the change in score, with or without a minimal clinically-important difference. In this thesis, we wanted to look more closely at factors associated with patient-reported outcome. More specifically, the association between the patient's experience with the overall health care process and clinical outcome. In addition to health care experience, we wanted to see whether the type of treatment affects the way patients score outcome.

Thesis outline

To meet the aims set out at the start of the Introduction, this thesis has been subdivided into three parts. **Part I** focuses on the changes in connective tissue in the carpal tunnel in carpal tunnel syndrome patients and why it would be interesting to measure nerve mobility. In **Part II**, we expand on the actual US measurements and describe carpal tunnel mobility and nerve elasticity assessments to address Aim 2. **Part III** discusses improving clinical outcome by both enhancing treatment itself as well as our understanding of success to address Aim 3.

Part I CTS pathophysiology and the role of connective tissue

This section is designed to introduce in detail the relevance of the subsynovial connective tissue. **Chapter 2** is a literature review summarizing what is known about the SSCT and how this plays a role in CTS etiology. A hypothetical model with an initial shear strain injury and a subsequent loop of increasing SSCT damage is proposed. **Chapter 3** is a descriptive animal model study in which we looked at SSCT morphology in fetal, young and adult rabbit tissue where we try to relate changes in architecture to age and function.

Part II (Dynamic) Ultrasound in the clinical assessment of a CTS patient

Based on the critical role that SSCT is assumed to play in idiopathic CTS development, this section follows the hypothetical model proposed in **Part 1** and explores the potential of its non-invasive measurement. In **Chapter 4**, we set out to test the reliability of an improved speckle tracking algorithm to track SSCT movement. **Chapter 5** is the subsequent application of that algorithm in a clinical setting in which we assess the relation between SSCT movement and disease severity as well as potential prognostic potential. **Chapter 6** shares the prognostic aim of the previous chapter, but utilizes a different perspective, namely the cross-sectional view of the carpal tunnel to describe the mobility of the median nerve. Finally, **Chapter 7** introduces a novel ultrasound technique to assess elasticity as a biophysical attribute of the median nerve with the goal of future use for patient-specific, non-invasive disease characterization.

Part III Improving CTS treatment outcome

Having looked at the tools available to the physician before commencing to invasive treatment, this final part focuses on improving these interventions, but also looks into the subjectivity of treatment success when defined by the patient and how health care experience of a patients influence these. **Chapter 8** addresses a novel area of investigation by looking at hydrodissection of the SSCT and preliminary data on clinical outcome. **Chapter 9** describes a new surgical approach in which ultrasound is utilized to safely perform a threaded carpal tunnel release. Besides refinement of medical techniques, attention to factors that influence success from the patient's perspective is given in **Chapter 10** and **11**. **Chapter 10** looks at the difference in patient-reported outcome when patients receive an injection versus surgery and **Chapter 11** identified health care experience factors that are associated with surgical results.

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PART I

2

CHAPTER 2.

The biomechanics of subsynovial connective tissue in health and its role in carpal tunnel syndrome

Festen-Schrier, V. J. M. M., & Amadio, P. C. (2018)

Journal of Electromyography and Kinesiology, 38, 232-239

Abstract

Carpal Tunnel Syndrome (CTS) is the most common surgically treated problem in the hand. Aside from the neuropathy itself, the most common findings are fibrosis of the subsynovial connective tissue (SSCT) and increased intra carpal tunnel pressure.

Normally, the SSCT is a multilayer tissue interspersed among the carpal tendons and nerve. As the tendons move, successive SSCT layers are recruited, forming a gliding unit and providing a limit to differential movement. Exceeding this limit, damages the SSCT as has been shown in both cadavers and animal models. This damage leads to a non-inflammatory response with progressive fibrosis and nerve ischemia leaving the SSCT more susceptible to injury. Although the direct consequences for patients are not fully understood, ultrasound research shows that this fibrosis restricts median nerve displacement during tendon loading.

This article aims to provide insights into the mechanical properties of SSCT described so far and place it in the context of CTS pathophysiology. A theoretical damage model concerning the SSCT is proposed showing a chain of events and vicious cycles that could lead to the nerve compression as it is found in CTS. Although not complete, this model could explain the pathophysiological pathway of idiopathic CTS.

Introduction

Carpal tunnel syndrome (CTS) is the most prevalent surgically treated problem in the hand said to affect 1-3% of the population[1,2]. It is commonly described as a compression neuropathy, affecting the median nerve at the level of the transverse carpal ligament. CTS has been linked to vibrations, highly repetitive and forceful finger-, hand- and wrist motions[3,4] and an increase in carpal tunnel (CT) pressure[5–10], but the exact cause remains unknown in most cases. Surrounding the tendons and the median nerve in the carpal tunnel is a multi-layered structure called the subsynovial connective tissue (SSCT). Histological examinations of the SSCT from patients with CTS most commonly show non-inflammatory fibrosis of the SSCT within the carpal tunnel[11–18]. It has been suggested that injury to the SSCT due to specific differential finger movements causes damage to the SSCT fibers with subsequent deposition of fibrotic fibers[16]. The damaged SSCT becomes thicker and accumulates interstitial fluid, resulting in a restriction of the normal median nerve movement during tendon loading. This all increases carpal tunnel pressure, causing the neuropathy. Also, since the fibrotic SSCT is less capable to carry any new movement loads, it increases the likelihood of repetitive fibrosis, forming a self-sustaining vicious cycle.

This review aims to provide a detailed description of the SSCT as an anatomical structure, its (biomechanical) contribution to the carpal tunnel and the relation with the surrounding tendons and nerve. Finally, a pathophysiological model is proposed, summarizing current views on the role of SSCT in CTS.

The SSCT is part of a gliding unit

The carpal tunnel is a flat tubular tunnel located at the base of the hand. It is bordered by the carpal bones dorsally and covered by the flexor retinaculum on the palmar side. It is roughly 1-1.5 cm wide and encompasses the median nerve, eight digital flexor tendons (superficial and deep flexor tendons for digit II-V) and the flexor pollicis longus (Fig. 1). Two bursae limit friction between the osseous structures and the tendons during movement. The digital flexor tendons are enveloped by the ulnar bursa and the thumb flexor is bordered by the radial bursa with communication between the two being quite common[19]. The carpal tunnel shows a unique arrangement where not only both bursae provide friction prevention but also the tissue between the structures and the visceral layer of the bursa[20]. This specific connective tissue is described as subsynovial connective tissue and in a non-pathological state will loosely connect the tendons and nerve to each other (Fig. 2)[21]. Guimberteau described that a single gliding unit is formed by the flexor tendon, the carpal bones, the visceral synovium that moves against the parietal synovium (which on the palmar side is fixed to the flexor retinaculum), and the underlying SSCT[22]. The SSCT consists of multiple layers of collagenous fibers in which the blood and lymphatic vessels are richly represented. The fibers within the SSCT have been visualized by electron microscopy and show multiple sheets of fibrous tissue running parallel to the tendon[11], connected to each other by loose fibers, consisting

of collagen types I, III, IV, V and VI bundles[23]. During flexor tendon movement, the vertical fibers of the layer closest to the tendon will stretch out first. When the tendon movement is continued, the next horizontal sheet is enlisted and subsequently the next layer of vertical fibers will be stretched out etc., resulting in progressive recruitment of the layers (Fig. 3). During movement, the SSCT provides an adaptive scaffold for the vascular and lymphatic vessels and similar structural organizations and can be found throughout the body[24].

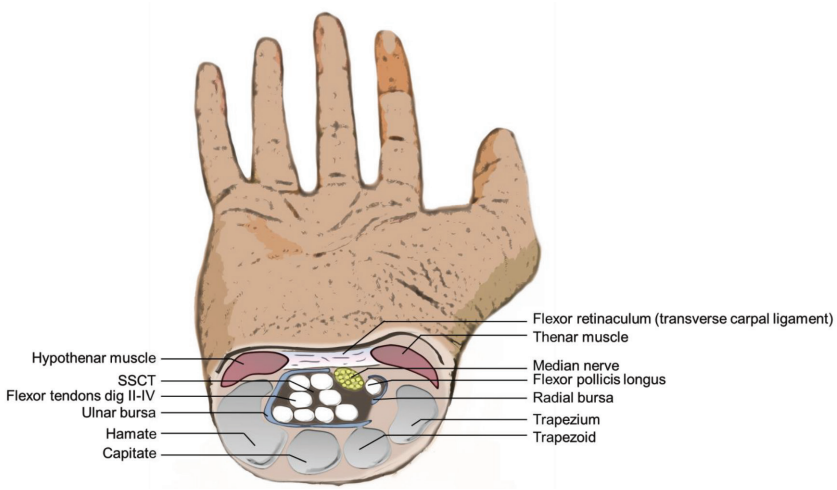


Figure 1: Cross section of the wrist with only the carpal tunnel structures visualized.

It has been shown that as the tendons move, the SSCT layers pose a limit to the differential movement of the adjacent tendons, inhibiting them more than what would be theoretically possible given normal tendon and joint excursions[25].

Interestingly, it has been suggested that there is a difference between the vertical orientations of individual SSCT fibers, which would result in only a certain amount of fibers processing the tension of tendon movement at any given time. If, due to for example excessive force, these vertical fibers tear, the next set of vertical fibers will carry the next tension load[26].

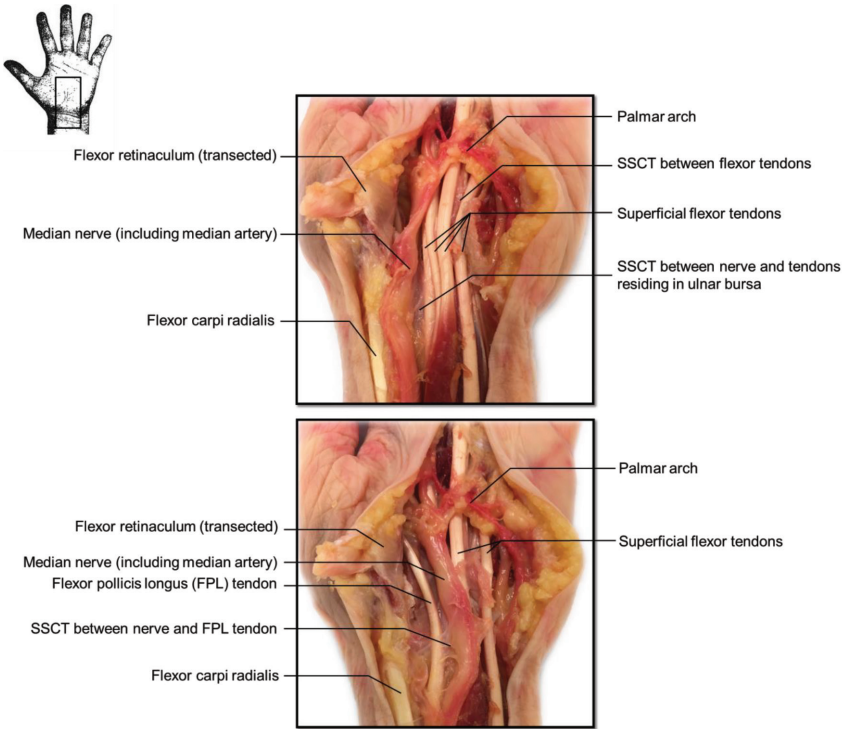


Figure 2: Anatomical dissection of a left cadaver wrist. Upper: The median nerve is placed on the radial side to show the deeper SSCT between both the nerve and the superficial flexor tendons as well as between the flexor tendons themselves. Lower: The median nerve is places on the ulnar side to show the SSCT between the median nerve and the flexor pollicis longus. Both pictures were taken after superficial dissection of the bursal tissue and superficial SSCT.

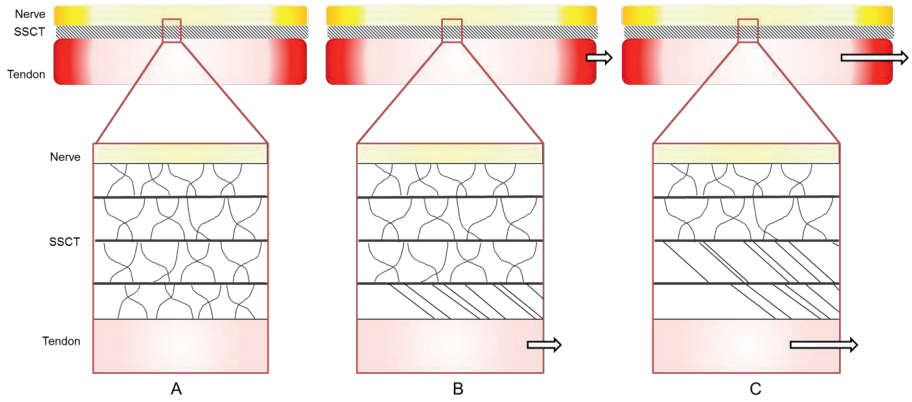


Figure 3: Schematic representation of sequential recruitment of SSCT layers during tendon excursion. On top, the distribution of three tissue layers is visualized from superficial to deep: the median nerve, SSCT and a flexor tendon. Below in the red rectangle, a more detailed version of the three layers is shown, showing the interconnecting vertical thinner fibers. A) In the starting position, without tendon excursion, the vertical SSCT fibers hang loosely. B) During the first part of tendon excursion, in this example to the right, the row of SSCT fibers closest to the tendon is loaded first stretching out the fibers. C) With increasing excursion, the second layer of SSCT fibers is recruited. If the motion would continue, all layers would be involved and eventually include the median nerve as well.

Biomechanical properties of SSCT

Research has shown that highly-repetitive and forceful motions are related to an increased risk of CTS development and therefore focus has been directed towards digit tendon motion and how it affects the surrounding tissue. Since the SSCT is the connecting layer between the nerve and the digital tendons, with the superficial flexor tendon of the third finger (FDS III) usually lying closest to the nerve, much of the research has been focused on assessing the consequences of FDS III tendon excursions on the SSCT structure, its force bearing potential and gliding resistance (GR). Although not an ideal setting, biomechanical testing on cadaveric hands has provided us with some interesting insights into the normal mechanical properties and responses. In such a setting, the hand is (partially) dissected and fixated to prevent uncontrolled movement. Flexion and extension are often mimicked by having an actuator pull and release load bearing tendons to a pre-programmed extent. A load cell is usually attached to measure force. The term gliding resistance is used to describe the amount of force necessary to displace a structure from point A to point B, like a tendon moving through the carpal tunnel. If the resistance to the displacement increases, this will be reflected in the increased necessary force and energy, meaning that high GR implicates that a relatively high amount of force (in this context usually measured with a load cell) needed to be applied. Another possible term to describe relative motion is shear index. This has been defined as the ratio of the difference in motion between structure A and B divided by the motion of structure A. In this context, structure A could be the FDS III tendon and structure B the surrounding SSCT[27]. This is a unit less description which decreases if two structures move in a similar fashion and increases the more they differ.

Starting with the normal physiology, Zhao et al. applied biomechanical testing on a cadaver model by using a custom build fixation apparatus. They showed an exponential increase in tendon GR relative to the distance of FDS III tendon displacement[28]. Findings were most profound in flexed wrist position and during isolated finger tendon motion. Seeing as the tissue surrounding the tendon is the main source of resistance, the SSCT thus seems to offer more impedance during longer excursions. This could be explained by the layered anatomical design and sequential recruitment of the SSCT. In order to look more detailed at these findings and possible damage to the SSCT, a similar set up was used to compare forces at separate cycles for different excursion rates. Irreversible damage initiated at 90% of the physiological tendon excursion, indicating that damage is possible within normal range of motion[25].

Oh et al. used Doppler ultrasound to measure the velocity at which the SSCT responds to FDS III tendon displacement. They showed that increasing the velocity of the tendon excursion, also resulted in the SSCT to move faster, but this happened at a slower rate compared to the tendon[29]. This data, and that from a comparable Doppler study[30], indicate that the SSCT does not move at a one on one ratio with the tendon, but rather has a response lag. This experiment was partially repeated in another study by Yoshii et al., who also introduced the concept of shear index as described above. They used

fluoroscopy, instead of Doppler, to measure excursion in mm and used that to calculate the shear index. Similar data were found, with the shear index being significantly higher at the highest tested speed for single finger motion[27]. So far a maximum tendon excursion velocity of 10 mm/s had been described, but Filius et al. also looked at 60 mm/s in order to mimic a more realistic fast-paced hand activity. At a low velocity they found the same initial damage at 90% excursion as was described before[25] but at high velocity, this damage occurred at 60% of normal excursion[31]. Finally, Kociolek et al. used even higher velocities, ranging from 50-150 mm/s. Increased tendon GR was found after exposure to increased tendon excursion and velocity underlining that SSCT provides a natural limitation that is more profound when the tendon moves faster and/or more[32].

Besides tendon excursion level and speed, the repetitive nature of finger motions also seems to increase the shear index between tendon and SSCT during single finger movement as tested in healthy male volunteers[33]. This effect was seen mostly in individual tendon movements.

Knowing that excursion repetition, velocity and distance affect the total GR within the carpal tunnel, a model was proposed by Filius et al. in order to isolate the different factors that sum up to the total GR. In this model, GR was defined as the summation of I. the resistance provided by the SSCT, II. the deformation of the tendon and III. the structural contact friction. This model allowed for differentiation between these three different components and their contribution to the total GR was measured at both a low (2 mm/s) as well as a higher (60 mm/s) velocity tendon excursion. The relative SSCT contribution to the GR ranged between 50% to almost a 100% at full physiological excursion and was most clearly present if in the experimental setting the fastest motions without relaxation time were used[34]. The SSCT influence on the decreasing total GR thus increases when moving the tendon relatively quickly and not allowing for SSCT recovery. If the SSCT is allowed to recover in between tendon motions, it has a chance to get back to its original shape and provide the same amount of GR for the next excursion. This behavior indicates a poro-elastic trait[35,36].

Epidemiological research first hinted that CTS is associated with high force, repetitive finger motions, which piqued interest in the anatomical structures within the carpal tunnel and their biomechanical influences on each other. Important to note is that the exact contribution of specific occupations and hand related activities are difficult to study. Epidemiological studies that aim to focus on isolating single causes (e.g. specific occupations, tasks or hobbies) often face difficulty accounting for confounding factors leaving conclusions on causality open for debate. In experimental designs, most of the normal physiological work has been done using cadaver hands which, although not an ideal comparison, did add to the understanding of movement ranges and limits. Tendon excursion, tendon speed, force, repetition of movement, and wrist position all seem to influence the total GR which makes these variables interesting to use in a comparison with the clinically idiopathic CTS situation.

SSCT and carpal tunnel syndrome

Chronic pressure elevation within the carpal tunnel is associated with nerve ischemia[37,38], but the cause of the elevation has been a topic of investigation. The fact that increased CT pressure and SSCT fibrosis are common findings in the SSCT of patients with CTS indicates that finding the cause(s) of the pressure difference and fibrosis might help illuminate some of the pathology behind CTS.

Animal models have been developed to mimic the increased pressure by either artificially increasing the carpal tunnel volume or decreasing the space. This provided valuable insight into the relationship between pressure differences and the neuropathy, but so far has not illuminated the cause for and/or maintenance of the nerve compression or ischemia. Combining the acquired data on the role of the SSCT and GR there was a need for a different animal model. Although rats have been successfully used to study CTS development[39], their SSCT structure differed from the human multi-layered SSCT as was also found for canine models[40]. Rabbits do have a similar SSCT structure and CTS models have been developed by either injecting a hypertonic glucose solution, or by inducing tendon injury[40–47]. The same rabbits, without the artificially induced CTS, were used to assess SSCT fiber thresholds and it was found that, like in humans, even low velocity tendon excursions within the physiological range can induce step-wise damage to the SSCT[26] and that this (micro tear) damage can indeed lead to progressive fibrosis[47,48]. The step-wise pattern fits the hypothesis that as tendons move, the connecting vertical SSCT fibers will sequentially be stretched out, up until the point where the load exceeds their bearing potential and will cause irreversible rupture of the fibers (Fig. 4). Cadaver work has added to that, that damage to the SSCT could arise within physiological movement range, implicating that SSCT rupture could be relatively common in humans[25,31,49]. This means that relatively small finger motions could already inflict SSCT damage, even though not everyone develops CTS. Therefore, examining SSCT derived from CTS patients would be the logical next step. Since it is undesirable to use undisturbed SSCT from living human control subjects for ethical reasons, most of the research on CTS has been focused on comparing CTS patients undergoing surgery versus cadaver wrists.

Looking in detail with light and electron microscopy at the scar formation in both cadavers and patient derived SSCT, tearing of the vertical fibers was seen including thickening of the horizontal SSCT sheets. This implies that after rupture, the fibers "stick" to the sheets, losing their bearing function and increasing the size of the sheets. Secondly, the most severe fibrotic SSCT changes in the patients were located where movement is most prevalent, closest to the tendon, suggesting that these changes may indeed be the result of a shearing injury[13]. On a molecular level, overexpression of transforming growth factor-β (TGF-β) and connective tissue growth factor and their receptors has been found in SSCT tissue in both a rabbit animal model and in CTS patients[50,51], suggesting that the mechanical vicious cycle of injury and fibrosis is exacerbated by overexpression of pro-fibrotic cytokines. This combination of vicious cycles- of injury and response with fibrosis, potentiated by overexpression of pro-fibrotic cytokines may

explain why the SSCT in CTS patients have increased elastin phagocytosis and collagen fiber size compared to healthy samples[12,23,52].

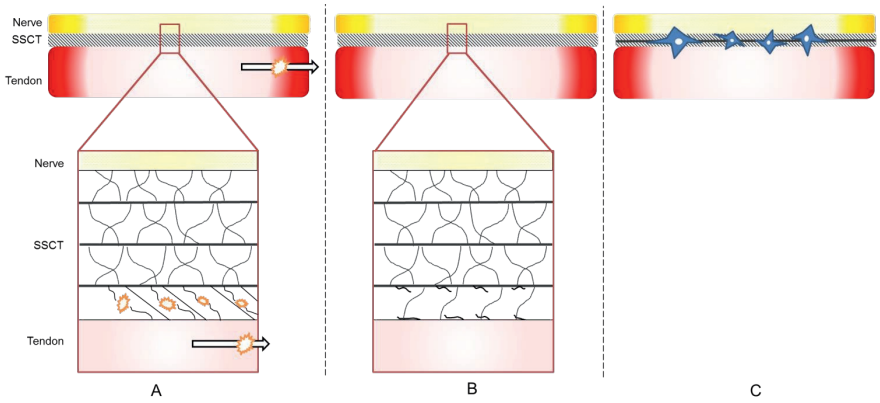


Figure 4: Schematic representation of the proposed self-sustaining SSCT injury cycle. A) External risk factors cause the interconnecting SSCT fibers carrying the highest load to rupture. B) The ruptured fibers are no longer functional during tendon excursion and will tend to "stick" to the horizontal sheets. C) The injury will induce a non-inflammatory reaction, amongst others activating fibroblasts, forming scar tissue, retention of interstitial fluid and cause subsequent increased carpal tunnel pressure. The fibrosis will also increase gliding resistance and inhibit the free movement of structures around the SSCT like the median nerve.

SSCT and fibrosis involvement on a macroscopic level was looked at by Ettema et al. by tagging the most superficial layer of the SSCT (visceral synovium) and the FDS III tendon with a surgical marker and following them during tendon excursion intra-operatively. No significant differences were found, but there was a trend visible showing a lagging difference between SSCT movements in CTS patients versus cadaveric controls[53]. Being able to visualize the relative SSCT motion and comparing healthy with CTS cases would be the next step. Dynamic ultrasound has been used to measure displacement differences in CTS patients versus healthy volunteers. In a transverse plane, movement of both the tendon and nerve within the carpal tunnel in CTS patients is inhibited[54–57]. Normally, the median nerve moves dorsally between the superficial flexor tendons during flexing motions but this restriction in movement in CTS patients limits the ability of the nerve to translate while the tendons are loaded (Fig. 5, left), resulting in its compression anteriorly between the flexor tendons and the flexor retinaculum (Fig. 5, right). Longitudinally, a decreased median nerve displacement, worsening with more severe nerve conduction study classification, was shown in a prospective study, but no significant differences in SSCT displacement could be found. The SSCT is a small structure of ~1mm[54], posing a challenge for current low-cost non-invasive visualization modalities to be visualized. Future studies should focus on using newer high-resolution ultrasound imaging techniques in order to track SSCT in order to measure a shear index.

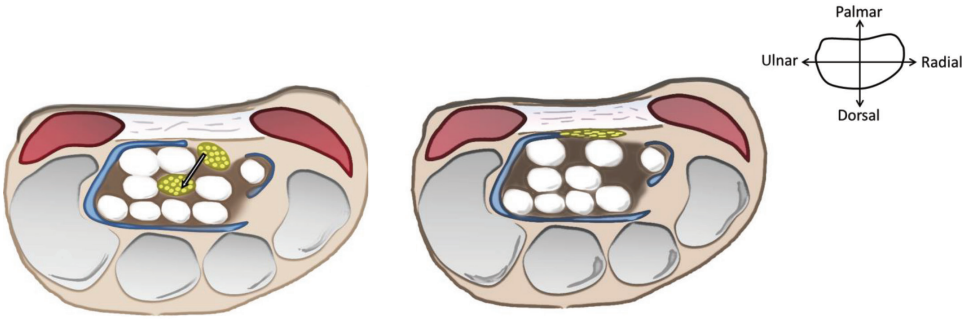


Figure 5: Left: Normally, during wrist and finger flexion the median nerve moves in a dorso-ulnar direction protected by the superficial tendons. Right: In CTS patients, the SSCT stiffens which inhibits this excursion and forces the nerve to translate in the path of least resistance which would be in palmar direction. It ends up between the dorsal surface of the flexor retinaculum and the loaded flexor tendons.

Also, increased SSCT thickness and increased shear strain between the SSCT and FDS III tendon has been found in symptomatic CTS patients compared to healthy controls[58], indicating that altered SSCT might be involved in symptomology. However, future studies with larger group sizes will need to show a more direct association. After *in vitro* carpal tunnel release, the shear index between tendon and SSCT decreases significantly (with the wrist in a flexed position), indicating that the release procedure not only decreases CT pressure but also shear strain[59].

Besides the known association with specific motions, elevated intracarpal pressure is also a familiar physiological difference found in CTS patients. Sud et al. showed that SSCT from CTS patients could absorb fluid at a much faster rate than that of the healthy controls[60]. Additionally, Osamura et al. showed that hydrostatic permeability of SSCT is relatively low, indicating that any leaked fluid (either blood from damaged vessels or lymph fluid) could be absorbed quickly by the SSCT after which it would be retained, contributing to the intracarpal pressure increase[61].

The chronological cause-effect model comprised of these findings might, in part, explain the pathogenesis of the nerve compression in idiopathic CTS: The carpal tunnel is a small space with multiple moving structures, facilitated by a gliding unit. Forceful, high velocity and repetitive finger and hand movements with differential movement of the flexor tendons induce shear stress which damages the SSCT. This damage results in SSCT fibrosis, stiffening, and enlargement of the tissue which increases carpal tunnel pressure. The increased pressure increases additional load on the tendons, adding shear stress to the already affected SSCT and together with the fibrotic SSCT prevents compensatory movement of the nerve to avert compression. The increase in pressure also induces episodes of ischemia and reperfusion, and alters SSCT fluid permeability. The already damaged SSCT would then be susceptible to additional damage, due to the chronic and repetitive nature of the aforementioned risks and consequences of the initial damage (Fig. 6). The chronic nature adds to the progression of asymptomatic median

nerve damage to a clinically relevant situation which is likely to worsen over time if no intervention is initiated.

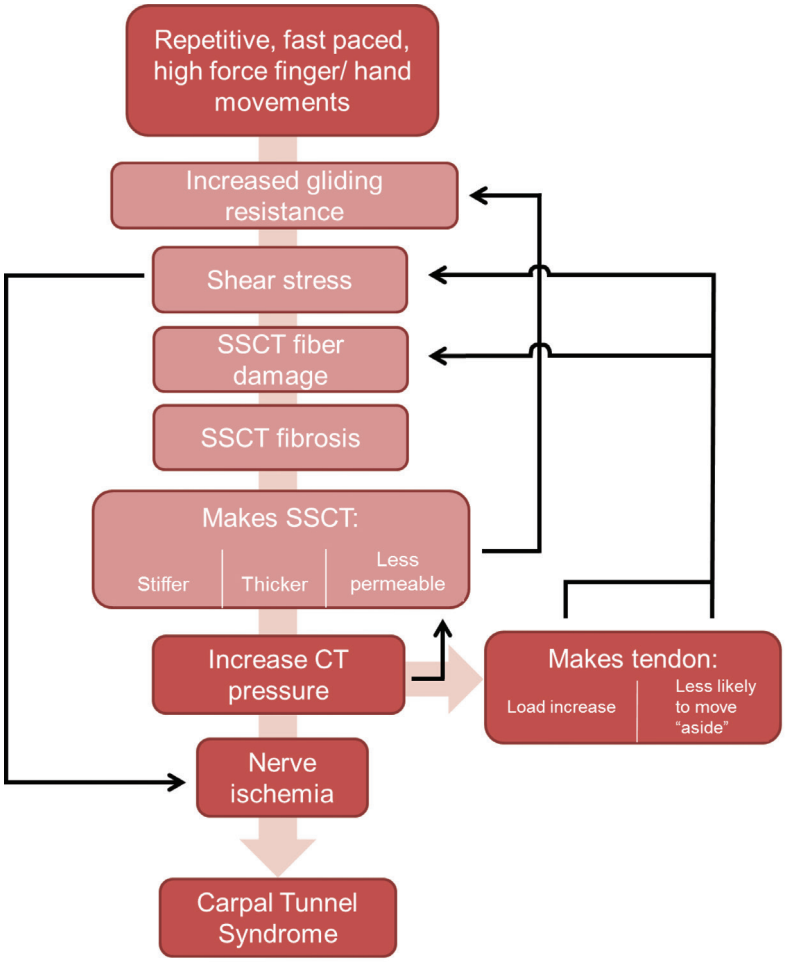


Figure 6: Different components of the proposed damage model: The lighter boxes are directly linked to SSCT and the darker boxes represent either causes or effects on surrounding tissue. The light arrow indicates chronological order; black arrows indicate a negative effect on earlier components which form pathological cycles.

Although this model provides a possible explanation for the pathophysiology of idiopathic CTS, it does not explain why not everyone with high force, repetitive hand activities develops a symptomatic carpal tunnel syndrome. Future research should focus more on inter patient differences and between CTS patients and non-CTS patients. This would help illuminate the precise role of SSCT and could aid in the prevention, prognosis, and treatment of CTS. Although surgical carpal tunnel release is an effective treatment, symptoms are not relieved to a satisfactory level in around 25% of cases[62], and although this percentage is prone to a large variation, it shows a gap that apparently is not treated by decreasing pressure alone. In light of preventing CTS, a finite element

model has been developed to help study the effect of various hand movements on SSCT stress and strain, and may help to identify potentially hazardous activities, which may then be modified to reduce the risk of developing CTS[63].

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CHAPTER 3.

Subsynovial connective tissue development in the rabbit carpal tunnel

Schrier, V.J.M.M.*, Vrieze, A.*, Amadio, P.C. (2020)

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Abstract

The carpal tunnel contains the digital flexor tendons and the median nerve, which are embedded in a unique network of fibrovascular interconnected subsynovial connective tissue (SSCT). Fibrous hypertrophy of the SSCT and subsequent adaptations in mechanical response are found in patients with carpal tunnel syndrome (CTS), but not much is known about the development of the SSCT. This observational study describes the morphological development of SSCT using histology and ultramicroscopy in an animal model at four time points between late-term fetuses through adulthood. A transition is seen between 3 days and 6 weeks post-partum from a dense solid SSCT matrix to a complex multilayered structure connected with collagenous fibrils. This preliminary data shows a developmental pattern that matches an adaptive response of the SSCT to loading and motion. Understanding the anatomical development aids in recognizing the pathophysiology of CTS and supports research on new therapeutic approaches.

Introduction

Carpal tunnel syndrome (CTS) is a very common compression neuropathy at the level of the carpal tunnel (CT), a fibroosseous channel in the wrist. CTS occurs most frequently in adults aged 45-55[1], and thus has a substantial socio-economic impact, with overall costs in the US alone reaching over $2 billion annually[2]. Most cases have no directly relatable cause and CTS pathophysiology is still a common topic of investigation.

The human CT consists of nine flexor tendons and the median nerve. These structures are enveloped by two synovial bursae and interconnected via a fibrovascular subsynovial connective tissue (SSCT). This connective tissue shows a unique arrangement in both humans as well as in animal models[3-5]. Multiple horizontal sheets surround each tendon to form a gliding unit (Fig. 1). The sheets are interconnected with smaller vertical fibrils that limit motion and prevent damage to the vascular and lymphatic structures[4]. This specific architecture also allows the absorption of mechanical stress as successive layers of SSCT are recruited sequentially with increased tendon excursion[6] preventing the nerve from excessive shear forces. The SSCT is hypothesized by some to play a central role in the pathophysiology of CTS[7-9]. Studies have shown that the SSCT is prone to irreversible damage even within physiological tendon excursion[6,7]. Under continued stress, the SSCT damage initiates a non-inflammatory fibrotic response as is seen in histological samples of SSCT from CTS patients[5,7,9-14]. Since movement and size of the SSCT can be measured non-invasively, it is an interesting topic of investigation for diagnostic and prognostic purposes[15,16].

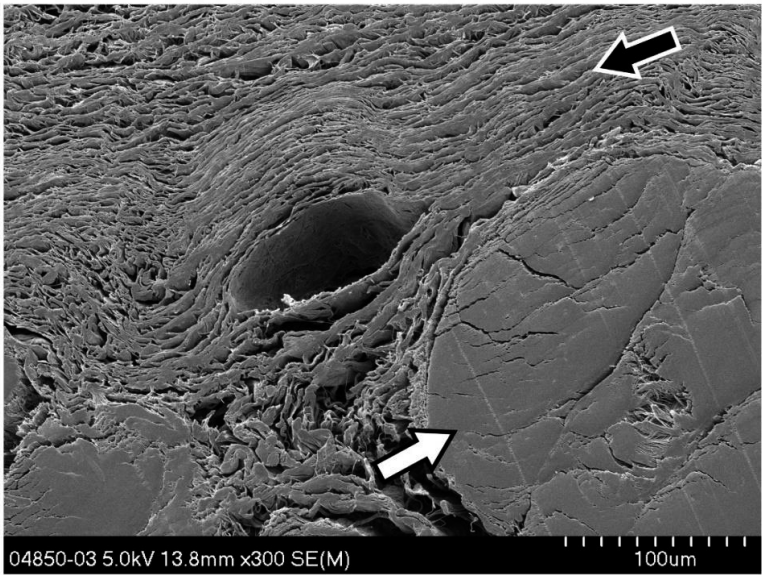


Figure 1: Transverse SEM view, x300, of a human superficial flexor tendon (white arrow) and the multilayered sheet organization of the SSCT (black arrow) as it is found throughout the carpal tunnel. A small vein present in the SSCT can be seen in the middle.

With the SSCT potentially playing a pivotal role in CTS development, a better understanding of its structural development over time is helpful. It is currently unknown whether the multilayer organizational structure of the SSCT develops purely in response to loading and motion over time or if it develops independent of stress loading during normal embryological tissue formation *in utero*. If the first case is true, it could be hypothesized that the SSCT starts as a simple structure that adapts over time due to finger, hand or wrist motion, similar to how bone and other tissue dynamically adapts to changes in force loading[17,18]. This could, in turn, suggest that the SSCT damage in CTS is a failure of an adaptive mechanism, which might be addressed by physical measures; similar to the way stress fractures can be avoided by avoidance of sudden increases in repetitive loading of bone. However, if the SSCT is present in its adult form at birth, prior to high force dexterity loading, it would mean that the histological characteristics as seen in CTS patients are less of a defective adaptation response but more fitting with a progressive damage model. Potentially, a preset amount of fibrils are established during gestation which, during a lifetime of hand use, are gradually lost to attrition. In this case, there would be less of an indication to apply activity modification as a treatment, with strategies better targeting minimalization of the subsequent formation of SSCT fibrosis. The question would remain why some people develop CTS whereas others can go a lifetime without any problems; these two scenarios pose different possible solutions for the management of CTS.

With the difficulty of obtaining human fetal tissue, using an animal model showing a similar micro-organization could help elucidate the development of the SSCT over time. A previous study described the comparability of the human SSCT to different animal models and concluded that the rabbit carpal tunnel was anatomically similar to humans[3]. This comparison study resulted in multiple CTS related rabbit studies[6,19-22]. Additionally, rabbit fetuses have been found to develop in utero in similar stages as humans[23]. Therefore, the rabbit model was chosen to visualize the organizational structure of the SSCT before and after it has been subjected to physiological stress and strain of normal life. This article contains subjective descriptions of rabbit carpal tunnel morphology based on macroscopic, microscopic and ultramicroscopic characteristics, starting from third trimester fetuses to fully mature rabbits. These findings can be used to help design animal studies focused on carpal tunnel pathology. Emphasis will be placed on structural development of the SSCT sheets and the connections between.

Materials and Methods

Animals

All animal protocols were approved by our institution's Animal Care and Use Committee (IACUC) and performed in accordance with the US National Research Council's Guide for the Care and Use of Laboratory Animals. Three male and three female adolescent (six weeks) New Zealand White (NZW; *Oryctolagus cuniculus L.*) and a pregnant NZW female at 27 days gestation were sedated intramuscularly with Ketamine (35mg/kg) and

Xylazine (5mg/kg) and then euthanized with an intravenous overdose of pentobarbital; seven fetus kittens were collected from the pregnant female postmortem. Additionally, six 3-day old rabbits and six 18-25 month old (four female and two male) NZW rabbit cadavers were donated from other IACUC approved studies from other laboratories; no interference between their research studies and our collected sample was expected. Sex in rabbits is not reliably visualized until 3-4 weeks of age therefore, sex was not noted for fetal and neonate groups. Immediately after sacrifice, both front legs were harvested at the point of the elbow and either frozen at -80°C or had the carpal tunnel excised and fixed according to imaging protocols below. Specimens were organized into the following age groups: fetal (27 day gestation; end of the third trimester), neonate (3 days), juvenile (6 weeks) and adult (18-25 months). To reduce the number of animals required, the group sizes were chosen for descriptive analyses only, and therefore objective quantification falls outside of the scope of this study.

Macroscopic features
Specimens were frozen at -80°C for at least 24 hours and cut with a diamond blade in transverse sections to acquire a full cross-sectional view of the carpal tunnel for macroscopic imaging under magnifications ranging from x2-10 (BLX51, Olympus, Japan). Macroscopic imaging of both fetal and neonate groups was not feasible due to limited tissue solidity and absence of discernable tissue characteristics at this age.

Microscopic features
Specimens underwent an *en bloc* style dissection of the carpal tunnel and flexor digitorum superficialis (FDS) of the third digit, taking care to not disrupt the SSCT. Sutures were placed at the proximal end for embedding orientation and to help maintain the integrity of the carpal tunnel. Samples were fixed in 4% neutral buffered formalin for at least 24 hours, dehydrated, cleared and subsequently paraffin-embedded. Then, 5 μm sections cut both transversely and longitudinally were mounted on slides for standard hematoxylin and eosin (H&E) staining and evaluated under a maximum of x200 magnification.

Scanning Electron Microscopy features
Fresh samples from each age group were prepared for scanning electron microscope (SEM) imaging. The carpal tunnel was dissected *en bloc* as described above with a suture to indicate orientation the tissue was fixed in Trump's fixative as per standard protocol[24] (1% glutaraldehyde, 4% formaldehyde and 0.1M phosphate buffer at pH=7.2). Samples were then treated with a series of dehydration and rinse steps using respectively ethanol and 0.1 phosphate buffer in a critical point dryer. Cross-sectional and longitudinal samples were mounted and sputter coated with gold-palladium; images were taken with a cold field emission scanning microscope at 3.0kV (Hitachi S-4-700, Hitachi High Technologies America, Inc., Pleasanton, CA, USA). In order to get both overview images as well as detailed organizational representations, magnifications between x80-8.00k were taken for independent evaluation by two reviewers with subsequent consensus through discussion. Results were subsequently confirmed by two senior researchers who have published on this topic.

Results

Gross Overall Anatomy – Human versus rabbit

The adult rabbit carpal tunnel has been described in detail before, including similarities with human anatomy[3]. In short, both carpal tunnels contain digital flexor tendons, the median nerve, in some cases a (persistent) median artery, and a transverse carpal ligament covering the volar side. In adult rabbits, the carpal tunnel is about 8mm proximal to distal compared to 21-30mm in humans. The transverse ligament has two harder triangularly shaped fibrocartilageous areas on the lateral edges and a thinner section overlapping the CT. A marked difference with humans however, is that the deep flexor tendon (FDP) in rabbits does not branch into the individual tendons proximal but distal to the carpal tunnel. Often, at the level of the CT, there are only three individual FDS tendons visible, of which one splits more distally into two. Additionally, the fifth metacarpal is located more proximally than in humans (dewclaw); the flexor pollicis longus lies dorsal to the FDP. The proximal carpal bones form the dorsolateral arch in which the CT lies. Rabbits have a total of nine carpal bones, with an extra central carpal bone in the distal row[25]. As seen in Figure 2, the carpal tunnel lies volar to the carpal bones and measures 5-6mm in width and about 4mm in height. In one of the adult front paws, the nerve split into a ramus medialis and ulnaris just before the CT, as described earlier by Ettema et al.. Based on our observations, the macroscopic CT layout of the juvenile rabbits was similar to that of the adult rabbits.

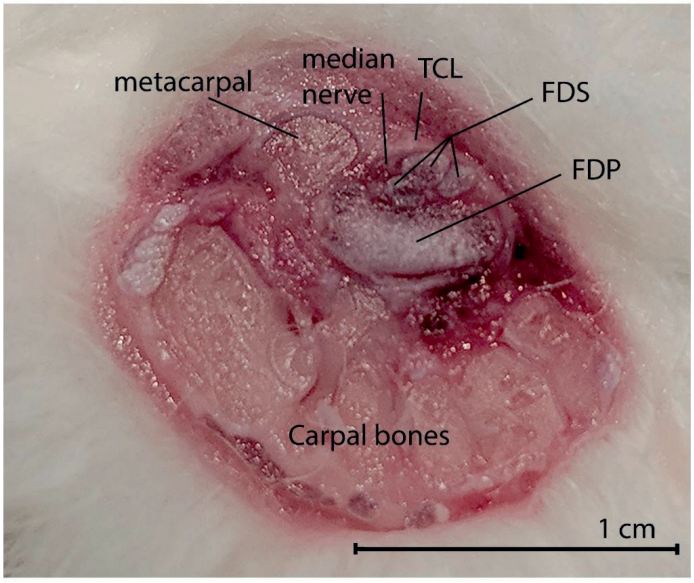


Figure 2: A macroscopic view of a transversely cut adult rabbit CT. Volar side at the top, radial side on the left of the image. There are three FDS tendons visible, superficial to the much larger singular FDP tendon. The median nerve is located just radial to the tendons.

Histological assessment of rabbit samples

Histological assessment of the fetal tissue showed fully developed carpal bones and the completed formation of the isolated carpal tunnel (Fig. 3A). The FDS tendons form a closely packed bundle superficial to the FDP. At higher magnification, the tendons display high cellularity with a small nuclei-matrix ratio, fitting the highly proliferative fetal stage (Fig. 3B, white arrow). A cellular lining of connective tissue was located circumferentially to the FDS bundle as well as in between the individual FDS tendons (Fig. 3B-C, black arrows). At 3 days post-partum, the tendons showed a decrease in cellularity and were more clearly defined compared to the fetal samples. Where the fetal samples had more densely packed tendon bundles, the post-partum samples showed more individualized tendons separated by a layer of connective cells. This connective layer between the individual FDS tendons was considerably smaller than the layer around the CT (Fig. 3E-F). At 6 weeks, all the carpal tunnel structures including the median nerve and the transverse carpal ligament had fully formed, comparable to the adult stage. The SSCT was present throughout the CT, again with a thicker layer around the CT compared to the layers in between the tendons (Fig. 3G-I).

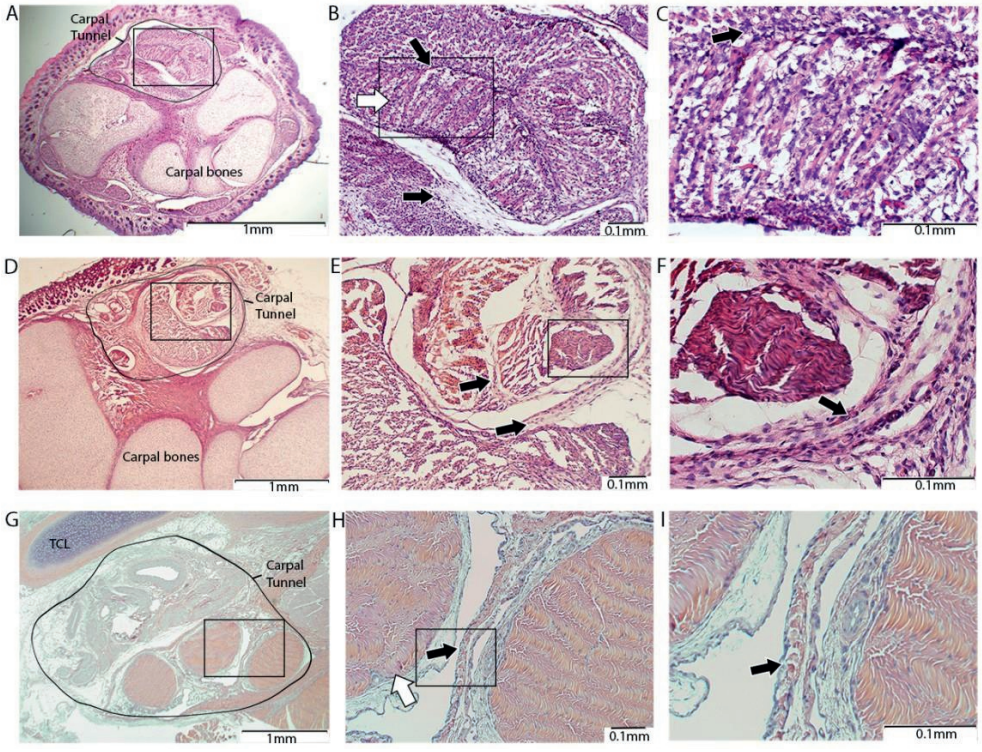


Figure 3: H&E staining of the CT of a rabbit at 27d gestation (top row), 3 days post-partum (middle row), 6 weeks post-partum (lower row). (A-C) The carpal tunnel is recognizable including the deep flexor tendon and the three FDS tendons which. Connective cell layers within and around the tendons are seen. (D-F) The tendons have separated, but are still closely related. The SSCT is better distinguishable and thicker around the circumference of the carpal tunnel compared to in between the tendons. (G-I) The FDS tendons are fully isolated with a connective cell layer of just a few cells wide separating them.

CT: carpal tunnel; FDS: flexor digitorum superficialis; TCL: transverse carpal ligament; SSCT: Subsynovial connective tissue. Rectangles: region of interest for subsequent pictures. Arrows: superficial flexor tendon (white), connective tissue (black). Magnifications of x20, x100 and x200 were used.

Electron Microscopic images of rabbit tissue

Fetal (27 day gestation)

Working chronologically from young to old, the SEM images of fetal CT showed distinguishable oval shapes of the flexor tendons (Fig. 4A), with a characteristic wave-like organization pattern in the longitudinal view. Regions of SSCT matrix were mostly selected around the flexor tendons, assuming that changes in organizational structure would most likely be presented in the areas subjected to the highest shear stress levels. At higher magnification (Fig. 4B), the surrounding connective tissue appeared as a dense, solid mass with sporadic connections to the tendon. These connections appeared as single-stranded thin fibrils with flat surfaces and large footprints on the tendon. No difference in connective tissue thickness or organization was seen at other regions in the carpal tunnel.

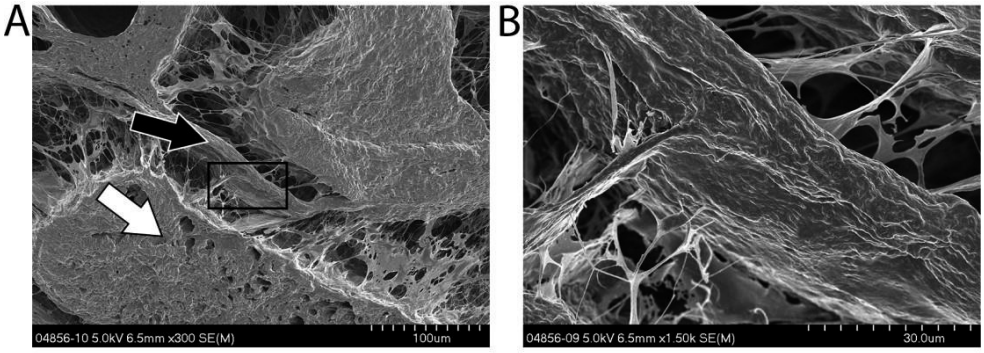


Figure 4: Transverse SEM views of the carpal tunnel structures of a third trimester rabbit kit. (A)The FDP tendon has been formed (white arrow) and has a dense, homogenous matrix on top (black arrow). (B) A higher magnification, broad sheeted fibers are connecting the tendon to the dense matrix.

Neonate (3 days)

SEM images from neonates showed that the flexor tendons have clearly formed and are separated by a thick, dense layer of connective tissue, as was seen in the fetal tissue (Fig. 5A). Fibrils looked similar in terms of morphology and gross count and were again represented as flat connectors, although not present throughout (Fig. 5B). It is noteworthy that rabbits are not ambulating at this age, but will show active motion and gross limb movement.

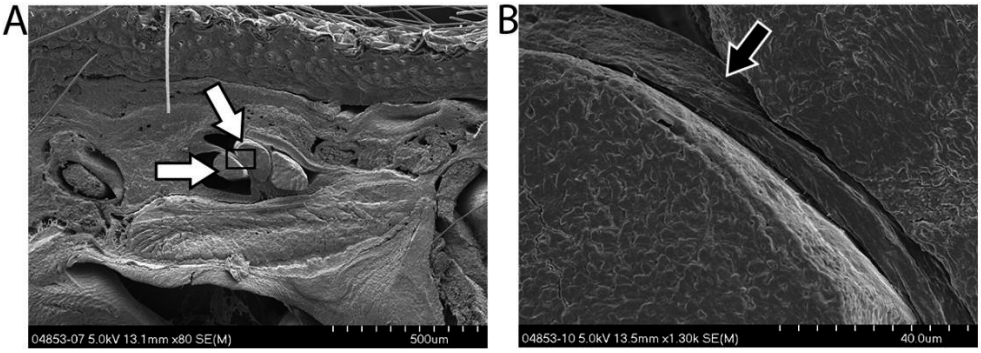


Figure 5: SEM images showing transverse carpal tunnel tissue from a 3 day old rabbit kit. (A) The FDS tendons are discernable (white arrows) and lie volar to the flattened FDP. The palmar side with the skin can be seen at the top. (B) A thin layer of dense tissue separates the flexor tendons (black arrow).

Juvenile & Adult (6wk and adult)

In these samples, a more profound difference in connective tissue morphology was visible. With the layered organization, the connective tissue at the juvenile age (Fig. 6A&B) more closely resembles what has previously been described in the adult (and human) SSCT. Instead of the densely packed homogenously appearing matrix, sheets now have formed, giving the transverse cross-section a more layered and separated appearance. However, the presence of the interconnecting fibrils is less apparent at the juvenile stage (Fig. 6C) than in the adult. At an x8.0k magnification, the transverse cross-sectioned sheets appear to be the product of flattened bundles of connective matrix, not interconnected but still closely stacked. In some of the samples, the sheet formation was not as profound and seemed more chaotic and interwoven (Fig. 7). However, none of the images showed the solidity of the SSCT as seen in the younger specimens. This was true for the SSCT between the FDS tendons as well as around individual tendons. The adult specimens had a more layered appearance with a higher count of sheets and larger spaces separating them (Fig. 8A). The original orientation of the fibrils within the sheets was still visible although the surface appeared smoother (Fig. 8B).

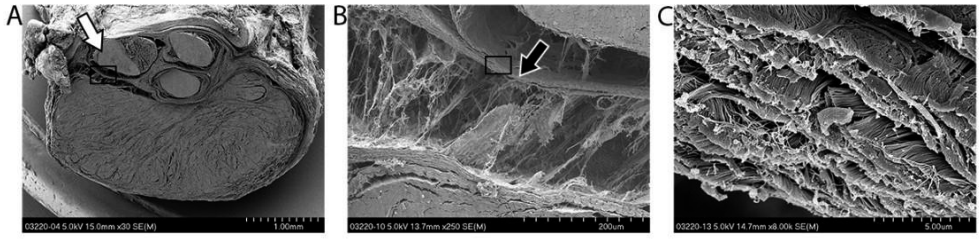


Figure 6: SEM images of a fully dissected carpal tunnel from a juvenile rabbit containing the FDP and three FDS tendons. Sample was cut transversely. (A) At low magnification, the individual FDS tendons (white arrow) are distinguishable with the much larger FDP bundle at the bottom. (B) In between the FDS and FDP, a thin layer of the SSCT is visible (black arrow), with loosely attached string-like fibers attached to the FDP. (C) At higher magnification, the intricate sub-organization of the sheet is visible, with flattened bundles of fibrils oriented parallel to the tendon.

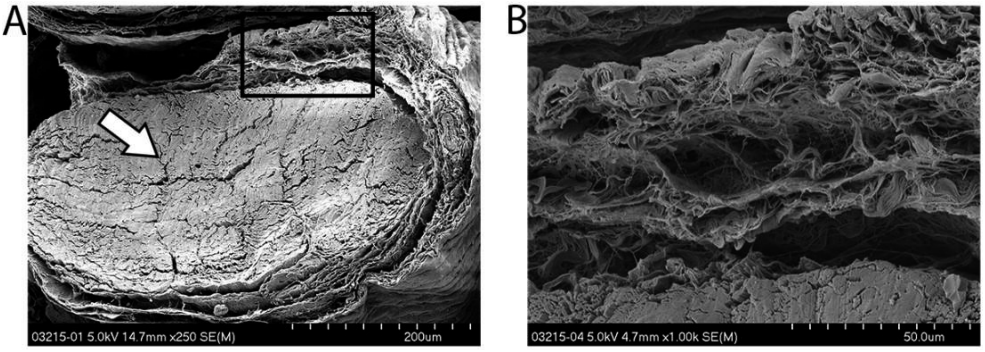


Figure 7: SEM images of a juvenile rabbit with (A) a single FDS tendon (white arrow) and the surrounding SSCT. (B) Initial layering starts superficial to the tendon with a network of crossing bundles and relatively thick sheets, partially merged together.

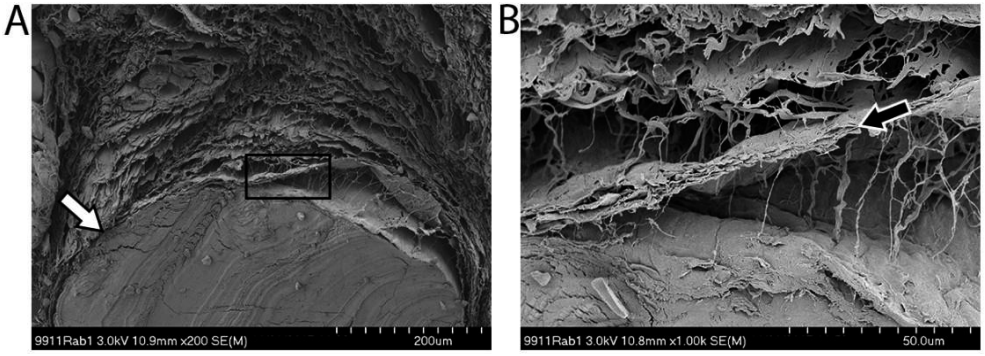


Figure 8: SEM image of adult rabbit carpal tunnel tissue, cut in the transverse plane. (A) The SSCT superficial to the third FDS tendon (white arrow) shows a structural organization similar to humans with multiple horizontal sheets (B) stacked on top of each other (black arrow) and linked through smaller singular fibrils.

Discussion

Between the final fetal stage and adulthood, the connective tissue in a rabbit carpal tunnel undergoes marked morphological changes, from a dense, thick matrix localized peri- and inter-tendinous to a complex micro-organization of interlinked sheets comprised of flattened collagenous fibrils. Assuming that there is a reasonable comparison to make between rabbit and human development[23], looking at the rise and timing of the shaping of the SSCT supports the adaptive development hypothesis described in the introduction, which may lead to important implications regarding the development and possibly even management of carpal tunnel syndrome.

One of the most interesting findings was the transition from a solid structure into the multi-layered one between the fetal and neonatal time points. As in tendon and ligament development, it seems plausible that SSCT development is intimately connected with the development of its anatomical context and the application of mechanical forces[26].

Tendons have been shown to react in cellular response to mechanical forces and increases in shear stress, adjusting the type of extracellular matrix deposited[17]. Animals that need independent locomotor movement quickly after birth show larger tendon fibril diameters than those who can remain inactive after birth[27]. Although our study lacks time points between 3 days and six weeks, it is not until after the first week that rabbits will start exhibiting controlled active movement and will start bearing their weight on the front paws[28], so it seems fitting that it was not until the latter time point that we found marked differences in SSCT structural organization. This is supported by an earlier study that looked at rabbit superficial flexor tendon and synovial sheath development, which noted that it was not until after the first month that larger tendon fibrils with more interconnections were formed[29]. The authors also described that the synovial membrane surrounding the tendon showed less mature fibroblasts compared to the tendon proper up to 112 days after birth. Although this sampling was done at a level more proximal than the CT and only reviewed tissue on microscopic level, this indicates that the connective tissue lags in development and, as the authors suggest, could be due to a response to direct loading of the tendon whereas the sheath is subjected to less and indirect loads[29,30]. Additionally shown was the tendon decreasing in cellularity to 5% of the original count within the first 4 months of life, which was reflected in our histological observations as well.

In order to link our anatomical findings to a functional purpose, we hypothesize that nutrition transportation and mechanical stress inhibition play prominent roles. First, it is important to note that flexor tendons in the carpal tunnel are unique in that they do not conform to the conventional categories of being either intra-, of extrasynovial. The loose paratenon (or epitendineum) commonly found lining the outer layer of a tendon provides nutrition for extrasynovial tendons[31], but this layer is incorporated in the SSCT at the level of the carpal tunnel. This combination of a synovial lubrication layer and the viscoelastic character of the SSCT allows for enhanced movement with less friction[32]. Guimberteau et al., (2010) describe the SSCT as a microvacuolar network containing blood and lymphatic vessels, showing highly variable polyhedral shapes when dissected. Chemical analysis of the SSCT showed a composition of predominantly proteoglycans (70%) and collagen type I and III (23%) which fits with the relatively high water content and the mucinous-like nature noticeable *a vue*. Secondly, if the main function of the connective tissue surrounding the flexor tendons can be summarized as (1) modulation of the transmission of stress (either in the form of shear or compressive) from the tendon to the surrounding tissue and (2) accommodating a vasculature network, then it would make sense that the SSCT structure is a combination of lubricant holding (i.e., the proteoglycans) and sliding (the SSCT layers) elements. The smaller interconnecting fibrils restrict movement, reducing the risk of damaging the nutritional sources[4]. When expanded, the SSCT provides an increase in maximal excursion by recruiting different layers, but is restricted when the edges reach the physical limitations of the preceding layer. This is comparable to extending a telescope. A multilayered system allows both the potential to capture fluid in between the layers and dampen stress with tendon

loading[16,33]. Cadaveric and clinical data has shown that the SSCT moves less compared to the tendon during finger flexion and extension and. Their ratio depends on tendon velocity, total excursion and acquisition method[34,35], with a stress-relaxation pattern suggesting visco-elastic behavior[6]. This would fit the above mentioned mechanism of shearing layers with incremental recruitment. Placing this in the context of CTS, it has already been shown that there is thickening and tearing of these fibrils in patients with CTS[9,36] which lead to the hypothesis that SSCT damage could play a significant role in CTS pathophysiology[37].

Assuming that one of its primary functions is to create a dynamically accommodating environment, it makes sense to look at a similar example, e.g. the muscle-fascia interface; in humans, fetal movement commences as early as seven weeks of gestation with individual limb motion around week ten[38]. A study on myofascial development in older human fetal tissue (25-33 weeks) shows a cyclic process of fascia depositions, originating from the skeletal muscles themselves, then thickening, and subsequently detaching. The repetition of the process produced multilayered fascia[39]. Hypothetically, a similar process could be present in the CT, where tendinous fibroblasts create layers of collagen matrix deposits in areas where friction is present (closest to the tendon), which would explain why the SSCT in the juvenile rabbits contained fewer layers and interconnecting fibrils compared to the adult rabbits.

A limitation of our approach is the absence of time points between 3 days and 6 weeks and the lack of biomechanical testing data. In humans, the SSCT does not exceed 1mm in width and has proven to be very difficult to reliably test biomechanically, especially in vivo. Based on our experience in this field, a preliminary observational study was deemed more feasible and so we wanted to limit the animal sample size. Most of our specimens were provided through other research studies, and the fetal and juvenile time points were chosen based on expected tendon development as reported by others. Nonetheless, these limitations prevent us from drawing any conclusions on causality and on how the SSCT is formed on a cellular level. As we wanted to appreciate not solely SSCT development, but also the relationship between the different carpal tunnel structures during development and gain some insights into levels of cellularity, we choose to visualize the specimens in multiple ways. Unfortunately, due to limited tissue size and rigidity, we were not able to acquire the macroscopic images of the fetal and neonatal groups. Additionally, a few side notes need to be addressed concerning the limitations of our methodological approach. For the staining and SEM acquisitions, tissue was prepared through dehydration and rinse cycles. We cannot exclude the possibility that the dehydration might have affected some of the structural integrity of the specimens. Additionally, for both approaches samples needed to be cut, which in some samples resulted in minor dissociation of tendinous structures from surrounding connective tissue. SSCT tissue was mostly spared, despite this being quite delicate, probably because shear stress was limited through the addition of anchoring sutures on the edges of each sample. Finally, because of the selected method of specimen collection and the

limited amount of tissue available per specimen, we were not able to fully correlate tissue characteristics between different visualization methods within one animal. Future research can build on our findings and further explore the developmental potential of a rabbit carpal tunnel model while answering clinically important questions. A follow-up research question would be to test the effect of short-term casting on SSCT morphology and strength, similar to what we do in conservative CTS treatment.

To summarize, the SSCT in the carpal tunnel is a unique structure with an important role in load transmission, which may be relevant in CTS pathology. Using an animal model, we have shown that, as in other musculoskeletal structures, the SSCT does not fully develop during the fetal phase but is instead more likely a functional adaptation to (post-natal) activity. We propose that the structural layout of the SSCT supports minimization of load-induced mechanical stress while facilitating nutritional flow, but further biomechanical studies are needed to confirm this hypothesis. We hope that a better understanding of the SSCT multi-level organization will be useful to those who study the development of CTS, as well as those interested in CTS treatment.

Conflict of interest

All the authors declare no conflicts of interest with the work presented here.

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PART II

4

CHAPTER 4.

Reliability of ultrasound speckle tracking with singular value decomposition for quantifying displacement in the carpal tunnel

Schrier, V. J.M.M., Evers, S., Bosch, J.G., Selles, R.W., & Amadio, P.C. (2019).

Journal of Biomechanics, 85, 141-147.

Abstract

Introduction
Inhibited movement patterns of carpal tunnel structures have been found in carpal tunnel syndrome (CTS) patients. Motion analysis on ultrasound images allows us to non-invasively study the (relative) movement of carpal tunnel structures and recently a speckle tracking method using singular value decomposition (SVD) has been proposed to optimize this tracking. This study aims to assess the reliability of longitudinal speckle tracking with SVD in both healthy volunteers and patients with CTS.

Methods
Images from sixteen healthy volunteers and twenty-two CTS patients were used. Ultrasound clips of the third superficial flexor tendon and surrounding subsynovial connective tissue (SSCT) were acquired during finger flexion-extension. A custom made tracking algorithm was used for the analysis. Intra-class correlation coefficients (ICCs) were calculated using a single measure, two-way random model with absolute agreement and Bland-Altman plots were added for graphical representation.

Results
ICC values varied between 0.73-0.95 in the control group and 0.66-0.98 in the CTS patients, with the majority of the results classified as good to excellent. Tendon tracking showed higher reliability values compared to the SSCT, but values between the control and CTS groups were comparable.

Conclusion
Speckle tracking with SVD can reliably be used to analyze longitudinal movement of anatomical structures with different sizes and compositions within the context of the carpal tunnel in both a healthy as well as a pathological state. Based on these results, this technique also holds relevant potential for areas where ultrasound based dynamic imaging requires quantification of motion.

Introduction

Carpal tunnel syndrome (CTS) is the most common compression neuropathy, with an estimated prevalence of 1-5%[1,2]. CTS is predominantly a clinical diagnosis, often supported by electrophysiological measurements. However, this is an invasive, uncomfortable method that has been criticized since it has limited negative predictive potential[3], with up to 50% of electrophysiological-negative patients still benefitting from treatment[4]. More recently, ultrasound (US) imaging has emerged as an interesting alternative with sensitivity and specificity rates almost matching electrophysiological testing[5].

Transverse and static ultrasound parameters have been studied most extensively, with median nerve area showing the highest sensitivity rates[6,7]. However, longitudinal and dynamic assessment of the carpal tunnel structures has also gained interest. A common finding in CTS patients is non-inflammatory thickening and fibrosis of the connective tissue around the median nerve and tendon flexors[8]. The fibrotic changes in this subsynovial connective tissue (SSCT) alter the mechanical response of the tissue surrounding the median nerve to loading of the flexor tendons. Previous research has focused on measuring these patterns of (relative) motion[9-11]. Compared to non-CTS volunteers, relative median nerve motion appears inhibited, worsening with more severe symptoms[12]. Since the decision for conservative or surgical treatment is influenced by disease severity, being able to measure this non-invasively could thus aid the clinical diagnosis process and support intervention choice. However, measuring the SSCT has been challenging due to its small size.

Speckle tracking is an image analysis technique that has been applied mostly to cardiac imaging[13] but is now also under investigation in the musculoskeletal field due to its ability to describe features of moving structures[14]. Doppler imaging is similar to this technique, but is limited by its angle dependency. This method tracks the displacement of speckle patterns, the grainy texture in the ultrasound image that results from interfering ultrasound waves that are backscattered by the inhomogeneity of the tissue. Speckle tracking of tendons has been described for the Achilles tendon[15-20], the tibialis anterior[21], the patellar[22], the flexor digitorum superficialis (FDS)[17,23,24] but also for the median nerve[12,25].

Relative SSCT motion has been assessed using commercial tracking software in both healthy volunteers[26] as well as in patients with CTS[11], but this type of tracking is limited because the software limits the settings that the user can change manually in order to optimize the tracking on individual patient basis.

Recently, a custom made speckle tracking algorithm was extended with a background suppression technique based on Singular Value Decomposition (SVD)[27] in order to minimize the effect of clutter and noise of stationary background. This improved

approach could provide a cleaner look at the differential movement of the FDS and how the relationship between the SSCT and neighboring structures changes in CTS. Since ultrasound imaging and speckle tracking are both subjected to operator interpretation, variability in image acquisition and analysis needs to be assessed. If reliability can be established, it also provides interesting potential applications in the image processing of any musculoskeletal assessment where dynamics and biomechanics play a role. Therefore, using the context of the carpal tunnel structures, this study evaluates three aspects of reliability of speckle tracking with SVD: 1) intra-rater, reflecting the variation in measurements done by a single rater, 2) inter-rater analysis, reflecting the variation between two raters who measure the same subjects[28], and 3) repeatability (also referred to as test-retest reliability), reflecting the variation in measurements acquired at multiple time points.

Materials and Methods

Data collection

Ultrasound images were obtained from a sample of patients with CTS and from volunteers without CTS, referred to as control group. The Mayo Clinic Institutional Review Board approved both studies (control group IRB#06-002950, patients with CTS IRB#14-003444). Written consent was obtained from all participants.

Control Group
Seventeen subjects between the ages of 18-85 years were included. Exclusion criteria were: history of CTS, rheumatoid arthritis, osteoarthritis or traumatic injuries of the ipsilateral hand or wrist. All imaging was done according to a preset imaging protocol described below.

Patients with CTS
A dataset was constructed by randomly selecting twenty-two patients included in a prospective randomized controlled trial (ClinicalTrials.gov, identifier: NCT02219555). Patients were recruited after being diagnosed with CTS in a hand clinic by any of the hand physicians. Diagnoses were made based on clinical presentation and EMG results as described in the guideline from the American Academy of Orthopaedic Surgeons[29]. Inclusion criteria were clinical diagnosis of CTS, age between 21-80 years, symptoms of numbness or tingling for at least 4 weeks, and indication for treatment with injection or surgical release. Exclusion criteria were a previous surgical release, tumor, deformity in hand/wrist, previous history of steroid injection, and any known risk factor for non-idiopathic CTS (including pregnancy, diabetes, rheumatoid arthritis). Clinical evaluations included two point discrimination, Phalen's test, Tinel's sign, manual muscle testing of the abductor pollicis brevis, and notation of the presence of thenar muscle atrophy.

Imaging protocol

Ultrasound recordings were collected from the patients prior to their treatment. Each subject was imaged in supine position on a bed with the elbow (of the affected hand in CTS cases) fully extended, the shoulder in abduction (70-80 degrees) and the forearm supinated stretched out on an acrylic glass board. One strap was used to minimize forearm movement and another to inhibit overextension of the third digit and flexion of the second and fourth digit. An ultrasound scanner Philips iE33 (Royal Philips Electronics, Amsterdam, the Netherlands) equipped with 15L7 linear array transducer was used. The transducer was placed at the wrist in a sagittal plane over the proximal wrist crease with the wrist in the neutral position. If necessary, a folded pillowcase was placed under the hand to straighten the wrist. The transducer was applied to the skin without additional pressure and with plenty of gel. Participants were asked to flex and extend the third digit corresponding to a frequency of fifty beats per minute under guidance of a metronome (Fig. 1, Suppl video 1). After a practice round, three ultrasound clips with each three flexion-extension cycles were recorded. All images were taken following the same ultrasound protocol, with the same machine by two different ultrasonographers (VS & SE) who were both trained in using the protocol.



Figure 1: Example of start (left) and end (right) position of the third digit during the flexion-extension cycle. Each recording contained three cycles.

Data analysis

After image acquisition, all images were analyzed using a Matlab based custom made algorithm developed at the Erasmus MC for speckle tracking with singular value decomposition which had already been tested in an animal model and validated for tendon tracking[23,30]. Before setting the region of interest (ROI), the complete clip was reviewed to identify the median nerve, the SSCT, the FDS and the flexor digitorum profundus (FDP). Then, image analysis took place in three stages (Fig. 2). First, the ROI was manually placed with its proximal border at the level of the radial head, covering the width of the tendon, but without incorporating movement of the adjacent FDP. The ROI was fixed during motion with a pre-set 1:4 size ratio, and could be rotated to ensure a position parallel to the tendon fibrils. Immediately after placement, a feedback video

would play with the ROI added for the analyst to review whether it correctly captured the desired structure throughout the entire clip (Fig. 3). In some cases, the tendon would show apparent movement in the volar-dorsal plane in which case, if possible, the ROI would be adjusted in size. Differences in ROI box sizes were negligible, since tendon widths were comparable. In cases where additional kernel size and number fine tuning were deemed necessary, images were excluded. For the SSCT, a similar method was used, but with a separate ROI covering the visible SSCT. The position of the ROI was placed directly volar to the tendon ROI to measure relative motion. The SSCT's organization shows multiple horizontal sheets[8] and to account for different speeds at different levels, the analysis was performed over five evenly-spaced vertical layers within the SSCT ROI. After placing both ROIs, a temporary video file was created that was used as input for the speckle tracking. The algorithm has been described in detail before[30], but in short, consists of first the SVD filtering followed by the speckle tracking. The SVD filter is an improvement on the more classically used high-pass clutter filter; It decomposes the sequence into specific motion components, which allows noise (incoherent-high frequency signals) as well as clutter removal (high intensity- low frequency), minimizing the signal of static and slow moving structures in the image sequence. Then, within the ROI, a set of overlapping 2D kernels was defined and block matching with normalized cross correlation was applied for each of them to find the frame-to-frame displacement vector. Finally, as described by Bandaru et al.[30], unreliable kernel results were removed based on their correlation values and discordant vectors. The average of the displacement vectors of the reliable kernels gave the final frame-to-frame displacement vector.

The analyses were done using Matlab (R2016a, The MathWorks Inc., Natick, MA, 2000). The Euclidean length of the total displacement vector was calculated in mm. The input used for reliability analyses were the excursion and shear index over 3 cycles (equal to six movements). The shear index is a measure for relative motion between the third FDS and SSCT and was defined as:

$$Shear\ index = \frac{Tendon\ Excursion - SSCT\ excursion}{Tendon\ Excursion} * 100\%$$

An index value of 0% indicates that the SSCT moved in equal amount with the tendon whereas 100% would indicate a complete dissociation. All analyses were done in random order and the rater who performed the analyses was blinded to the results of their previous assessment and the results of the other rater.

Three aspects of reliability were analyzed:
Intra-rater reliability: For measurement of the main rater-dependable factor of the speckle tracking (placement of region of interest), the recordings from both groups were analyzed in random order twice by the same rater with a time interval of three-four weeks. The first set of results was also used for the inter-rater reliability and the repeatability. *Inter-rater reliability:* In order to measure the variance between different raters, a second analyst measured the same set of clips derived from both the control group and the CTS patients. *Repeatability:* Repeatability was defined as the variation in

measurements derived from the same subject, under equal circumstances, directly after an initial clip was recorded. Any differences measured therefor must be the summation of the variance in the recording and the analysis.

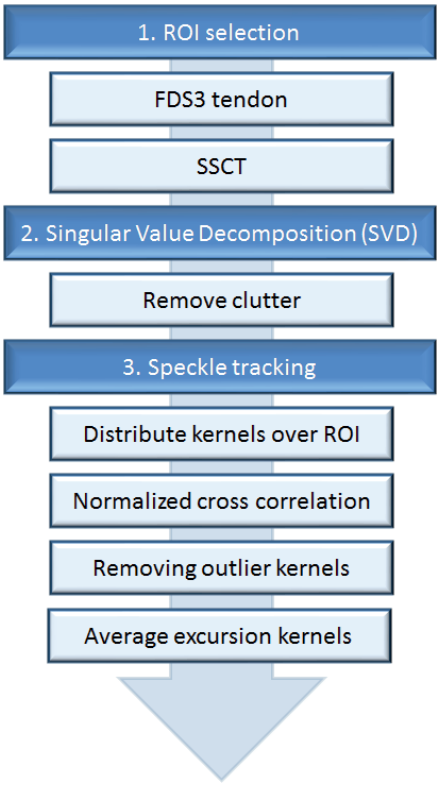


Figure 2: General overview of the image analysis sequence including the singular value decomposition.

Statistical Analyses

To quantify reliability, intra-class correlation coefficients (ICCs) including the 95% confidence interval were calculated, using a two way random effects model with single measure and absolute agreement. In general, ICC values above 0.75 are considered as excellent, values between 0.40–0.74 are fair to good and values below 0.40 are considered as poor, in accordance with the classification proposed by Fleiss[31]. Agreement was evaluated using Bland Altman plots with 95% limits of agreement[32]. Distribution of the difference of means was tested for normality visually with histograms and normal quantile plots and statistically with the Shapiro-Wilk test of normality[33]. In case of normality, the plots were made with limits of agreement as calculated by the mean difference ± 1.96 x SD of the difference. In case of non-normal distribution, a logarithmic transformation of the data was performed after which the same calculation for the limits was done[34]. Statistics were done using IBM Statistical Package for Social Sciences software version 22 (SPSS, Chicago, IL, USA).

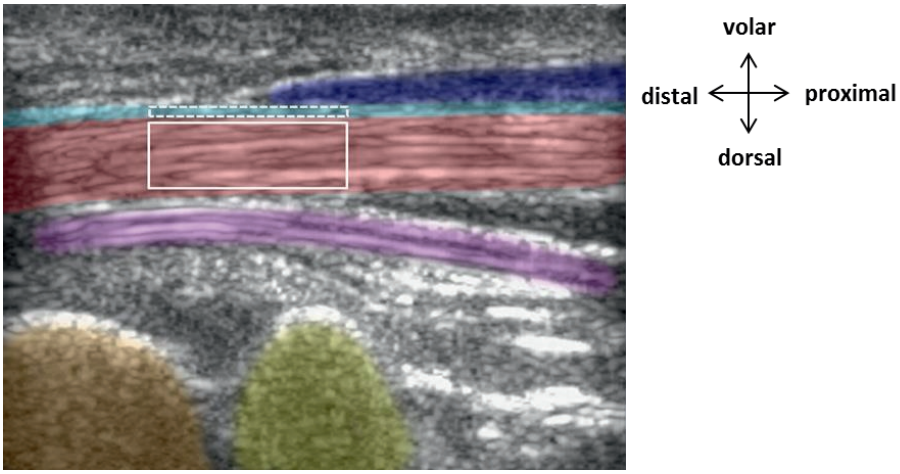


Figure 3: Ultrasound B-mode image with color overlay to indicate the anatomical structures in a sagittal plane. From volar to dorsal, dark blue: median nerve, light blue: SSCT between tendon and median nerve, red: FDS3, purple: FDP 3, orange: lunate, yellow: radius. The box with the solid demarcation depicts an example of a ROI over the entire width of the superficial tendon. The box with the interrupted line shows the ROI for the SSCT.

Table 1: Summary of absolute displacement results and shear indices in both groups. Means were calculated based on measurements over three consecutive flexion-extension cycles derived from the first data set from one of the raters. FDS: Flexor digitorum superficialis, SSCT: Subsynovial connective tissue, Shear index: ratio of the difference in motion between FDS and SSCT over the total motion of the FDS.

	Control group	CTS patients
Number of participants	16	22
FDS displacement in cm; Mean (SD)	6.8 (2.4)	8.4 (2.5)
SSCT displacement in cm; Mean (SD)	1.5 (0.7)	2.1 (0.8)
Shear index in %; Mean (SD)	78 (9.7)	73 (13.2)

Table 2: Reliability of structure displacement measurements including relative motion (shear index) for both the control and the CTS group. ICC's were calculated using two way random model with absolute agreement and include the 95% confidence intervals. Control group n=16, CTS patient group n=22. FDS: Flexor digitorum superficialis, SSCT: Subsynovial connective tissue, CI: Confidence interval, Shear index: ratio of the difference in motion between FDS and SSCT over the total motion of the FDS.

	Control group						CTS patients					
	FDS		**SSCT**		**Shear index**		**FDS**		**SSCT**		**Shear index**	
	ICC	95% CI	**ICC**	95% CI	**ICC**	95% CI	**ICC**	95% CI	**ICC**	95% CI	**ICC**	95% CI
Intra-rater	**0.94**	0.84-0.98	**0.93**	0.82-0.98	**0.87**	0.66-0.95	**0.98**	0.95-0.99	**0.81**	0.59-0.92	**0.82**	0.61-0.92
Inter-rater	**0.95**	0.86-0.98	**0.82**	0.56-0.93	**0.74**	0.41-0.90	**0.82**	0.59-0.92	**0.70**	0.38-0.87	**0.66**	0.32-0.85
Repeatability	**0.89**	0.72-0.96	**0.82**	0.55-0.93	**0.73**	0.40-0.90	**0.88**	0.74-0.95	**0.74**	0.47-0.88	**0.82**	0.61-0.92

Results

In total, images from seventeen healthy volunteers and twenty-two CTS patients were analyzed. The clips of one control participant were labeled as too low image quality to include since the tendon and SSCT moved significantly in volar-dorsal direction and out of plane, causing the structures of interest to fall out of the ROI. After exclusion, the remaining sixteen healthy volunteer clips were used for analyses.

The absolute values for the tendon, SSCT and shear index are summarized in Table 1. The shear indices are arguably the parameter of most interest since it is the disturbance of the relative motion due to fibrosis of the SSCT that would underlie the hypothesized pathophysiology of idiopathic CTS. The shear index values found in the control versus the CTS group ranged between 56%-90% with an average of 78% versus 41%-93% with an average of 73% respectively.

The intra-rater, inter-rater and repeatability ICCs of the tendon and SSCT are shown in Table 2. All tendon ICC values can be classified as excellent, as well as the majority of the SSCT comparisons. Only SSCT inter-rater and repeatability in the CTS patients classified as good with 0.70 and 0.74 respectively. In the control group, values for the shear indices ranged between good and excellent (intra-rater: 0.87, inter-rater: 0.74, repeatability: 0.73). This was also found in the CTS group except for a lower ICC value of 0.66 for the inter-rater reliability. Bland Altman plots for the intra-, inter-rater and repeatability reliabilities of the shear index are shown in Figure 4A-F. By plotting the average between two measurements against the difference between those measurements, any funnel-like shapes would indicate a more profound disagreement with either a decrease or increase in the average value. Only in the inter-rater comparisons and the repeatability of the CTS group (Fig. 4C, D & F) can a slight tendency of more disagreement be found with lower values. However, most values still fall within the 95% confidence limits.

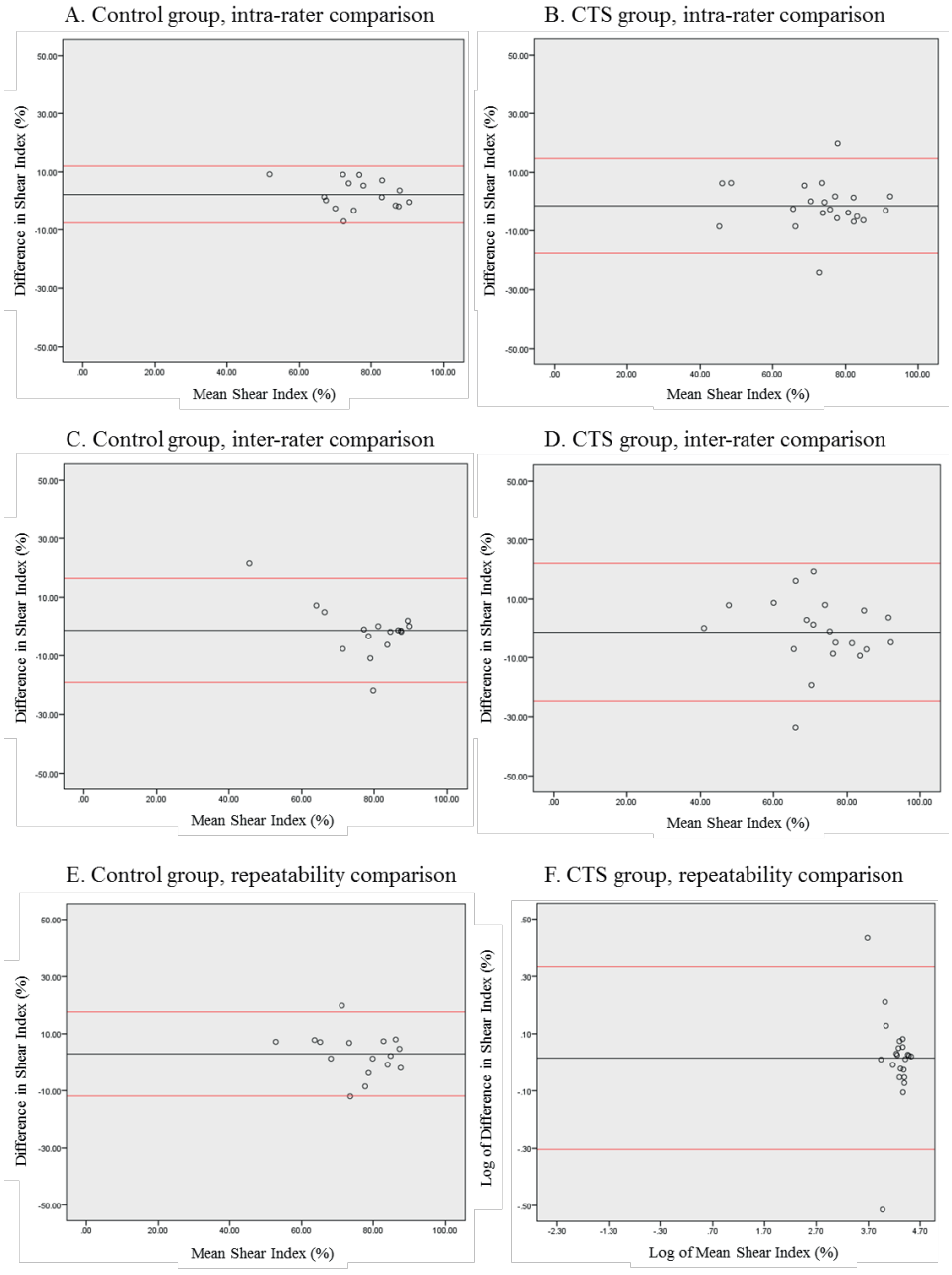


Figure 4A-F: Bland Altman plots showing the intra- (A and B), inter-rater (C and D) and the repeatability (E and F) data of the shear index for both tested groups. These plots have the mean of two measurements on the x-axis plotted against the difference between the two values. The black line and red lines indicate the average difference and the 95% upper and lower limit respectively for the compared samples. Data for figure F was logarithmically transformed due to the non-normal distribution of the data.

Discussion

This study assessed the reliability of a speckle tracking algorithm with singular value decomposition to track absolute and relative motion of longitudinal structures inside the carpal tunnel. Based on our sample of both CTS and non-CTS subjects, we found mostly good to excellent reliability. The lowest ICC value, for the inter-rater comparison of the shear index, can still be classified as moderate to good.
Although several publications describe speckle tracking for analysis of carpal tunnel structures, not many have described reliability. Filius et al. have used a similar speckle tracking analysis without SVD and published test-retest values for both the third FDS tendon and its superficial layer of SSCT. They note ICC values of 0.70 and 0.73 respectively[12]. Their repeatability measurements were done in a larger group (n=50) and reliability data was from both healthy and CTS patents combined. Using Doppler imaging, FDP excursion reproducibility was assessed in both a healthy group[35] as well as a patient group with tendon injury[36]. They presented ICC values of 0.81 and 0.88 respectively at ten days post intervention. Despite the difference in imaging depth, their results are similar to ours.

In general, our SSCT ICC values tended to be lower than those for the tendon which was also the case in similar research[12]. This can in part be explained by the anatomical differences; The FDS tendon has a width of about ten times the SSCT (<1mm) and thus can accommodate a larger ROI which is less susceptible to movement in the volar-dorsal plane. In addition, the SSCT is a layered structure, so during tracking the displacement can differ based on the position. This should receive extra consideration when imaging patients with CTS, because higher levels of fibrosis have been found to occur in the layers closest to the tendon[37].

In this study, an average of 1.1 cm (control group) and 1.4 cm (CTS group) excursion of the tendon was found per flexion / extension movement (data shown in the results is total over three cycles with two movements each), which is comparable to what colleagues have found; Korstanje et al. used a similar algorithm, without SVD, on CTS patient-derived clips and found a mean tendon excursion of 1.98 cm but do not report any ICC values[10]. Filius et al. published two studies with CTS patients[9,12] where they used the same algorithm without SVD, and found average tendon excursions of 1.88 cm and 1.44 cm (calculated based on published values). Control data showed 1.56 cm and 1.43 cm FDS3 excursion. To test whether the difference in tendon excursion in the present study could be due to anthropometric differences between the groups, post hoc hand measurements from the distal wrist crease to the tip of the long finger were made. This indeed showed a small but significant size difference (CTS: 20.3±1.8 cm, controls: 18.7±1.0 cm). However, the quality of the CTS patient pictures was suboptimal, leading to less accurate measurements. Perhaps more importantly, we do not have data on the individual amount of finger flexion during the trials. Someone with a large hand could still have a small tendon excursion if their maximum finger flexion was smaller. Although there is evidence that hand morphology is associated with CTS, it seems to be the ratio

between wrist width and depth that differs most between controls and CTS patients[38]. So far, this seems to hold limited diagnostic and prognostic value[39].

Excursion values for SSCT were published in two articles, 1.55 cm and 0.74 cm for CTS groups and 1.30 cm and 0.76 cm in control groups[10,12]. Although Korstanje found a significant difference in SSCT excursion between the most and least affected hand (p=0.025), this was not found in the study by Filius et al. and explained by possible greater inter-subject variability. Our study found an average of ~0.3 cm SSCT excursion in both the patients with CTS as the healthy group, but all results are susceptible to noticeable standard deviations. Additionally, our study was not primarily designed for absolute value comparison and our absolute values represent the total excursion over three cycles. Previously, a validation of the speckle tracking[30] was done for tendons and ideally the same would be done for the SSCT. However, this is challenging, even with a phantom or cadaver model, due to the small size of the SSCT and the likelihood of disrupting the complex microstructure with markers.

Strengths of this study are that we tested an innovative technique to measure ultrasonically captured motion of small anatomical structures, expanding the boundaries of what speckle tracking can detect and be utilized for. Insights gained into the limitations of speckle tracking can help contextualize results whether acquired for research or clinical purposes. In addition, utilizing dynamic features and interrelations of the carpal tunnel structures to support CTS therapy choice is a novel approach. Limitations of the study include that speckle tracking inherently does not take out of plane motion into account, which may result in underestimation of the actual motion. We also used a high frequency probe to allow visualization of the SSCT, which might have supported the high ICC values. Additionally, it was reported that the tracking depends on the balance between tendon velocity and acquired frame rate, with a decrease in validity if these parameters fall outside of predetermined boundaries[30]. In our situation this was, prevented by adding a metronome during acquisition, but this does not eliminate the variation in the participant's cooperation and ability to perform a repeatable movement. If, in a clinical application baseline images are compared to a follow-up, changes in finger movement should be taken into account via either a rigid acquisition protocol or an outcome measure insensitive to total excursion (like shear index). Additionally, the ICC values presented in this study do indicate that future data on shear index in the carpal tunnel should be re-evaluated within different contexts. For example, for a study-based purpose, a single rater would be fine, but for clinical application, more research including multiple raters would be needed.

In conclusion, this study shows that SVD enhanced speckle tracking can reliably be used to analyze (relative) longitudinal SSCT displacement both in participants with and without CTS. Together with the validation data, our results imply that dynamic ultrasonic measurements of carpal tunnel structures can be used to further explore the potential of using this technique to help guide and predict CTS treatment outcomes. Although our study focuses on CTS related anatomical structures, the principles of the

image analysis could be extrapolated to other research areas involving pathologies where the assessment of dynamic imaging is of relevance.

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Conflict of interest statement

None of the authors have any commercial associations that might pose or create a conflict of interest with information presented in the submitted manuscript. None of the authors received payments or other benefits or a commitment or agreement to provide such benefits from a commercial entity. Data presented in this manuscript has been presented on poster at the Orthopedic Research Society conference, New Orleans, Louisiana in 2018.

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CHAPTER 5.

Relative motion of the connective tissue in carpal tunnel syndrome: the relation with disease severity and clinical outcome

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Abstract

Excursion of the median nerve and the surrounding subsynovial connective tissue (SSCT) is diminished in patients with carpal tunnel syndrome (CTS). This study sought to determine if SSCT excursion could be utilized to predict surgical outcome. Idiopathic CTS patients were reviewed with ultrasound and electrodiagnostic tests at baseline. A speckle tracking algorithm was used to determine SSCT relative to tendon motion (shear index). Analyses of variance tests were used to compare SSCT motion with disease severity at baseline. Adjusted linear regressions were used to test the association with patient-reported outcome. A total of 90 CTS patients were analyzed and showed an average shear index of 79% (95%CI: 76.3-81.6%). SSCT motion was lower in CTS patients with increasing electrophysiological severity ($p=0.0475$). There was no significant association of preoperative SSCT motion with symptomatic improvement ($p=0.268$). Overall, SSCT motion is decreased in CTS patients, but shows limited correlation with clinical severity.

Introduction

Carpal tunnel syndrome (CTS) is a compression neuropathy of the median nerve at the level of the carpal tunnel. The diagnosis is most commonly made based on clinical presentation supported by functional evaluation of the nerve with electrodiagnostic studies. Recently, ultrasound (US) has gained a more profound role in the CTS work-up due to its painless nature, cost-effectiveness[1], and comparable sensitivity and specificity[2-5] when using increased nerve cross-sectional area as the parameter of interest[6]. However, these changes in nerve morphology are most likely the physical manifestation of a longstanding process of nerve compression, the initiating factors of which are not well understood.

In clinical practice, disease severity is an important factor in the decision to initiate conservative treatment or to proceed to surgical intervention. Having a non-invasive method of both determining severity and predicting the likelihood of success of operative or non-operative treatment would have additional benefit for the individual patient. Although surgery is highly effective, there is a wide variation in reported success rates[7], suggesting again that better case selection might improve treatment outcomes. Electrodiagnostic studies are the gold standard, but these tests are invasive and their usage has been a topic of debate[8,9].

Besides the well-described increase in median nerve size, a common feature in CTS patients is non-inflammatory fibrotic thickening of the subsynovial connective tissue (SSCT)[10-12]. The SSCT lies below the visceral layers of the bursae in the carpal tunnel and consists of multiple layers of interconnected collagenous fibers, including blood and lymphatic vessels, which directly surround the flexor tendons and the nerve. This structure is a unique feature seen only in the carpal tunnel[13,14]. As the fingers flex and extend, the SSCT layers are gradually recruited, transmitting and absorbing the mechanical stress[15,16], and providing a dynamic framework to protect the supply of nutrients and the nerve from excessive strain[17]. Normally, this results in a situation where the SSCT moves in coordination with, but at a lower velocity than, the underlying flexor tendon[18-20]. Typically, the SSCT is less than 1 mm thick, but in patients with CTS, it thickens and the connections between the layers are disrupted[10] causing changes in the layered gliding system[21]. Peripheral nerves have a physiological reserve when it comes to longitudinal stretching, but with increased perineural fibrosis[21] this may be diminished, and sections of the median nerve could be prone to increases in local strain. With as little as 6% strain already shown to affect nerve function[22], alterations in mechanical SSCT response are a relevant topic of investigation and may help explain the genesis of nerve compression and ischemia in patients with CTS[11,23].

SSCT motion has been measured by Doppler ultrasound as well as by both commercial and custom speckle tracking algorithms[18-21,24-27], showing the feasibility of these methods as well as increased SSCT/tendon shear strain in patients with CTS. However, these were

either done using cadaveric specimens[18,19] or focused solely on the diagnostic potential of relative SSCT motion[20,21,24,25].

Our group has recently validated and tested the reliability of an extension of a previously reported speckle tracking algorithm[28] optimized to track small, fast-moving tissue such as the SSCT[29]. The present study describes the application of this algorithm to a large prospectively-gathered dataset of US images in order to evaluate flexor tendon and SSCT motion in CTS patients undergoing carpal tunnel release. The two main aims of this study were to 1) test whether there is a difference in relative SSCT motion between healthy participants and CTS patients of different severities and to 2) determine whether these preoperative measures were associated with clinical outcome after carpal tunnel release.

Materials and Methods

General

This is a prospective clinical (level I) study utilizing the surgical arm of a larger research trial (CT.gov identifier NCT02219555) which was reviewed and approved by our Institutional Review Board (S-1). Written informed consent was obtained from all participants in accordance with the Declaration of Helsinki[30].

Subjects and disease characteristics

The control group included seventeen subjects between the ages of 18-85 years. Exclusion was based on self-reported absence of history of CTS, rheumatoid arthritis, osteoarthritis or traumatic injuries of the ipsilateral hand or wrist. All imaging was done according to a preset imaging protocol described below.

CTS patients were recruited at our institution at a specialized hand clinic. CTS diagnoses were made by orthopedic or plastic surgeons based on a combination of symptoms at presentation, clinical tests, and nerve conduction studies with/without needle electromyography, referred to as electrodiagnostic studies (EDS). Inclusion criteria were age between 21-80 years, symptoms of numbness or tingling for at least four weeks in at least two digits in one hand including the thumb, index, long or radial border of ring finger, ability to understand spoken and written English, and a decision for surgical release. Exclusion criteria were a previous carpal tunnel release in that hand, tumor, space occupying lesion or deformity in the hand/wrist, pregnancy-induced CTS, and any of these diagnoses: cervical radiculopathy, peripheral nerve disease, thyroid disease, rheumatoid arthritis or other inflammatory arthritis, osteoarthritis in the wrist, diabetes, renal failure, sarcoidosis, amyloidosis or major trauma to the ipsilateral hand or wrist. Clinical evaluations included sensibility (two-point discrimination) in the median nerve distribution of the hand, Phalen's test, Tinel's sign, pinch and grip strength, and assessment of thenar muscle atrophy. In cases of bilateral CTS, only the more severe hand was included. As part of the routine diagnostic workup, most patients received electrodiagnostic tests, in each case according to the guidelines of the American

Association of Neuromuscular and Electrodiagnostic Medicine[31]. Disease classification was done with a five scale categorization (normal, mild, moderate, severe, very severe)[32].

Patient characteristics and CTS data

Patient demographics were collected at baseline and included age, sex, affected side, bilateral presence of symptoms, dominance, disease duration, BMI, visual analog scale for pain, and mood scores assessed using the Center for Epidemiologic Studies Depression scale[33]. For this study, disease severity was classified in two ways: EDS-based classification and patient-reported outcome using the Boston Carpal Tunnel Questionnaire (BCTQ)[34]. The BCTQ was completed at baseline, and at 1 and 3 months post-surgery, with the 1 month follow-up done via mail and the 3 month follow-up completed in clinic. The BCTQ consists of two subscales, resulting in a symptom severity score (SSS) and a functional severity score (FSS). Scores range between 1 (no symptoms) to 5 (most severe) and for this study were categorized based on a system proposed by Storey et al. [35]: 1.0 ='asymptomatic', 1.1-2.0 = 'mild', 2.1-3.0 = 'moderate', 3.1-4.0 = 'severe', and 4.1-5 = 'very severe'. For the outcome measures of the post-surgical association, the difference in average BCTQ scores was used (baseline score - follow-up score) and averaged separately for the two subscales.

Ultrasound acquisition protocol

Patients and controls were tested in the supine position with the upper extremity on a plastic support board at a ~ 45° shoulder abduction angle. Subjects were then asked to flex and extend the third digit to a frequency of fifty beats per minute, paced by a metronome. After a practice session, three clips were recorded in the longitudinal view at the carpal tunnel inlet using the lunate and the distal radius as anatomical landmarks. All recordings were made using a Philips iE33 ultrasound scanner (Royal Philips Electronics, Amsterdam, the Netherlands) equipped with a 15L7 linear array transducer, set at a focal depth of 1.5cm. All measurements were done by ultrasonographers with a minimum of 1 year of experience in this field and two physicians (VS, SE) who were blinded to the clinical outcome and who were trained by the same ultrasonographer using a predetermined protocol.

Image analysis

A full and detailed description of the image analysis protocol has been published previously and showed high reliability[36]. Images were analyzed using a Matlab (R2016a, The MathWorks Inc., Natick, MA, 2000) based custom-made speckle tracking algorithm[28] enhanced with a singular value decomposition filter[29]. A region of interest (ROI) was manually placed on both the third flexor tendon (FDS) and the overlying SSCT (Fig. 1). Then, the ROI was automatically segmented in 40 and 24 kernels for, respectively, the tendon and SSCT. A normalized cross-correlation analysis was applied to calculate the frame-to-frame displacement. Finally, as described by Bandaru et al.[29], unreliable kernel results were removed based on their correlation values and discordant

vectors. The average of the displacement vectors of the reliable kernels gave the final frame-to-frame displacement vector. Analyses were done by a single rater blinded to patient data and clinical outcome. The average length of the x-axis vector was calculated over the course of a single movement based upon three cycles (three flexion and extension). The shear index is a measure introduced in previously published literature[27] and describes the relative motion between the FDS and SSCT. It is defined as ([tendon excursion] – SSCT excursion])/[tendon excursion]*100. A value close to 0% indicates a 1:1 movement between the SSCT and the tendon, whereas 100% would indicate a complete dissociation. Additionally, the ratio between maximum velocities of the SSCT and FDS was calculated based on the average peak values from three cycles.

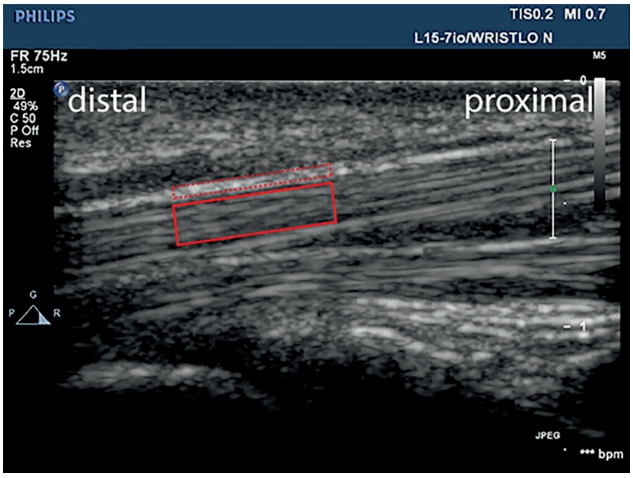


Fig. 1: Longitudinal ultrasound image showing the tendon and overlying hyperechoic SSCT. The overlays represent regions of interest of the third superficial flexor tendon (continuous line) and SSCT (interrupted line). Images were corrected for rotational angle before speckle tracking analysis.

Statistical analysis

Patient demographics were reported based on their distribution either as means including standard deviation or medians including their first and third quartiles. Categorical data is shown as counts and percents. Descriptive statistics of the excursions and US outcomes were expressed in means and standard deviation. To test aim 1: The relation between the shear index or maximum velocity ratio and disease severity was assessed using a one-way analysis of variance or Kruskal Wallis test based on data distribution followed by multiple means comparisons correction (Tukey HSD or Steel Dwass , respectively). For aim 2, two approaches were used to test the association between baseline US parameters and clinical outcome. First, a linear regression model was applied using the absolute change in separate BCTQ scores as main outcomes. Tested covariates as potential confounders included the baseline BCTQ score, sex, age, disease duration, BMI, and baseline depression score. To obtain consistent estimates of the variance of the

model caused by heteroscedastic residuals, White robust variance estimators[37] were used. Secondly, we categorized patients based on their shear index as either "low", "normal", or "high" using the 95% confidence interval of the healthy control group as thresholds. This was used as the independent variable of a logistic regression model adjusted for the same confounders. Here, we dichotomized the BCTS-SSS scale to "success" and "not success" based on the relative minimal clinically important difference[38]. Type I error rate was set at 0.05. All analyses were done using SAS™ (version 9.4; SAS Institute, Cary NC) and JMP (Version 14, SAS Institute Inc., Cary, NC, 1989-2007.).

Results

Descriptive Overall Results

A total of 124 participants completed the baseline assessment, of whom 90 (71%) had US images which could be analyzed for SSCT movement. The main reason for image exclusion was out-of-plane motion of the SSCT or the tendon. In this sample, follow-up rate was 72% and 90% for 1 and 3 months follow-up, respectively. The demographics and the distribution over the different disease severities are described in Table 1. At baseline, US measures gave a mean of 79% (SD: 13%) for the shear index, based on 1.2 mm (SD: 0.4 mm) excursion of the tendon and 0.24 mm (SD: 0.15 mm) total excursion of the SSCT (Table 2).

Table 1. Baseline characteristics of CTS patients

Age, years (mean, SD)	55.8±14.6
Sex (female)	55 (61%)
Duration of symptoms, months (median, IQ_{1-3})	24 (12-44)
Surgery on dominant hand (yes)	55 (63%)
Bilateral CTS (yes)	52 (60%)
BMI, kg/m^2 (mean, SD)	31.8±6.4
Pain score, visual analogue scale (median, IQ_{1-3})	3 (2-5)
EDS severity (%)	
Normal	2 (2%)
Mild	25 (28%)
Moderate	41 (46%)
Severe	19 (21%)
Very severe	1 (1%)
No NCS available	2 (2%)
Symptom Score severity (%)	
Asymptomatic	0 (0%)
Mild	7 (8%)
Moderate	35 (39%)
Severe	40 (44%)
Very severe	7 (8%)
Missing/ incomplete	1 (1%)

BMI: body mass index, IQ: Interquartile, EDS: electrodiagnostic study, SD: Standard deviation. N=90.

Table 2. Ultrasound parameters of CTS patients at baseline

Tendon excursion, cm (mean, SD)	1.2±0.4
SSCT excursion, cm (mean, SD)	0.24±0.15
Shear index, % (mean, SD)	78.9±12.8
Maximum velocity ratio, cm/s (mean, SD)	0.30±0.15

SD: Standard deviation.

Shear Index and Disease Severity at Baseline

Figure 2 shows the shear indices relative to CTS severity based on either EDS results or patient-reported assessment. With only small sample sizes in the symptom score-based 'mild' and 'very severe' categories, these groups were merged with 'moderate' and 'severe' respectively, since these were found to be clinically most alike. A single patient who had a 'very severe' EDS was merged with the 'severe' group, but the two cases with 'normal' EDS were excluded from subsequent analyses. Analyses of variance showed a significant statistical difference in average shear index between categories based upon EDS severity (p=0.0475). Post-hoc mean comparisons showed that the mean shear index in the moderate EDS severity group was 8.6% higher than that of the milder CTS cases (p=0.0389). However, no association of shear index with the initial symptom severity score from the BCTQ (p=0.609) was found. The shear index averages, the 95% confidence intervals per group, and the results of the pairwise comparisons can be found in the Supplements (S-1). For the maximum velocity ratio (MVR), overall analysis of variance showed a similar pattern as the shear index, with the EDS based disease severity having a greater association with MVR (p=0.072) compared to the symptoms score comparison (p=0.207). None of the pairwise comparisons with the maximum velocity ratio showed statistical significance.

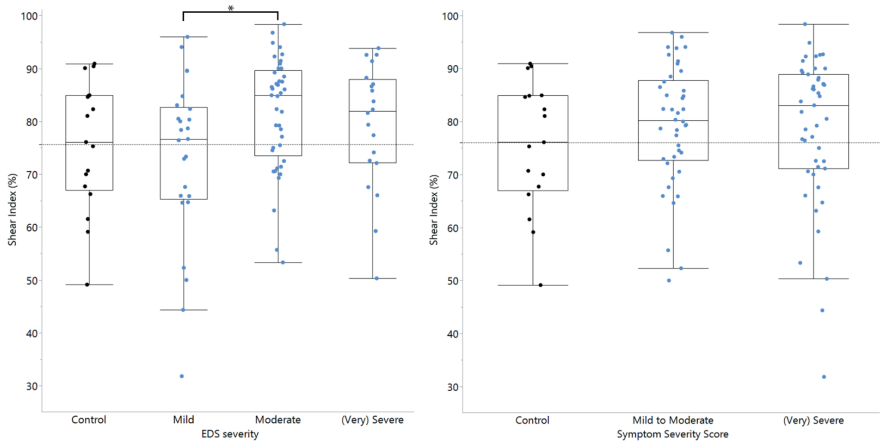


Fig. 2: Boxplots showing individual samples of shear index values at baseline. They are categorized by CTS severity defined by electrodiagnostic studies (left) or patient-reported symptom severity score (right). Dashed lines represent mean value of the control group. Overall ANOVA results showed difference in means for EDS based severity (p=0.0475), but not for symptom based severity (p=0.609). *p=0.0389 based on pairwise means comparison corrected for multiple comparisons.

Shear Index and Association with Surgical Outcome

BCTQ scores improved for all patients after surgery for both the SSS ($p<0.001$) and the FSS ($p<0.001$); The SSS improved from 3.1 (SD: 0.7) to 1.6 (SD: 0.5) at 1 month and 1.5 (SD: 0.5) at 3 months post-surgery. The FSS improved from 2.5 (SD: 0.8) to 1.7 (SD: 0.5) and 1.5 (SD: 0.6) for the same time points respectively (S-2). Adjusted linear regression models showed no apparent association between either of the US parameters with symptomatic or functional improvement at one or three months follow-up (Table 3). To also test extreme values of the shear index for prognostic potential, the shear index was split into three groups (high, middle, low) and used as an independent variable. The surgical outcome was dichotomized into "success" and "not success" groups using the minimal clinically important difference. Out of those who completed the follow-up time points, 39 patients (61%) were categorized as successful at one month and 52 patients (66%) at three months. Again, no statistically significant associations were found between shear index and surgical outcome at 1 month ($p=0.42$) or 3 months ($p=0.60$). More detailed chart reviews were done of the five patients with the highest and lowest shear indices but they did not stand out from the others in terms of symptom severity score changes, intra-operative findings, or nerve size and dynamics.

Table 3. Linear regression model for baseline SI versus clinical outcome

	Δ Symptom Severity Score				Δ Functional Status Score			
	1 month		3 month		1 month		3 month	
	Estimate (β)	p-value*	Estimate (β)	p-value‡	Estimate (β)	p-value‡	Estimate (β)	p-value#
Shear index, %	0.003	0.49	-0.004	0.27	0.007	0.18	-0.004	0.25
Maximum Velocity Ratio	-0.02	0.58	0.51	0.10	-0.15	0.74	0.67	0.06

Modeled with linear regression, adjusted for *baseline BCTQ and age; ‡ baseline BCTQ; # baseline BCTQ and CES-D score

Discussion

This study looked at the added value of longitudinal dynamic ultrasound in the assessment of disease severity and the association with post-surgical outcome. We found a significant decrease in SSCT motion (i.e. increase in shear index) between mild and moderate CTS patients, suggesting that the SSCT became less adherent to the tendon as severity increased. However, there was no association between pre-operative shear index and self-reported clinical symptom (improvement) after carpal tunnel release surgery.

Current literature suggests that diminished nerve excursion is present in patients with CTS[39] during finger and wrist motion in both the transverse[40-45] and in the longitudinal plane[25,44,46,47]. In addition, mechanical studies in cadaver tissue have shown that the SSCT is prone to damage even with normal finger tendon excursion (for example, if adjacent fingers are moving in opposite directions)[15,16,48] and that progressive fibrosis of the SSCT is present in synovial biopsies from CTS patients[18]. Combining these observations, it has been suggested that the SSCT could play a central role in the initiation and progression of CTS[23,49]. A previously published pilot study of intra-operatively measured relative SSCT movement in CTS patients reported a bimodal distribution where the SSCT excursion was either very closely related to the tendon motion or dissociated from it[50]. This led to a hypothetic model that pathological SSCT changes could be divided into two categories: (1) With increasing fibrosis, the SSCT loses the mechanical response characteristic of a viscoelastic structure and adheres to the tendon during motion and (2) with progressive damage and continued repetitive, high force tendon motion, eventually the fibrotic connections between the closest layers of SSCT and the tendon are disrupted, causing a physical dissociation from the tendon and subsequent absence of SSCT motion (Fig. 3). Both descriptions have been observed in CTS patient samples[10,50] but have never been correlated to US findings. The first category of patients would be associated with a lower shear index while the second description would produce a higher shear index value. In our dataset, we did find some patients who showed low shear indices, but most of these values were within the range of the normal controls. The dominant trend was a higher shear index with increasing severity, which fits with this concept of SSCT dissociation from the tendon. Currently, the US resolution is not adequate to characterize the SSCT in more detail, but in the future, higher-resolution imaging options will become available to enhance image processing.

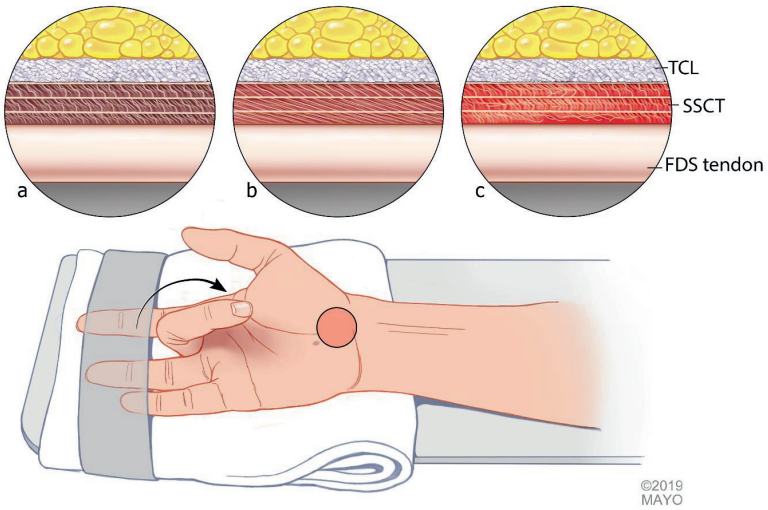


Fig. 3: Hypothetic model with two stages during CTS. A) During anatomical rest position, the absence of tendon loading leaves the SSCT in a relaxed state. B) During finger flexion and extension, longitudinal motion of the tendon causes the recruitment of SSCT layers. In CTS, fibrosis of the SSCT could alter this motion to a more closely related movement. C) With time, additional damage to the SSCT disrupts the architecture and causes dissociation between the movement of the tendon and the surrounding structures.

The first aim of this study was to test the SSCT as a potential proxy for disease severity, as has been done before for the cross-sectional area of the nerve. Compared to the median nerve, however, non-invasively visualizing and quantifying the SSCT behavior is complicated due to its thinness, viscoelastic properties, and the potential effects of variability in the joint range of motion. So far, multiple studies have assessed SSCT motion longitudinally using either Doppler[19-21] or a post-processing speckle-tracking based algorithm either using commercially available software[26,27] or custom-designed[24,25]. Doppler is easy to use but loses its validity when the moving structures start to approach an angle perpendicular to the probe's surface. Speckle tracking algorithms are not limited by this since it is a post-acquisition processing technique that follows acoustic signals from frame-to-frame with a pattern matching principle.

Four of the previously-mentioned studies included CTS patients[21,24-26]. From these, Tat et al. looked at eleven patients with self-reported CTS symptoms and compared them to an equally sized healthy group. Sample sizes were too small to directly associate the BCTQ scores to the shear index, but they reported a statistically significant higher shear index (30.2% versus 21.7%, p=0.016) in the CTS patients[21]. This was in agreement with data from van Doesburg et al., who assessed 18 CTS patients and compared the shear index to 22 healthy volunteers. Using commercial speckle tracking software, they found a shear index of 48% in the CTS patients, which also was higher than that of the controls (36%, p<0.05) [26]. Again this study was not aimed at testing a direct association with disease severity and the authors commented that their method required further optimization to increase sensitivity for SSCT tracking. The other two studies were larger

in their sample size and aimed at testing disease severity with SSCT measures, utilizing a similar speckle tracking algorithm as was used in this study. Korstanje et al. included 55 CTS patients and compared the most affected with the least affected hand based on clinical and NCS classification. An increase in absolute SSCT excursion was reported in the most affected hand (p=0.025), but this was also the case for the FDS tendon (p=0.008), thus it remains unclear whether the relative shear index was different. Finally, a larger clinical study where US images were analyzed with speckle tracking showed that the majority of patients had higher shear indices than the controls[24]. Interestingly, their shear index values ranged between 41-61% whereas the majority of our values ranged between 72-88%. This difference was mostly caused by the higher SSCT displacements Filius et al. registered. Possible explanations for this discrepancy could be the noise filter that was added to our speckle tracking algorithm[29], which prevents overestimation of motion detection by eliminating background noise, and also the difference in imaging protocol since speckle tracking is dependent on velocity and total excursion amount. Overall, these studies confirm our finding that CTS is associated with increased shear index and thus decreased SSCT motion, albeit with outliers who do not necessarily correlate with different initial severity or subsequent outcome.

The major advantage of this study is the larger clinical population used to specifically address the clinical usefulness of relative SSCT displacement. We aimed to combine pathophysiological findings with innovative non-invasive evaluation tools in order to find a patient-centered non-invasive tool that might correlate a biomechanical property with clinical parameters, including the ability to predict outcomes. Both symptomatic and functional evaluation measures were used as reference points for disease severity at baseline. Limitations of our approach include those intrinsic to the speckle tracking method and the small size of the SSCT, which resulted in a relevant loss of ultrasound data: out of plane motion cannot adequately be corrected and the ROI is a set framework through which the structures move, rather than identifying the speckles and dynamically tracing them. We found our method to have higher reliability in terms of intra- and inter-rater repeatability[36], a feature which is highly desirable when looking at potential diagnostic and prognostic applications, but the trade-off was that speckles could fall outside the ROI, resulting in a less accurate measurement. In addition, speckle tracking is based on the assumption that the SSCT can be viewed as a solid structure, which allows averaging the different read-out layers during speckle tracking. Nonetheless, given the current resolution of the images, this simplification likely had a limited effect.

Finally, the dataset was limited to CTS patients undergoing surgical intervention. This could have biased the baseline to be more severe than is found in the average CTS population, for whom conservative treatment plays a prominent role. Carpal tunnel release surgery has been known to be highly effective, although reports on wide variation in success have also been published. During design, we anticipated a symptomatic improvement almost twice as small as what we actually found. In addition, we observed less variation than expected, so only a relatively small proportion of our patients

showed less optimal results. This complicated our ability to correlate the shear index with outcome. We do plan to assess the shear index in a sample of non-operatively treated CTS patients and in those with other forms of CTS (for example, associated with diabetes, where the fibrosis may be more profound and clinical outcomes less predictable) to further test the possible value of the shear index in identifying the best treatment for the right patient.

Conclusion

The SSCT moves less in patients with CTS, resulting in a higher shear index, which significantly increases with increasing disease severity. This fits the underlying pathophysiological model of SSCT disruption and disconnection from the tendon. The relative movement of SSCT can be measured, but we were unable to show a clear association with patient-reported outcome after surgical intervention, possibly because our patients did exceptionally well after surgery, and also due to the fact that SSCT thickness is close to the limit of resolution of current US technology. Given these findings, dynamic ultrasound of the SSCT seems to have limited prognostic value in patients undergoing surgery for idiopathic carpal tunnel syndrome. We plan to investigate this in an injection cohort to see if this measure performs differently in patients who are treated non-operatively, where treatment failures are substantially more common.

Summary

The connective tissue in the carpal tunnel is less responsive to tendon motion in CTS patients with increased electrophysiological severity. This can be assessed with dynamic ultrasound, but inter-patient values present in a wide variation and seem not to be associated with short-term patient-reported outcome in a surgical cohort.

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Supplements

Suppl. 1. US parameters and pairwise comparison

		Shear index			Maximum Velocity Ratio		
Summary data	**N**	**Mean ± SE**	**95% Confidence Interval**		**Mean ± SE**	**95% Confidence Interval**	
EDS Severity							
Controls	17	75.6 ± 3.1	69.6 – 81.6		0.37 ± 0.04	0.30 – 0.44	
Mild	25	72.9 ± 2.5	68.0 – 77.9		0.35 ± 0.03	0.29 – 0.41	
Moderate	41	81.5 ± 1.9	77.7 – 85.4		0.27 ± 0.02	0.22 – 0.32	
(Very) Severe	20	79.2 ± 2.8	73.7 – 84.8		0.30 ± 0.04	0.23 – 0.37	
Symptom Severity Score							
No CTS	17	75.6 ± 3.1	69.4 – 81.8		0.37 ± 0.04	0.30 – 0.44	
Mild to Moderate	42	79.2 ± 2.0	75.3 – 83.1		0.30 ± 0.02	0.25 – 0.35	
(Very) Severe	47	78.5 ± 1.9	74.8 – 82.2		0.29 ± 0.02	0.25 – 0.34	
Anova data		**Mean difference ± SE**	**95% Confidence Interval**	**p-value***	**Mean difference ± SE**	**95% Confidence Interval**	**p-value***
EDS Severity							
Controls vs							
Mild		2.6 ± 3.9	-7.6 – 12.9	0.91	0.02 ± 0.05	-0.10 – 0.15	0.97
Moderate		5.9 ± 3.6	-3.5 – 15.3	0.35	0.10 ± 0.04	-0.01 – 0.22	0.11
(Very) Severe		3.6 ± 4.1	-7.1 – 14.4	0.81	0.07 ± 0.05	-0.06 – 0.20	0.50
Mild vs							
Moderate		8.6 ± 3.2	0.3 – 16.9	0.04	0.08 ± 0.04	-0.02 – 0.18	0.18
(Very) Severe		6.3 ± 3.7	-3.5 – 16.1	0.34	0.05 ± 0.05	-0.07 – 0.17	0.72
Moderate vs							
(Very) Severe		2.3 ± 3.4	-6.6 – 11.2	0.91	0.03 ± 0.04	-0.08 – 0.14	0.90
Symptom Severity Score							
No CTS vs							
Mild to Moderate		3.6 ± 3.7	-5.1 – 12.4	0.58	0.07 ± 0.04	-0.04 – 0.17	0.27
(Very) Severe		2.9 ± 3.6	-5.7 – 11.5	0.70	0.08 ± 0.04	-0.03 – 0.18	0.20
Mild to Moderate vs							
(Very) Severe		0.7 ± 2.7	-5.7 – 7.2	0.96	0.01 ± 0.03	-0.07 – 0.08	0.97

EDS: Electrodiagnostic studies; SE: standard error. *ANOVA means comparisons with p-values corrected for multiple comparisons using Tukey Kramer.

Suppl. 2. PROM scores before and after carpal tunnel release

	N	Mean ± SD	Mean difference (95% CI)	p-value*
BQCTS - Symptom score				
Baseline	89	3.1 ± 0.7		
1 month	64	1.6 ± 0.5	-1.4 (-1.6; -1.3)	<0.001
3 month	79	1.5 ± 0.5	-1.5 (-1.7, -1.4)	<0.001
BQCTS - Function score				
Baseline	86	2.5 ± 0.8		
1 month	63	1.7 ± 0.5	-0.8 (-1.0; -0.6)	<0.001
3 month	78	1.5 ± 0.6	-1.0 (-1.1, -0.7)	<0.001
VAS – pain				
Baseline	89	3.5 ± 2.2		
1 month	63	1.7 ± 0.7	-1.8 (-2.3, -1.2)	<0.001
3 month	82	1.5 ± 0.9	-2.0 (-2.4, -1.5)	<0.001

All participants (100%) improved after surgical intervention.
SD: standard deviation, PROM: patient-reported outcome measure, BQCTS: Boston questionnaire for carpal tunnel syndrome, VAS: Visual analog scale. Mean differences shown are relative to baseline value.
* Paired t-tests with statistical significance reached at p<0.05

6

CHAPTER 6.

Median nerve transverse mobility and outcome after carpal tunnel release

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Ultrasound in medicine & biology, 45(11), 2887-2897.

Abstract

Introduction
Nerve movement is decreased in patients with carpal tunnel syndrome and can be assessed with ultrasound. In addition to morphological features, this study describes a novel approach by assessing nerve movement and the association with short-term patient-reported outcome.

Methods
Ultrasound images at the carpal tunnel inlet were acquired during finger and wrist flexion. Linear regression models were used with the Boston Carpal Tunnel Questionnaire as main outcome.

Results
85 Patients were included, 93% completed 3-month follow-up. Pre-surgical mean nerve area was 14.5±4.2mm^2 and decreased to 13.3±3.8mm^2 ($p<0.001$). Displacement in dorsal direction with wrist flexion increased from 1.9±1.3 to 2.4±1.3mm ($p<0.01$). A pre-surgical larger nerve area was associated with more functional improvement ($\beta= -0.024$, $p=0.02$), but baseline mobility was not.

Conclusion
Change in excursion with finger flexion was associated with symptomatic improvement, but with a small effect ($\beta=-0.05$, $p=0.01$). This indicates that there is limited prognostic potential for dynamic transverse ultrasound in carpal tunnel syndrome.

Introduction

Carpal tunnel syndrome (CTS) is a common compression neuropathy with an estimated prevalence of 3-12% in a working population[1,2]. Carpal tunnel release (CTR) surgery is the best option for some patients in order to prevent progression of motor nerve dysfunction, but also the next treatment in line after conservative treatment fails[3]. Although CTR is effective in reducing pressure, there is wide variation in patient-reported success rates of CTR, ranging from as low as 27% to as high as 100%[4]. Possible reasons for failure as mentioned by Dr. Bland and others[5] include misdiagnosis, intra-operative errors, suboptimal results as found in very severe cases, and the variation in the definition of success. The range also indicates the relevance of adequate selection of surgical candidates. To date, it has been difficult to identify specific factors that support surgical outcome prediction[6-8]. It seems likely that not a single, but a multitude of preoperative factors, could potentially help support consideration for surgical intervention.

So far, the role of ultrasound has been established for the diagnosis of CTS with patients showing a larger cross-sectional nerve area with subsequent ligament bowing and decreased flattening just proximal to the carpal tunnel inlet[9]. However, the prognostic potential, of these static, morphological characteristics has shown conflicting results on short term and little relation with long-term surgical outcome[10,11]. In addition to nerve size, transverse dynamic ultrasound has successfully been used to evaluate the flexor tendon and nerve movement in response to finger and hand movement[12-15].

There is evidence that fibrosis of the subsynovial connective tissue (SSCT) plays an important pathological role in CTS[16-19]. The SSCT is a multilayered tissue surrounding the tendons and nerve within the carpal tunnel. As the tendons move, the SSCT moves along, but is prone to damage with excessive excursions[20-22], which can lead to a non-inflammatory response with progressive fibrosis[23-25]. Although the presence of causality or direction of association between CTS development and SSCT fibrosis is still unknown, CTS patients have been found to have thicker SSCT compared to controls[26] and dynamic ultrasound studies have shown decreased mobility of the nerve in patients with CTS[27-32]. Hypothetically, these differences could be attributed to the decrease in nerve size[33] and the mechanical restriction of the fibrotic SSCT, with the nerve "sticking" to the transverse carpal ligament. An increase in CTS severity has been associated with a decrease in nerve gliding movement[34] and Nanno et al. showed significant changes in median nerve motion after CTR[35]. Nerve dynamics may thus hold useful information pertaining to CTS pathophysiology, but so far there has been limited research associating this with clinical outcome.

In this study we aim to build on these findings by exploring the prognostic potential of a combination of static and dynamic nerve characteristics using the change in patient reported outcomes as the primary outcome. In addition, nerve mobility before and after

CTR will be compared, hypothesizing an increase in excursion at the three month time point.

Materials and Methods

General

This is a prognostic, prospective (level I) study with a sample of surgical patients from a larger clinical trial (CT.gov identifier NCT02219555) which was reviewed and approved by our Institutional Review Board. Written informed consent was obtained from all participants in accordance with the Declaration of Helsinki.

Subjects and disease characteristics

All patients with idiopathic CTS were approached and recruited from a single institution with a specialized hand clinic. The diagnosis and surgical indication was determined by the attending hand physicians based on a combination of symptoms at presentation, clinical tests and supporting electro-diagnostic tests. Clinical evaluations included sensibility (two point discrimination) in the median nerve distribution of the hand, Phalen's test, Tinel's sign, pinch and grip strength, and thenar muscle atrophy. After the diagnosis of CTS was made and the decision for surgical treatment was agreed upon, patients were approached for the study. This was an open label study and no restrictions based on type of surgical procedure or post-operative protocol were applied. Patients were recruited if they met the following inclusion criteria: clinical diagnosis of CTS, age between 21-80 years, symptoms of numbness or tingling for at least four weeks in at least two digits on one hand including the thumb, index, long or radial border of ring finger, full understanding of English language, and indication for surgical release surgery. Exclusion criteria were a previous surgical release, tumor, mass or deformity in hand/wrist, pregnancy induced CTS, and any of these diagnoses: cervical radiculopathy, peripheral nerve disease, thyroid disease, rheumatoid arthritis or other inflammatory arthritis, osteoarthritis in wrist, diabetes, renal failure, sarcoidosis, amyloidosis or major trauma to ipsilateral hand or wrist. If patients experienced bilateral CTS, only the hand with most severe symptoms was included. No exclusions based on previous treatment, except surgical intervention, were applied.

As part of routine diagnostic medical care, all patients underwent electrodiagnostic tests using the Viking Select or Viking EDZ EMG machine (CareFusion, San Diego (CA), USA) according to the guidelines from the American Association of Neuromuscular and Electrodiagnostic Medicine. Part of the recording included measuring the sensory latency, velocity and sensory nerve action potential amplitude, the distal motor latency, velocity in forearm and compound muscle action potential from the abductor pollicis brevis muscle. The severity grade was based on the results of the individual components with values ranging between 1 and 5 with 1 indicating normal test results and 5 very severe CTS[36].

Patient reported outcomes

The Boston Carpal Tunnel Questionnaire (BCTQ)[37] was used to assess patient reported clinical outcome at both baseline level and three months after surgery. This questionnaire consists of 19 questions divided over symptom severity (SSS; 11 questions) and functional status scales (FSS; 8 questions). Each answer can range from 1 (no symptoms) to 5 (most severe) and are averaged to result in two separate scores.

Ultrasound acquisition protocol

Ultrasound images were acquired at two time points; before surgery and three months after surgery. Patients were placed in supine position with the affected arm stretched out in 70-80° abduction on a Plexiglas board. A Velcro strap was used around the mid forearm to minimize movement and pillow cases under the hand were used to place the wrist in neutral position. All images were made using a Philips iE33 (Royal Philips Electronics, Amsterdam, the Netherlands) ultrasound machine with a L15-7io linear transducer. Images were taken, all in a transverse plane, just proximal to the level of the carpal tunnel inlet defined as the proximal margin of the flexor retinaculum. To minimize compression to the carpal tunnel structures, the transducer was applied to the skin without additional pressure and with a surplus of gel.

Patients were asked to perform two tasks: finger flexion and palmar wrist flexion (Fig. 1). For the first, starting from a neutral position, patients were asked to make a fist; start from a full extension position, flex the four fingers, without thumb adduction, over the course of seven seconds. For the latter, again starting from a neutral position, patients were asked to keep the fingers extended and flex the hand until a maximum range of motion was reached. Movements were chosen based on their clinical relevance, with CTS patients often complaining of symptom aggravation after a period of wrist flexion and physicians using this feature as part of their diagnostic exam (Phalen's test). No formal quantification of range of motion was done, assuming similar movements within patients and reflecting clinical practice. After a practice round, a set of three clips was recorded for each movement and the best quality image selected for analyses.



Figure 1. Hand motions. From left to right: Wrist in neutral position, finger flexion, wrist flexion.

All measurements were done by ultrasonographers with a minimum of 1 year US experience and two physicians (SE, VS) who were blinded to the clinical outcome and who were trained by the same ultrasonographers while using a predetermined protocol.

Image analysis

Images were analyzed using an in-house software package (Analyze v12.0, Biomedical Imaging Resource, Mayo Clinic, Rochester MN, USA). All parameters were measured by manually placing a polygon region of interest (ROI) around the inner edge of the nerve epineurium. Parameters acquired from static images included median nerve cross sectional area (CSA), perimeter and circularity. A perfect circle would present with a value "1" whereas non-circular / irregular shapes have higher values. Nerve mobility parameters were based on Cartesian coordinates x and y from the center point of the median nerve polygon (Fig. 2). Overall displacement is graphically presented in a two dimensional plot including an ellipse showing the standard deviation standard ellipse as well as Hotelling's (95%) confidence ellipse of the average start and end positions. The initial start position of the nerve varies per patient and time so this point was used for normalization (coordinate 0,0). Additionally, Pythagorean theorem was used to calculate the gross displacement including *overall displacement* during a movement (end position – initial position) and *summed displacement*. A randomly selected set of ten images from the study cohort were used to confirm intra- and inter rater reliability; the test-retest assessment images were acquired with an identical protocol from seventeen volunteers without CTS symptomology. This was done in order to test repeatability without surgical interference.

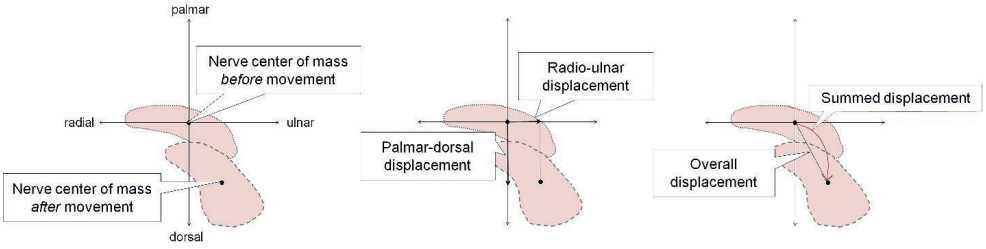


Figure 2. Nerve mobility parameters. Left: The nerve's center of mass was determined first in a dual plane field and used to normalize the start position in neutral wrist and finger posture. Middle: Displacements in radio-ulnar and palmar-dorsal direction. Right: Mobility was defined both as the difference in end-start position of the nerve (black arrow; overall displacement) and as a cumulative parameter describing the entire course of the nerve (red arrow; summed displacement).

Statistical analysis

Continuous variables were compared before and after surgical treatment using paired t-tests. Association of the baseline US parameters as well as the change in US parameters versus the change in BCTQ were tested using sex and baseline BCTQ-adjusted linear regression models. Potential confounders tested were age, sex, BMI, duration of symptoms, NCS based severity, and the respective BCTQ baseline scores. The dependent variable was the change in BCTQ scales after three months. White robust variance estimators[38] were used to obtain consistent estimates of the variance of the model parameters due to heteroscedastic residuals. For secondary analysis, logistic regression models were created using a cut-off score of 1 on the SSS subscale as dependent variable, based on the minimal important difference reported for this subscale[39,40]. These were also adjusted for baseline SSS and sex. Intra-rater data was collected by a single rater (VS) by measuring the same images twice with a four week interval. Inter-rater measurements were based on the same images made and analyzed by two raters (VS, SE). For the test-retest data, two clips were collected from healthy volunteers with a four-week interval between them and then analyzed in sets by either of the two raters (VS, SE) who were not blinded to the origin of the images. All agreement measurements were calculated using intra-class correlation coefficients (ICCs) including the 95% confidence interval using a two-way mixed effects model assuming single measure and absolute agreement[41]. ICC values above 0.75 are considered as excellent, 0.40–0.74 are fair to good and values below 0.40 are considered as poor[42]. Type 1 error rate was set at 0.05; no multiple testing corrections were done. Analyses were done using SAS™ (version 9.4; SAS Institute, Cary NC) and JMP (Version 13. SAS Institute Inc., Cary, NC, 1989-2007.).

Results

Patient characteristics

A total of 85 patients were included in the study of whom 79 (92.9%) completed the three month follow up visit. Baseline characteristics including age, sex, duration of symptoms, whether the included hand was the dominant hand and NCS severity are summed in Table 1.

Table 1. Patient demographics (n=85)

Age, years (mean, SD)	57.1 (13.5)
Sex (female)	65%
Duration of symptoms, months (median, IQ_{1-3})	24 (10-44)
Surgery on dominant hand	58.3%
BMI (mean, SD)	30.9 (6.5)
NCS severity	
Normal	3.5%
Mild	24.7%
Moderate	49.4%
Severe	15.3%
Very severe	2.4%
No NCS available	4.7%

BMI: body mass index, NCS: Neuro conductive study

Reliability

Overall, reliability of the ultrasound images of the CTS patients was labeled good to excellent with ICC values ranging between 0.75-0.99. Test-retest data using ultrasound images of healthy volunteers was high for the static measurements (ICC: 0.88-0.96), but moderate for the dynamic assessment (ICC: 0.56-0.76). All ICC values are described in Table 2.

Table 2. Ultrasound reliability

	Intra-rater ICC (95% CI)	Inter-rater ICC (95% CI)	Test-retest ICC (95% CI)
Nerve CSA	0.94 (0.76-0.99)	0.94 (0.79-0.84)	0.96 (0.90-0.99)
Nerve perimeter	0.97 (0.81-0.99)	0.75 (0.18-0.93)	0.91 (0.76-0.97)
Nerve circularity	0.97 (0.89-0.99)	0.92 (0.41-0.98)	0.88 (0.69-0.95)
Finger flexion – overall displacement	0.99 (0.98-0.99)	0.99 (0.98-0.99)	0.69 (0.31-0.88)
Finger flexion – summed displacement	0.99 (0.96-0.99)	0.99 (0.97-0.99)	0.69 (0.21-0.89)
Wrist flexion – overall displacement	0.99 (0.97-0.99)	0.99 (0.97-0.99)	0.56 (0.13-0.82)
Wrist flexion – summed displacement	0.98 (0.93-0.99)	0.98 (0.93-0.99)	0.76 (0.45-0.91)

Intra-class correlations (ICC) for ultrasound parameters. CI: Confidence interval, CSA: Cross sectional area.

Changes in patient reported outcome and US parameters after CTR

Patient reported outcomes improved (decreased in score) significantly after carpal tunnel release surgery to a mean score of 1.5 on both the SSS and FSS (mean difference -1.6 and -1.1 resp. with $p<0.001$ for each). Fig. 3 shows the mean differences of the ultrasound parameters (exact data can be found in Appendix A). Nerve CSA was on average 14.5±4.2 mm^2 before surgery and decreased significantly to 13.3±3.8 mm^2 ($p<0.001$). Of the dynamic measurements, the abstracted palmar dorsal axis showed a significant increase in mean displacement from 1.9 to 2.4 mm in dorsal direction (mean difference: 0.54 mm, p=0.003). A visual example of the change in nerve displacement is given in

Appendix B. Additionally, the summed displacement of the nerve during wrist flexion increased on average with 0.7 mm (p=0.038). Fig. 4 shows the end position of the nerve after flexion of the fingers and wrist relative to where it started. Note that these plots do not depict the entire pathway but only the end position and that these are limited to the individual representation of both baseline and follow up data points. These plots indicate that finger flexion causes a median nerve translation in mostly ulnar and slightly palmar direction which does not change significantly after surgery. Wrist flexion induces a movement that results in a more in ulnar-dorsal direction. The individual data points show that there is a large variation between patients in nerve end points in wrist flexion both before as well as after surgical intervention.

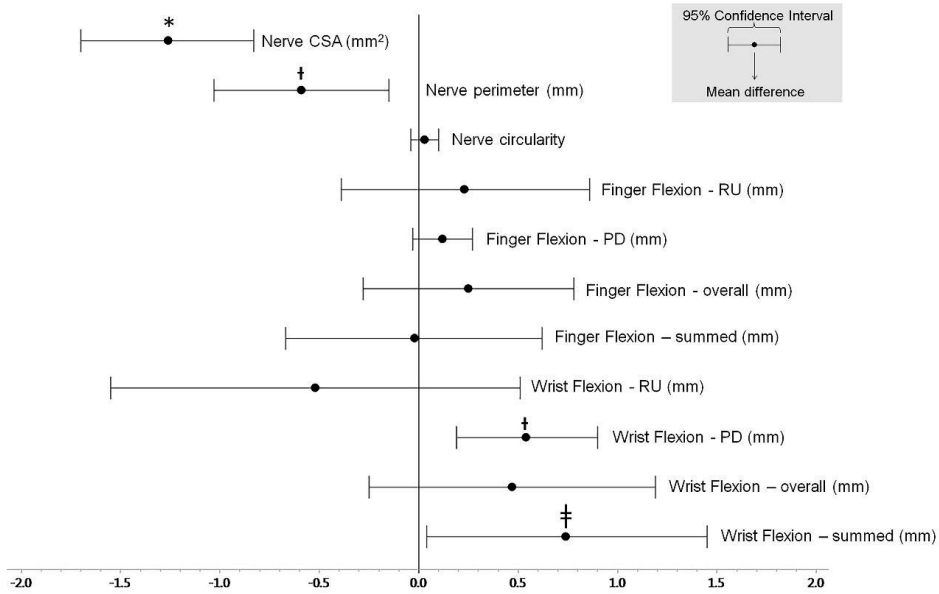


Figure 3. Change in US parameters after carpal tunnel release surgery. Data visualized as the mean difference with the 95% confidence interval. Units are defined behind the data labels. Mean differences were calculated by subtracting baseline values from follow up values. RU: radial-ulnar, PD: palmar-dorsal, CSA: cross-sectional area, *p<0.001, ǂ p<0.01, ǂp<0.05.

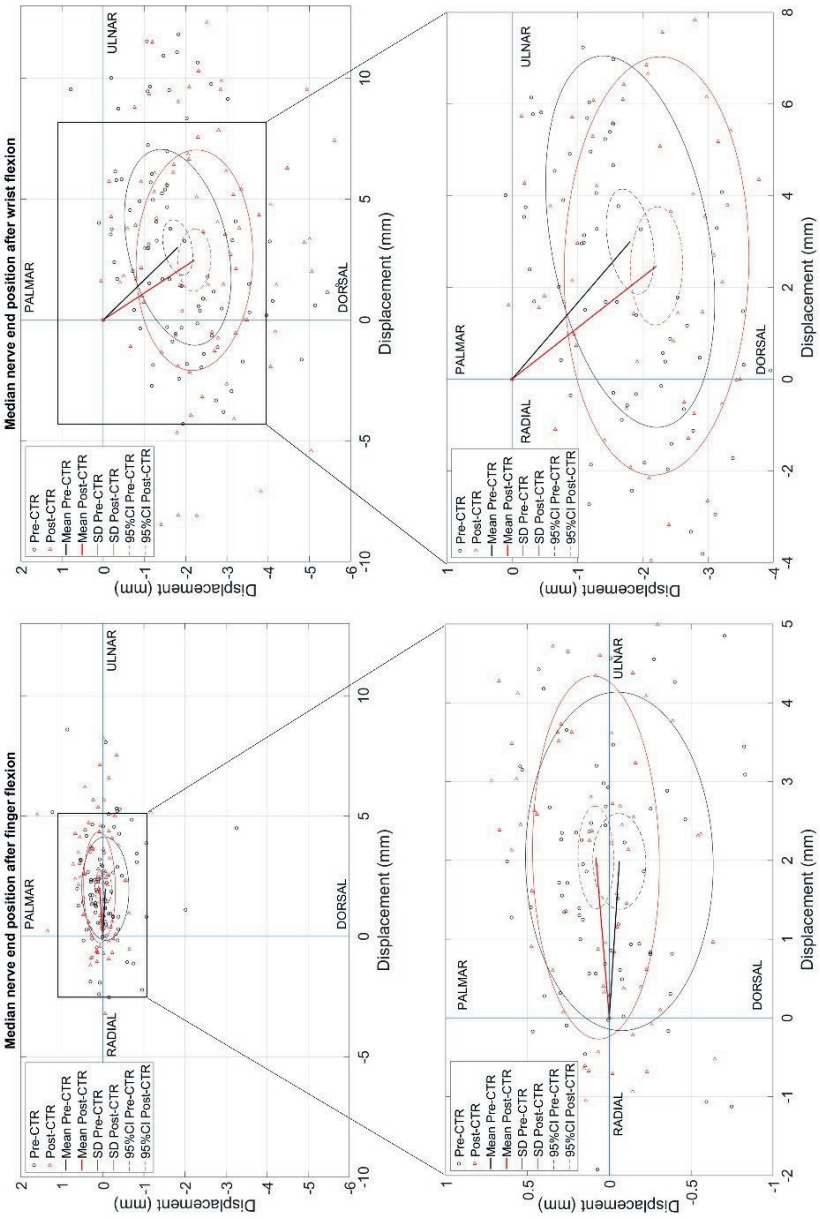


Figure 4. Cartesian plots of the individual and the average nerve displacement end points before and after surgery. Within the plots, both the baseline (red) and the follow up (black) data are depicted. Circles and triangles represent the individual patients with the solid straight line showing the average difference in end position of the nerve. Note that all end points have been normalized to where the nerve originally started. The standard deviation (SD; solid ellipse) and 95% confidence ellipse (interrupted ellipse) are shown as well. Upper plots show all patients, lower plots are zoomed in representations of the average data. The left plots show the data for the finger flexion and the right plots show the data for the wrist flexion. CTR: Carpal tunnel release.

Association between baseline US and BCTQ changes

All static and dynamic US parameters have been assessed in linear regression models adjusted for their corresponding BCTQ baseline scores and sex. The outcome of choice for the primary analysis was the change in the individual BCTQ scales. Detailed results are shown in Table 3. Out of the static measures, only nerve CSA and perimeter showed a significant association with the function scale difference (β= -0.024, p=0.02 for CSA and β= -0.049, p=0.02 for perimeter). None of the baseline measures showed a significant association with the change in symptom severity score. Using the minimal clinically important difference as the cut-off, 20% (n=17) of patients had a poor outcome. This is plotted in a gradient map where the size of the change in symptom score per individual is indicated in relation to the displacement end point (Figure 5). No obvious subsets of patients were identified as having either an extremely poor or successful outcome. Similar plots were also made for the FSS, but no subsets were visible (figures not shown). A formal comparison using logistic regression also indicated no parameters to be associated with a specific type of outcome.

Association between changes in US and BCTQ changes

Results for the changes in US parameters between baseline and three months post surgery and the association with changes in the BCTQ scores are described in Table 4. The change in overall displacement of the nerve during finger flexion showed a significant association with change in function score (β= -0.060, p=0.04). The same parameter as well as the change in displacement in radio-ulnar direction during finger movement were associated with the change in symptom severity score (β= -0.050, p=0.01 and β= -0.035, p=0.03 respectively). All associations were also tested using a percent change in score, but except for losing significance in the function score, no additional differences regarding significant associations were found.

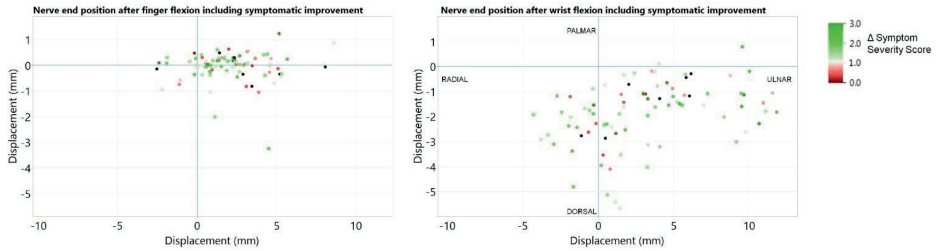


Figure 5. Nerve mobility and symptom score improvement. Cartesian plots showing the end position of the nerve after finger (left) and wrist (right) flexion. The color gradient indicates the change in SSS score between baseline and follow up measurement.

Table 3. Associations between baseline US and change in clinical outcome

	Δ Symptom Severity Score		Δ Functional Status Score	
Static parameters	**Estimate (β)**	**p-value (adjusted for baseline symptom score and sex)**	**Estimate (β)**	**p-value (adjusted for baseline function score and sex)**
Nerve CSA	-0.006	0.44	**-0.024**	**0.02**
Nerve perimeter	-0.042	0.70	**-0.049**	**0.02**
Nerve circularity	-0.011	0.49	-0.225	0.13
Dynamic parameters				
Finger flexion - RU displacement	0.024	0.27	0.060	0.15
Finger flexion - PD displacement	0.019	0.80	0.077	0.52
Finger flexion – overall displacement	0.036	0.17	0.075	0.13
Finger flexion – summed displacement	0.015	0.40	0.037	0.18
Wrist flexion - RU displacement	-0.003	0.76	0.011	0.55
Wrist flexion - PD displacement	0.023	0.42	0.039	0.28
Wrist flexion – overall displacement	0.001	0.94	0.031	0.22
Wrist flexion – summed displacement	0.004	0.79	0.030	0.23

CSA: Cross sectional area, RU: radio-ulnar, PD: palmar-dorsal.

Table 4. Associations between changes in US measures and changes in clinical outcome

	Δ Symptom Severity Score		Δ Functional Status Score	
Static parameters	**Estimate (β)**	**p-value (adjusted for baseline symptom score and sex)**	**Estimate (β)**	**p-value (adjusted for baseline function score and sex)**
Δ Nerve CSA	0.008	0.69	0.046	0.06
Δ Nerve perimeter	0.005	0.98	0.034	0.85
Δ Nerve circularity	0.001	0.98	0.025	0.43
Dynamic parameters				
Δ Finger flexion - RU displacement	**-0.035**	**0.03**	-0.036	0.12
Δ Finger flexion - PD displacement	-0.016	0.83	-0.111	0.41
Δ Finger flexion – overall displacement	**-0.050**	**0.01**	**-0.060**	**0.04**
Δ Finger flexion – summed displacement	-0.019	0.23	-0.035	0.11
Δ Wrist flexion - RU displacement	-0.012	0.22	-0.001	0.95
Δ Wrist flexion - PD displacement	-0.018	0.49	-0.043	0.40
Δ Wrist flexion – overall displacement	-0.016	0.32	-0.022	0.29
Δ Wrist flexion – summed displacement	0.000	0.99	0.000	0.99

CSA: Cross sectional area, RU: radio-ulnar, PD: palmar-dorsal.

Discussion

This study provides evidence that the median nerve gains mobility in a transverse plane in dorsal direction in CTS patients after surgical intervention as measured with US. The predictive value of baseline static parameters was found for functional improvement, but pre-operative dynamic parameters alone had no significant association with patient reported outcome. An increase in radio-ulnar displacement and higher overall nerve displacement after surgery during finger flexion showed an association with improved symptoms, but effect sizes are small and the dynamic measures were prone to noticeable physiological variation as reflected in moderate repeatability ICC values.

As our main outcome, we choose a patient reported outcome measure to keep the patients' perspective as a primary goal. It was decided to keep the two subscales of the Boston Questionnaire separated to allow conclusions on the different aspects of clinical improvement. Interestingly, it was on the FSS scale that we found a positive association between larger pre-surgical nerve size and increased improvement. We hypothesized that underlying pathological severity would be reflected in increased nerve size and would be directly correlated to clinical symptoms as tested in the SSS, but we found no significant association.

Dynamic imaging of the wrist has been done before, using imaging techniques including MRI and ultrasound. In non-CTS volunteers, Wang et al. showed that finger flexion alone does not induce a large nerve displacement, but that wrist flexion with extended fingers causes about 2.7 mm displacement[43]. We found similar magnitudes, but where they note a straight dorsal translation with wrist flexion, we found the nerve on average to move just as much in an ulnar direction. Whether this difference in angle is due to the CTS is difficult to say since their sample was relatively small (n=10), but their findings did illustrate that normally during wrist flexion, the nerve is inclined to move in dorsal direction, potentially to "escape" compression between the superficial flexor tendons and the transverse carpal ligament during tendon loading. A previous study from our group compared CTS patients with non-CTS controls (n=20 CTS cases, n=10 controls) and showed significantly inhibited dorsal translation for the cases[27], but this effect was lost in a follow-up study with a larger sample size (n=90 CTS cases, n=42 controls)[34]. Notably, van Doesburg et al. found no significant differences in nerve excursion during all four finger flexion between CTS and controls[44]. The fact that our study found a significant increase in dorsal translation after surgery only after wrist flexion supports the notion that the nerve regains mobility in that direction and is not induced by all finger or hand movements.

Our nerve mapping system was based on the hypothesis that after CTR, any increases in displacement would be visible by using the center point of the nerve as the start position. This poses a limitation if one of the effects of the surgery is not disruption of the SSCT, but more predominantly the repositioning of the nerve. In a study done by Nanno et al., the transverse displacement of the nerve was assessed using the hamate and trapezium to

define the absolute position of the nerve within the carpal tunnel. They report that after surgery, the nerve relocated to a more palmar position[35]. In contrast to our data, they find a decrease in nerve displacement after CTR during similar motions. This could in part be due to differences in location of the measurement as well as methodological timing as they acquired their imaging at the mid-level of the carpal tunnel at one month post-surgery.

So far, there has been conflicting evidence on the predictive potential of nerve CSA ranging between a negative association (smaller nerve is associated with better outcome)[45,46], no predictive value[11,47], and a positive association (larger nerve is associated with better outcome)[10,48]. Bland et al., like in our study, used the two isolated subscales as their main outcomes, and discussed that based on all the conflicting evidence it seems very unlikely that a single US parameter would provide enough evidence to support treatment counseling for the individual patient[47]. The other studies mentioned used a variety of outcomes, making direct comparisons challenging. Marschall et al. looked at a >25% increase in the overall result on the Boston questionnaire and their surgical sample contained only a small subset of the total sample (n=23)[10]. They found that a larger nerve size at baseline did not maintain its positive association at 12 months follow-up. El Miedany et al. found that a nerve CSA > 14mm^2 before surgery was associated with a poorer outcome and persistence of symptoms at 6 months[45]. Smaller nerve sizes were associated with a better response at 6 months based on a functional assessment and VAS score.

Possible explanations for the lack of association with post-surgical outcome and absence of differences in dynamic measurements at follow up could be the disconnection between US based features of the nerve and symptoms as reported by patients as well as timing of the assessments. Follow up was conducted at three months and although this might have been potentially too short term to find significant changes in the mobility of the nerve, we did see a significant decrease in nerve size, as reported by many before us. Pressure decrease in the carpal tunnel is found directly after surgery[49], indicating that neuro-edema caused by intra-carpal tunnel compression can start normalizing after a relatively short time, but it is currently not known what the time line is for any changes to occur in the fibrotic SSCT or what it would constitute. Assuming that either nerve size or the fibrosis play a pivotal role in the mechanism of the nerve being confined in its movements, it is perhaps the lack of understanding of the healing process that we see reflected in the results.

An important challenge with the dynamic recordings of the nerve as it translates to a different position is the large variation between patients and the lower ICC values between measurements of the same participants. After wrist motion we found movement in an almost 180° field, indicating that we are testing a heterogeneous variable. Notably, the test-retest reliability data came from healthy volunteers and indicate that even without surgical intervention, there is a physiological variation in the mobility of the nerve during our finger and hand movements. In an earlier study, a median nerve dynamic test-retest correlation of 0.82 had been reported[13], which is higher than what we found. However, these images were taken consecutively at the same time point, whereas our dataset had a several week time gap to mimic the timeline of the surgical cohort. Since the nerve moves

in a three dimensional plane, it would be interesting to see if the addition of a sagittal view would be a better proxy for nerve mobility. Intra-and inter cross correlations found in this study were excellent for both static as well as dynamic parameters. Possible explanation for the high values could be the absence of noise in the selected US parameters and the detailed protocol for the image analyses. Similarly high values have been reported before in healthy participants[12].

Limitations of the study include the focus on idiopathic CTS in patients undergoing any type of surgical release, limiting translatability to those with CTS not due to secondary causes or specific surgical approaches. Nerve size can decrease up to six months post-surgery[50], so this study can only conclude on short term effects. Additionally, no follow-up NCS data was collected, preventing any conclusions on biological recovery of the nerve. The change in nerve excursion was presented as the only US-based measure of surgery-induced changes, but future studies including healthy contralateral hands as controls should be done to draw any conclusions on how much the excursion restoration resembles a physiological state. Finally, conservative treatment including splinting and corticosteroid injections have a profound role in the management of mild-moderate CTS, but were excluded in this study and should be topic of future studies. With an expected wider spread in patient reported outcome, injection could yield a more complete picture to use as a primary regression outcome. Strengths of the study include the homogeneous nature of the patient sample, thorough analysis of the transverse mobility with the addition of a follow up ultrasound, the inclusion of reliability data, the high follow up rate and the large sample size.

Summary

In summary, after carpal tunnel release the median nerve mobility increases in CTS patients in a dominantly dorsal direction at a level proximal to the carpal tunnel. Not only on average but also at the extremes, our data indicates that there is no clear nerve movement threshold that evidently captures a subpopulation that is enriched in either clinically excellent or less excellent outcomes. There is considerable physiological variation in nerve displacement with finger and hand flexion which makes the measurement of small changes challenging. With high frequency ultrasound systems becoming more popular in daily clinical use, and increasing possibilities with image analyses, more opportunities for studies on small anatomical structures and differences are ahead of us. Also, with more studies prospectively collecting a wide array of data including pre-intervention patient characteristics, patient reported outcomes, longer term follow-up, and a better understanding of previously unknown confounders, a multivariable model serving a clinical advisory role for treatment seems possible. We anticipate that the added prognostic value of ultrasound, both statically and dynamically, acquired in multiple planes could add to such a model, rather than have added value as a single modality.

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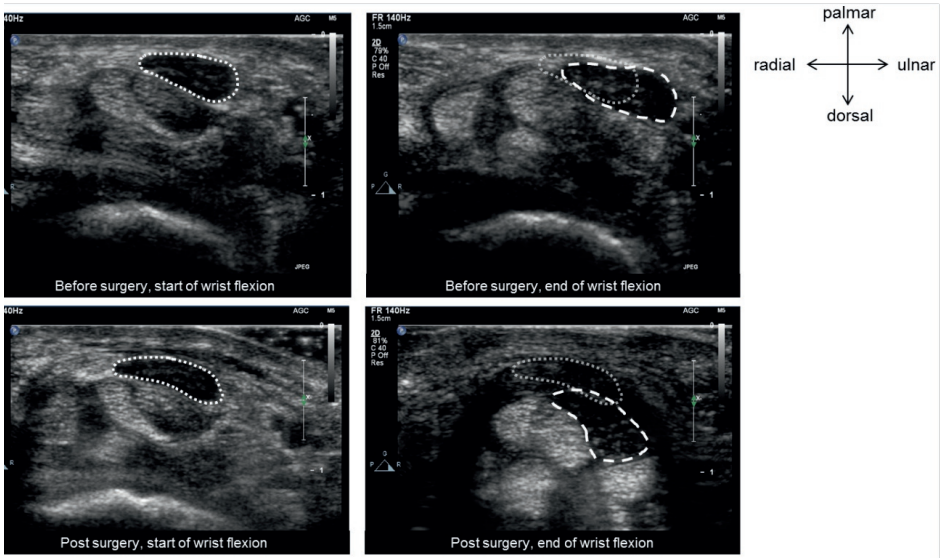
Supplements

App.A. Changes in BCTQ and US parameters after CTR

	Mean pre-operative (SD)		Mean post-operative (SD)		p-value	Mean difference (95% CI)
BCTQ						
Symptom Severity Scale	3.1 (0.7)		1.5 (0.5)		**<0.001**	-1.6 (-1.8, -1.5)
Functional Status Scale	2.6 (0.8)		1.6 (0.6)		**<0.001**	-1.1 (-1.3, -0.9)
Static US parameters						
Nerve CSA, mm^2	14.5 (4.2)		13.3 (3.8)		**<0.001**	-1.3 (-1.7, -0.8)
Nerve perimeter, mm	17.7 (2.6)		17.1 (2.8)		**0.009**	-0.6 (-1.0, -0.2)
Nerve circularity	1.7 (0.3)		1.8 (0.3)		0.41	0.03 (-0.04, 0.10)
Dynamic US parameters						
		Directionality		**Directionality**		
Finger flexion - RU displacement, mm	2.0 (2.2)	Ulnar	2.2 (2.3)	Ulnar	0.46	0.23 (-0.39, 0.86)
Finger flexion - PD displacement, mm	0.1 (0.6)	Dorsal	0.1 (0.4)	Palmar	0.12	0.12 (-0.03, 0.27)
Finger flexion – overall displacement, mm	2.4 (1.8)	Dorso-ulnar	2.6 (1.9)	Palmo-ulnar	0.35	0.25 (-0.28, 0.78)
Finger flexion – summed displacement, mm	4.8 (2.2)	NA	4.7 (2.3)	NA	0.94	-0.02 (-0.67, 0.62)
Wrist flexion - RU displacement, mm	3.1 (4.1)	Ulnar	2.7 (4.7)	Ulnar	0.32	-0.52 (-1.55, 0.51)
Wrist flexion - PD displacement, mm	1.9 (1.3)	Dorsal	2.4 (1.3)	Dorsal	**0.003**	0.54 (0.19, 0.90)
Wrist flexion – overall displacement, mm	4.9 (2.8)	Dorso-ulnar	5.4 (2.7)	Dorso-ulnar	0.20	0.47 (-0.25, 1.19)
Wrist flexion – summed displacement, mm	7.2 (2.5)	NA	8.0 (2.8)	NA	**0.038**	0.74 (0.04, 1.45)

Overall displacement is the excursion of the nerve defined as the difference in position between the start of the finger/wrist flex movement and the end. Summed displacement is defined as the summation of segmented nerve excursion over the full finger/wrist flex movement. Comparisons done using paired t-tests, absolute values pre and post-operative have been rounded to the nearest single decimal.
RU: radial-ulnar direction, PD: palmar-dorsal direction with the directionality of the displacements indicated in the next column. NA: Not applicable, since the summed parameter represents the summation of different vectors, no directionality is applicable. BCTQ: Boston Carpal Tunnel Questionnaire, SD: Standard deviation, CI: Confidence interval.

App.B. Example of ultrasound images and median nerve displacement during wrist flexion



App. B: Four transverse carpal tunnel ultrasound images from the same patient with moderately severe CTS. Upper row shows the start (left) and end (right) position of the nerve before and after wrist flexion with extended fingers. The row below shows two frames from de clip acquired three months after surgery. The median nerve is indicated with the interrupted line. In the post-operative images the change in end position to a more dorsal level can be seen.

7

CHAPTER 7.

Shear wave elastography of the median nerve: a mechanical study

Schrier, V. J.M.M., Lin, J., Gregory, A., Thoreson, A. R., Alizad, A., Amadio, P. C., & Fatemi, M. (2020).

Muscle Nerve, 61(6):826-833.

Abstract

Introduction
Shear wave elastography (SWE) shows promise in peripheral neuropathy evaluation but has potential limitations due to tissue size and heterogeneity. We tested SWE sensitivity to elasticity change and the effect of probe position in a median nerve cadaver model.

Methods
Ten specimens were used to measure median nerve elasticity under increasing loads using SWE and indentation. Measurements were compared using repeated-measures ANOVA.

Results
Indentation and SWE-based longitudinal nerve elasticity increased with tensile loading ($p<0.01$), showing a similar relationship. Acquisition in a transverse plane showed lower values compared to longitudinal measurements, mostly under higher loads ($p=0.03$), as did post-dissection elasticity ($p=0.02$). Elasticity did not change when measured proximal to the carpal tunnel.

Conclusion
Longitudinal SWE is sensitive to changes in median nerve elasticity. Measuring elasticity of peripheral nerves noninvasively could elucidate intra-neural pathology related to compression neuropathies, and proof to be of added value as a diagnostic or prognostic tool.

Introduction

Ultrasound elastography measures tissue elasticity by utilizing high-frequency sound waves to assess static or dynamic deformation behavior of tissue after a stimulus is applied[1]. This technique has been described most commonly in the context of isotropic tissues such as the liver and in neoplastic diseases[2] but is now becoming of more interest in anisotropic tissues, such as those in the musculoskeletal system[3,4].

Carpal tunnel syndrome (CTS) is a common peripheral compression neuropathy[5] and a frequent topic of investigation. Regardless of cause, a characteristic finding of CTS is increased pressure[6-11] in the carpal tunnel, typically associated with fibrosis of the connective tissue located around the nerve and flexor tendons[12-17]. As a result of the increase in pressure, the nerve is not only prone to ischemia, but the mechanical properties of the nerve are also altered over time[17]. Additionally, despite the natural capacity of peripheral nerves to adapt to longitudinal strain, median nerve mobility is diminished in CTS patients[18]. As there are currently no biomarkers that can reliably measure nerve elasticity changes or predict CTS treatment outcome[19], non-invasive measures of the disease are interesting topics to investigate. Elastic properties of nerves could also be used to evaluate changes in dynamic nerve response to physical therapy regimens that purport to mobilize peripheral nerves including nerve gliding exercises and nerve stretches, as often provided by hand therapists during conservative treatment.

Previous publications have described the potential of elastography for CTS, with the majority reporting increased nerve stiffness (ratios) in patients with CTS[20-32]; three research studies reported no significant difference[33-35]. In addition, ultrasound elastography has been used to assess carpal ligament stiffness[36,37], intra carpal tunnel pressure[38,39], the correlation between median nerve area and strain[40] and the difference in median nerve elasticity before and after carpal tunnel release[41]. These studies support the notion that the nerve's mechanical properties change in CTS patients, but the majority used *strain* elastography with manual compression in order to induce the tissue strain. This is a commonly used method, but has a major limitation: it does not provide absolute elasticity measurements but rather ratios and estimations due to the lack of consistent and reproducible stress in the soft tissue.

Currently, shear wave elastography (SWE) is the only clinically available method capable of obtaining quantitative elasticity measurements *in vivo*. Ultrasound (US) SWE is based on the propagation characteristics of waves induced by acoustic radiation force generated by an external vibrator or by focusing US beams to "push" the tissue. Previous studies have shown that SWE can detect both the differences in median nerve elasticity with increased nerve strain as induced by postural changes[42,43] as well as pathology-associated changes with sensitivity and specificity ranges between 76.4-96.6% and 62.5-100%, respectively[21,29-32].

SWE-measured elasticity is derived from wave propagation speed, but this estimation is mostly accurate for elastic, isotropic, homogenous, and large (compared to the shear wavelength) tissues like liver. Consequently, commercial US elastography systems are mathematically based on the assumption that measurements are carried out on tissues of such nature[44]. However, musculoskeletal tissues, tendons and nerves included, are often smaller than the wavelength, heterogeneous, anisotropic, and display a combination of elastic and viscous properties[45], and thus could decrease the validity of elasticity measurements. Specific to the application in nerves, probe orientation might affect elasticity values[46], exhibit boundary artefacts to their limited dimensions[47] (due to wave reflections at the nerve boundaries and acoustic impedance mismatching) and close proximity to highly reflecting structures such as tendons, bones, and ligaments[43,48]. Despite these confounding factors, it has been shown in tendons that the elastic modulus can still be used as an objective indicator of relative tissue elasticity as long as comparisons are done between similar tissue types, under similar conditions[49,50]; for nerves, this is currently less known. [4].

This is a mechanical study designed to assess the sensitivity of SWE to detect increases in nerve elasticity by testing the hypothesis that changes in tensile load will result in a different relative elasticity modulus. As a control, we used a compression indentation test directly on the median nerve. Secondarily, reflecting clinical relevance, the effect of probe orientation, position and interference of surrounding structures was tested.

Materials and Methods

Experimental Set up

This study was approved by our institutional biospecimens committee. Ten freshly frozen cadaveric forearms from our institution's anatomical bequest program were collected; donor medical charts were screened for any clinical indication of carpal tunnel syndrome, other upper extremity peripheral neuropathies, traumatic injuries, or surgical interventions in the wrist. Specimens were thawed out at least 24 hours in advance of testing and kept overnight at 4°C until the experiment. A single freeze-thaw cycle was applied, and all experimental data was gathered within a 24-hour timeframe. Previous publications have shown that biomechanical properties of soft tissues are preserved under similar freeze-thaw conditions[51,52]. Specimens were prepared with a circumferential incision 8 cm proximal to the distal wrist crease to expose the ulna and radius. The median nerve and flexor tendons were dissected, freeing the distal ends for at least 3 cm in length. Sutures were secured into the flexor tendons that were in close proximity to the median nerve (flexor superficialis tendons II-IV, profundus II and flexor pollicis longus). A circumferential suture was then placed in the median nerve to evenly distribute the weight of the tensile loads. The specimen was kept moist during the entire dissection by spraying exposed tissues with saline periodically. All cadaver hands

were fixated in supination using a custom-built platform that stabilized the ulna and radius with the wrist maintained in a neutral position.

Ultrasound and Shear Wave Elastography

US and shear wave measurements were performed using a GE Logiq E9 US machine equipped with a 2-8 MHz linear array 9LD transducer (GE Healthcare, Wauwatosa, WI) set on penetration mode with estimated target depth at 1.5 cm. In order to avoid compression artifacts, a custom-built probe holder was utilized, with ample coupling gel to keep the transducer stable and stationary during acquisition. B-mode imaging was first used to assess the presence of enough gel between the probe and the tissue and subsequently to identify the position, diameter and cross-sectional area of the nerve. Then, transverse and longitudinal images of the nerve were acquired just proximal to the carpal tunnel inlet (defined as the proximal margin of the transverse carpal ligament between the pisiform bone and scaphoid tubercle). In addition, in the longitudinal plane, images were taken approximately 5 cm proximal to the carpal tunnel. The order of SWE acquisition was alternated to avoid any effect of testing order on the results. Then, the transverse carpal ligament was divided, the connective tissue dissected, exposing the median nerve, and a coupler gel pad was placed on a layer of gel to allow direct SWE measurements in the longitudinal view. The exposed nerve measurements were used to assess the potential interference of connective and ligamentous tissue, as a gross reflection of surrounding tissue interference. At the start of SWE testing, 50 g weights were attached over individual low friction pulleys to the above-specified flexor tendons to maintain tension throughout the entire SWE portion of the experiment. To simulate a physiological change in median nerve elasticity, the nerve was connected to four different weights (10 g, 50 g, 100 g, 200 g), in increasing order, again including a pulley system to prevent friction and maintain tension (Suppl. Fig. 1A-B). After applying the 200 g weight, a 20 minute period was allowed for nerve tissue relaxation[53].

The Logiq E9 utilizes an acoustic radiation force mechanism; shear waves are created with multiple simultaneous push pulses aimed at targeted tissue[54] after which the propagation velocity of the resulting shear waves is detected in a rectangular field of view selected by the user. From the shear wave velocity, Young's modulus can be derived using the equation[55]:

$$E = 3C_s^2\rho$$

where E is Young's modulus, C_s is the speed of the shear wave and r is the tissue density, which was assumed to be 1075 kg/m^3 for the median nerve[56]. Note that this formula is an approximation of elasticity, limited by the assumption of isotropic, homogenous tissue and absence of boundary artefacts.

The preinstalled software on the GE Logiq allows for user-defined overlays of rectangular elastograms that display the tissue elasticity in a color scale. The size of the elastogram was adjusted during the first acquisitions so that at each position of interest, the entire median

nerve was centered in the rectangle. After that, elastogram size was kept constant for each specimen and probe orientation. Since the SWE measurements are dynamic, time was allowed to obtain a stable shear wave map before proceeding to acquire SWE images. Once a stable map was reached, a minimum of five SWE images was captured. Circular regions of interest (ROIs) were placed within the elastogram, with diameters adjusted to median nerve size. A single ROI was used for transverse images and double ROIs were placed for long-axis views over which averages were calculated (Suppl. Fig. 2).

Indentation protocol

The median nerve is surrounded by connective tissue in the carpal tunnel, but shows significant mobility with finger and wrist movement[18], indicating that the nerve can move and extend relatively freely. In order to test compressive elasticity in a non-destructive manner, while still allowing the addition of tensile load, an unconfined transverse indentation test was used as a control method. A similar approach has been used on tendons before[57]. The median nerve was dissected, isolated and terminated just before it branched to the digits for mechanical indentation testing.

A second circumferential anchoring suture was placed on the proximal nerve end with the total length of the sample spanning 6-8 cm. Unconfined compression testing was done using a 250 g capacity load cell (TA Instruments, New Castle, Delaware) with a 5.0 mm diameter plane indenter. The load cell was attached to an actuator, allowing for a controlled force increase (Suppl. Fig. 1C-D). Displacement was done at a constant rate of 0.05 mm/s with data collected at a sampling rate of 100 Hz. Loading was applied to the areas of the nerve corresponding to that assessed with SWE. Mechanical testing was performed while the nerve was subjected to the same four tensile loads, to impart changes in tissue elasticity. The indentation data were processed using MATLAB (R2016a, The MathWorks Inc., Natick, MA, 2000) and a stress-strain curve was used to abstract the slope with a linear fit between 0-15% strain, from now on referred to as the tangent modulus of the lower strain area (Suppl. Fig. 3). While this area is within the "toe region" and not the classic linear region of a stress-strain curve (thus not the direct equivalent of Young's modulus) the higher strains normally used to calculate Young's modulus are not achieved clinically. Therefore, for this study the lower strain region was selected as a better approximation of the elastic behavior of the nerve. During the first indentation experiment, two measurements were taken to establish repeatability, which was found to be high. Throughout indentation testing, the nerve was kept moist using a saline spray and moistened gauze. The total indentation time was roughly 20 minutes. All testing was conducted at room temperature (22°C).

Statistical Analysis

Summary statistics are presented as mean ± standard deviation. The mean elasticity values at each load for each method were calculated and visualized along with their corresponding standard error in order to gain insight into the shape of the relationship between tensile load and elasticity.

Two aims were tested in this study. First, we wanted to know whether increases in elasticity measured by SWE were proportional to those measured by indentation. Longitudinal SWE was selected as the main SWE approach to investigate, as this type of SWE application is the most common method in clinical and research use. With the same specimen measured multiple times, a repeated-measures analysis of variance (rm-ANOVA) was selected as the statistical model, with tensile load and the two acquisition methods (longitudinal SWE and indentation) as independent variables, specimen and nerve size as random factors, and elasticity as the dependent variable.

The second aim was to test the effect of different acquisition methods (SWE in the longitudinal plane, transverse plane, proximal location, and post-dissection) on SWE-measured elasticity. To do so, a second rm-ANOVA was performed, with US acquisition method as an independent variable, individual specimens and nerve cross-sectional area as random effects, and elasticity as the dependent variable. For both analyses, post-hoc paired comparisons, including Dunnett's procedure for multiple testing, were done using JMP (Version 14, SAS Institute Inc., Cary, NC, 1989-2007.). P-values equal to or less than 0.05 were considered significant. During indentation testing of the first specimen of the study (specimen no. 1), the specimen was noted to have dried out during the 200 g tensile load, resulting in an extremely high tangent modulus (>2 x IQR), which was not reflected in the shear wave data; this data point was subsequently excluded from further analyses[58]. During a later indentation test (specimen no. 7), we experienced technical difficulties with the actuator. After data review, it was found that data for this specimen also showed clear outlier behavior (>2x IQR); indentation data from this specimen was also removed from the analysis.

Results

A total of 10 specimens were tested; with the exception of one indentation data set and a single indentation data point noted above, all collected data were included in the analyses. The mean age at death was 75.3 years (range 63-93 years), with six male and four female specimens (Suppl. Table 1). The average nerve anterior-posterior diameter and cross-sectional area were 3.1±0.3 mm and 0.10±0.02 cm^2 respectively.

Elasticity and tensile load

Nerve indentation stress-strain curves were J-shaped, allowing measurement of the tangent moduli of the low strain region. Moduli generally increased with increasing tensile loads, but data showed larger variations with higher values (Fig. 1). The incremental effect between 10 and 50g was limited, as can be seen in the overlapping 95% confidence intervals. Indentation elasticity ranged from 3.4±1.2 kPa, 4.7±2.0 kPa, 8.4±5.1 kPa, 14.0±5.1 kPa for the 10 g, 50 g, 100 g, and 200 g loads, respectively. Both SWE and indentation moduli were plotted with a fitted linear regression model (Fig. 2). Analysis of variance indicated a significant effect of tensile load on both the indentation-based elasticity and longitudinal SWE ($p<0.001$) (Suppl. Table 2-A). The indentation elasticity was found to be lower for all loads compared to SWE ($p<0.001$). Subsequent interaction between measurement method and tensile load showed no significant differences ($p=0.22$), indicating that there is insufficient evidence to conclude that the relationship between tensile load and elasticity as measured by SWE is different from the relationship between tensile load and elasticity as measured by indentation.

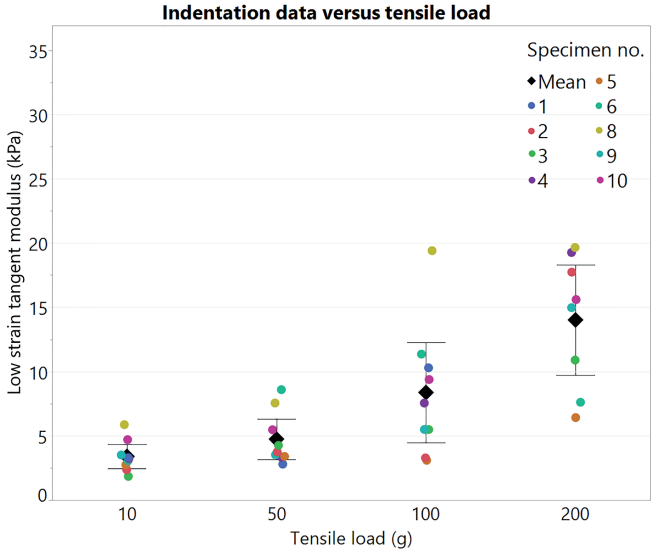


Figure 1: Data plot of indentation values for individual specimens with diamonds showing mean values and error bars representing the 95% confidence interval.

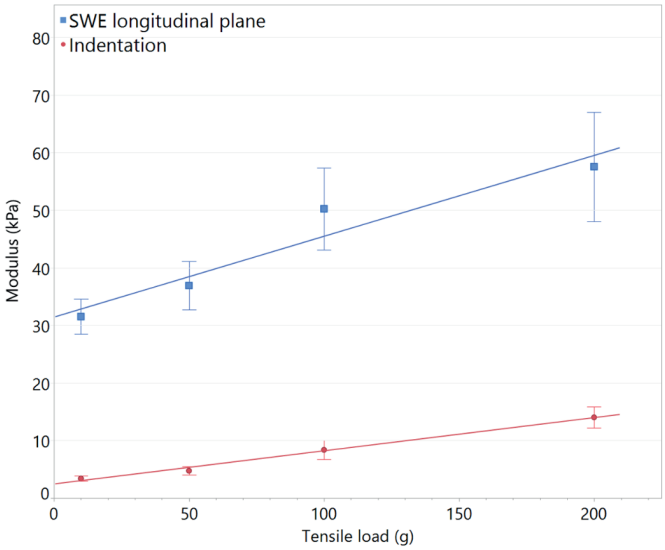


Figure 2: Relation between increased tensile load and measured moduli as acquired by longitudinally acquired SWE versus the indentation data. Data points show means with error bars representing the standard error of mean. Linear fit specifics: SWE= 31.43 + 0.1*tensile load, R2: 0.21, p<0.01; Indentation = 2.45 + 0.06*tensile load, R2: 0.56, p<0.01.

Changes in type of SWE acquisition

All SWE acquisitions are shown in Figure 3. Irrespective of the type of SWE acquisition, median nerve elasticity increased with added tensile load, and higher loads showed bigger inter-specimen variation. Except for the 200 g load (p=0.02), the analysis of variance showed no significant differences between the types of acquisitions under any of the tensile loads. Subsequent pairwise comparisons within the 200g group showed that elasticity values in the longitudinal plane (mean: 57.6 kPa, SD: 30.0 kPa) were significantly higher compared to the transverse plane (mean difference: 20.4 kPa, p=0.03) and the fully exposed nerve (mean difference: 22.2 kPa, p=0.02). All absolute SWE values are reported in Suppl. Table 2-B.

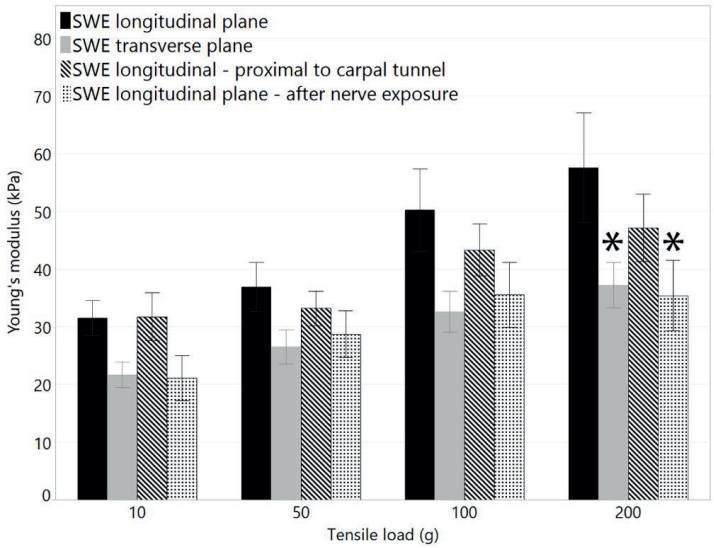


Figure 3: Mean longitudinal SWE data compared to transverse plane acquisition, proximal median nerve measurement, and after dissection. Error bars represent the standard error of mean. *p<0.05 compared to SWE longitudinal plane.

Discussion

This study tested two aims designed to elucidate the validity of SWE used in the context of carpal tunnel syndrome. We found that SWE measures nerve elasticity in a similar fashion to indentation testing, and that nerve elasticity measures are affected by probe orientation as well as characteristics of the tissue surrounding the nerve.

Measuring Nerve Elasticity

Previous publications have shown the applicability of the SWE assessment of the median nerve in CTS patients[21,29-32]. With reported values ranging between 8.2-43.7 kPa for the healthy controls, it seems that differences in the type of acquisition, study population, operator-dependencies, and other effects result in widely different absolute values. By looking at the interaction of tensile load with two different approaches to measure elasticity, we were able to see that despite absolute differences, SWE can detect increases in a similar fashion as direct indentation testing. For the values we reported (~30-60 kPa), we now have more substantial evidence that an increase in nerve elasticity measured clinically reflects an actual change in nerve physical properties. Our experimental SWE data matches the higher ranges reported in other studies, which also showed that inter-rater agreements are generally high (lowest reported ICC: 0.81)[21,29,30].

A previous study tested an *ex vivo* nerve under 300 g tensile load and showed a Young's modulus value of 13 kPa[59], which was comparable to our data but again much lower than elasticity measured clinically. This dissociation between lab-based and clinical

measures can in part be attributed to the differences in how data is acquired to calculate elasticity. Nerves have been characterized in previous studies to show slow stress increases at smaller strains followed by a gradual increase until a constant modulus is reached at >20% strain[53,60]; we found a similar profile in our indentation data. During SWE measurements, the elasticity of the nerve will be related to where in the toe region you are (may depend on wrist or finger position), as well as to unique characteristics of that nerve and that location including the relative amount of fibrous tissue in the nerve and perineural fibrosis. In addition, SWE relies on wave propagation which is affected by limited propagation space, fiber orientation, and adjacent tissues with differing material properties. Another key difference between the indentation and SWE methods is that the excitation mechanism and therefor frequency is different; with indentation, elasticity is measured as a result of incremental compression on a specified tissue region whereas in SWE, an acoustic radiation force is used to create waves that travel perpendicular to the excitation direction. Since elasticity values also depend on measurement frequencies[45,61], it was not surprising that we found significantly lower elasticity moduli with indentation.

The amount of variables involved underlines the difficulty of translating lab-based material properties to ultrasonographic findings. Interestingly, Ju *et al*. used a small cuff around rabbit sciatic nerves to apply circular compression which resulted in much higher elasticity values, around 67 kPa[60], suggesting that this approach might be more appropriate to directly relate to SWE data.

Effect of changing type of SWE acquisition

One of the sub-aims of this study was to look at the difference in probe orientation. Acquisition in both the transverse and longitudinal planes has been reported in the literature, so the fact that we found that transverse imaging generally leads to lower measurements is valuable information for protocol development and SWE standardization. Due to the small dimensions of peripheral nerves, transverse plane imaging is preferable, since its visualization is less dependent on getting an adequate section of the nerve in the plane. However, as was found before in muscles and tendons[62,63], we expected that the waves created during SWE, of which shear waves are one type, to propagate faster over the longitudinal axis of the nerve, parallel to the nerve fibers. This was confirmed in our data, most predominantly under higher tensile loads. With clinical data showing better reliability for longitudinal SWE than for transverse SWE[43], we recommend acquiring SWE data in a longitudinal plane, and to be mindful of the challenges with the validity of these values.

Our other sub-aim focused on seeing whether surrounding tissue might interfere with the longitudinal SWE signal, which we tested by creating an artificial carpal tunnel release scenario and by measuring in both a location close to and further proximal from the carpal bones. Neither variation showed more than a trend of lower elasticity measurements, but interestingly, after full exposure of the nerve, the elasticity decreased.

This could imply an interfering effect of the surrounding connective tissue and carpal ligament, but we cannot rule out a possible measurement artifact since we were measuring the nerve directly, with the coupling gel effectively functioning as a new anatomical border. If the first is the case, caution would be warranted in studies where post-surgical measurements are compared to pre-surgical results. We expected to have more interference closer to the distal radius and lunate bone, but the interference was relatively minor, indicating that at this level measurements can be performed reliably.

Our study has some limitations that could have influenced our results. We choose a cadaveric model instead of a phantom to ensure close anatomical and biomechanical resemblance to the human carpal tunnel. With the objective incremental tensile loading as a testing parameter came the necessity to dissect the nerve proximal to the carpal tunnel, which may limit the translation to the clinical situation. Further SWE validation is necessary to draw stronger conclusions on the effect of increasing disease severity, potentially with a pressure model instead of a longitudinal strain model. A potential approach could include using pressure points on the palm to induce intra-carpal tunnel pressure[64]. Our specimens were relatively old, and although medical records were screened, this does not fully rule out disease presence. Only one specimen had an enlarged nerve (no. 5), but nerve areas might have been overestimated since they were measured under a minimal amount of loading. Also, linear-elastic characteristics of the targeted nerve were assumed. As mentioned before, most of the soft tissues measured in the musculoskeletal field are not linear-elastic, resulting in a potential misrepresentation of the true elasticity. Our experimental set-up emphasized longitudinal nerve strain to induce elasticity changes, which we induced by using four standardized increasing weights which resulted in SWE values that were comparable to those reported in the literature. However, clinically, change in longitudinal nerve stretch is often quantified as strain. Lack of strain data is in fact a limitation of our present study. It is therefore important that future studies include the stain data. We found that both the SWE and the indentation data were prone to large variation between specimens, implying that there may be more value in repeated SWE measurements within the same person under different conditions as well as the usage of ratios, as proposed earlier[31]. Finally, although possibilities for alternative techniques are few, the indentation data was of limited value as a control. As discussed, elasticity values depend on strain and measurement frequencies[61], so we anticipated that the absolute values for indentation and SWE measurements would be substantially different. It has been proposed that in vivo, nerves are under a standard anatomical load and with additional (e.g. posture-induced) loading, the nerves show characteristics of a linear stress-strain curve, omitting the toe region phase[65]. Once excised, nerves do show a toe region typically seen in visco-elastic materials, but it is important to be aware of this discrepancy in mechanical behavior when comparing clinical and experimental situations. This is also one of the reasons why we chose to focus on the increase in elasticity in relation to tensile load rather than absolute measures.

Conclusion

In conclusion, SWE is a promising technique to assess changes in biomechanical properties of the median nerve. Clinical studies will be needed to assess diagnostic and prognostic usefulness, but our study supports the notion that SWE measurements reflect true increases in elasticity and that longitudinal image acquisition is the preferred plane of imaging. Any clinical application should be done with a standardized protocol and with due consideration of the limitations of measuring shear waves in anisotropic tissues.

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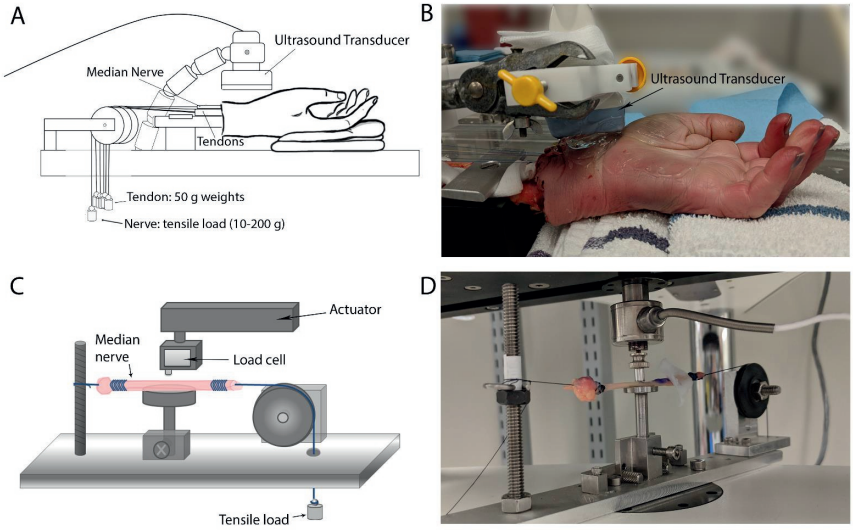
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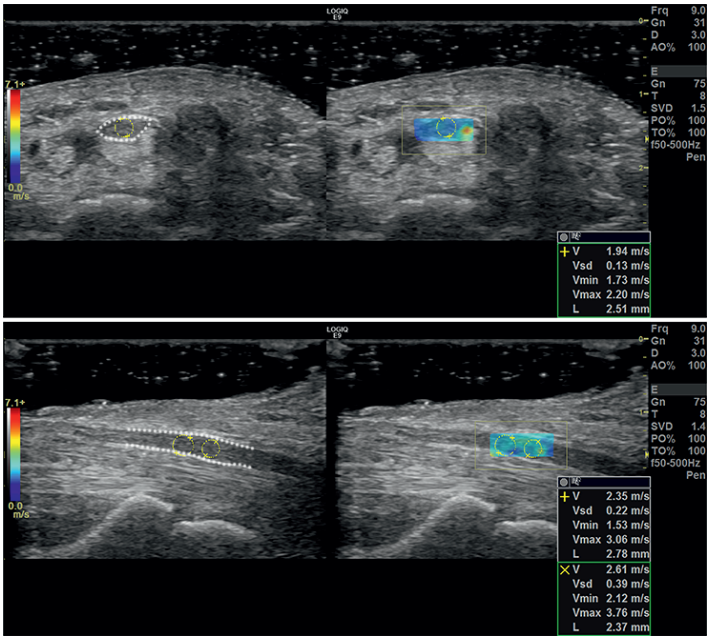
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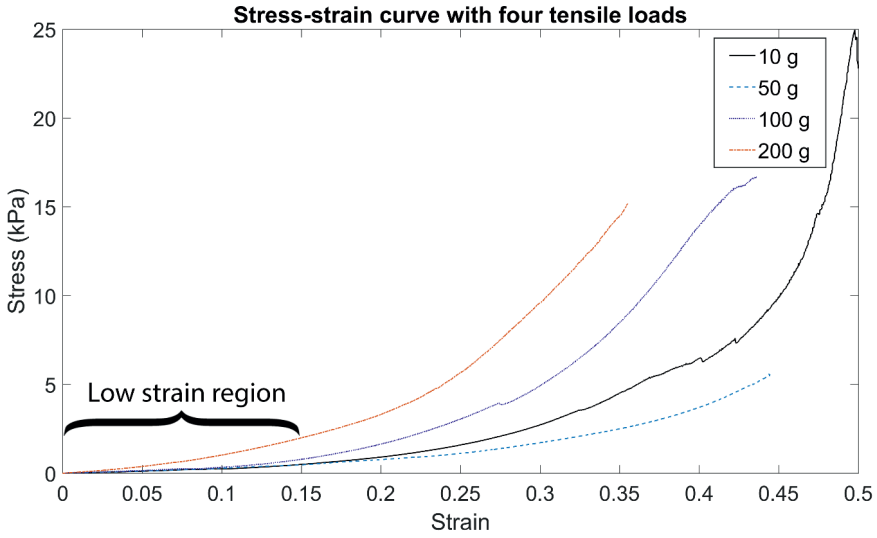
Supplements



Suppl. Fig. 1: A-B) Representation of experimental set up during the SWE testing. A) Schematic version of the total set-up. The median nerve was attached to increasing tensile loads. The five flexor tendons in closest approximation to the nerve were attached to 50g weights. B) A surplus of gel was used to prevent excessive pressure. C-D) The set-up during the indentation testing on the *ex vivo* nerve.



Suppl. Fig. 2: Example of shear wave ultrasound acquisition frames (specimen nr 8) at the transverse (above) and longitudinal planes (below). The median nerve is visible (white interrupted line) at the level of the carpal tunnel inlet. A single region of interest was selected for the transverse and two for the longitudinal images (yellow circles) with at least five acquisitions per tensile load. Data are shown in m/s but were converted to kPa in the analyses.



Suppl. Fig. 3: A set of stress-strain curves as measured on one of the nerves with uncompressed transverse indentation while under 10, 50, 100 or 200 g tensile load. The tangent modulus was measured between 0-15% strain.

Suppl. Table 1 Descriptive statistics for ten specimens.

Specimen	Sex	Age (yr)	Anterior-posterior nerve diameter (mm)	Nerve cross- sectional area (cm^2)
1	Male	74	3.4	0.11
2	Male	63	2.8	0.09
3	Male	63	2.8	0.10
4	Female	69	3.7	0.08
5	Female	80	3.3	0.15
6	Male	87	2.9	0.07
7*	Female	77	3.0	0.09
8	Male	82	2.9	0.12
9	Female	65	3.0	0.13
10	Male	93	3.4	0.10
Average		75.3	3.1	0.10
SD		10.4	0.3	0.02

*indentation data from this specimen excluded from analyses
SD: Standard deviation

Suppl. Table 2 A. Summary of analysis of elasticity by load and acquisition method

Factor	Level	LS means (SE)	p-value*	p-value‡
Method	Indentation	4.18 (5.1)	*<0.001*	
	SWE - longitudinal	31.5 (4.8)		
Load	10g	17.9 (3.9)		reference
	50g	21.2 (3.9)	*<0.001*	0.777
	100g	29.7 (3.9)		*0.022*
	200g	35.9 (4.0)		*<0.001*
Method * Load	Indentation, 10g	4.2 (5.1)		
(interaction)	Indentation, 50g	5.5 (5.1)		
	Indentation, 100g	9.1 (5.1)		
	Indentation, 200g	14.3 (5.3)	0.216	NA
	SWE - longitudinal, 10g	31.5 (4.8)		
	SWE - longitudinal, 50g	36.9 (4.8)		
	SWE - longitudinal, 100g	50.3 (4.8)		
	SWE - longitudinal, 200g	57.6 (4.8)		

LS: least squares

* p-value from global F-test for fixed effect of acquisition method and load

‡ Pairwise comparisons adjusted using Dunnett's procedure to compare all levels to reference

Suppl. Table 2 B. Summary of analysis of elasticity by SWE acquisition method

Load		Method	N	Mean (SD)	p-value*	p-value‡
10g	1	SWE - Longitudinal	10	31.5 (9.7)	0.050	reference
	2	SWE - Transverse	10	21.7 (7.0)		0.130
	3	SWE - Longitudinal-proximal	10	31.7 (13.0)		1.000
	4	SWE - Longitudinal-exposed	10	21.1 (12.4)		0.104
50g	1	SWE - Longitudinal	10	36.9 (13.4)	0.164	NA
	2	SWE - Transverse	10	26.5 (9.4)		
	3	SWE - Longitudinal-proximal	10	33.2 (9.4)		
	4	SWE - Longitudinal-exposed	10	28.7 (12.8)		
100g	1	SWE - Longitudinal	10	50.3 (22.5)	0.078	NA
	2	SWE - Transverse	10	32.6 (11.3)		
	3	SWE - Longitudinal-proximal	10	43.3 (14.2)		
	4	SWE - Longitudinal-exposed	10	35.5 (17.7)		
200g	1	SWE - Longitudinal	10	57.6 (30.0)	*0.021*	reference
	2	SWE - Transverse	10	37.2 (12.4)		0.031
	3	SWE - Longitudinal-proximal	10	47.1 (18.6)		0.387
	4	SWE - Longitudinal-exposed	9	35.4 (18.2)		0.016

* p-value is from global F-test for fixed effect of acquisition method

‡ Pairwise comparisons adjusted using Dunnett's procedure to compare all levels to reference

PART III

8

CHAPTER 8.

Ultrasound-guided hydrodissection with corticosteroid injection in the treatment of carpal tunnel syndrome: A pilot study

Schrier, V.J.M.M., Brault, J.S., Amadio, P.C. (2020)

J. Ultrasound in Medicine [epub ahead of print]

Abstract

Objectives
Corticosteroid injections can provide (temporary) relief in patients with mild to moderate carpal tunnel syndrome (CTS). Hydrodissection as part of an injection has been associated with positive clinical outcomes but data for CTS so far has been scarce. This study is designed to assess patient tolerance and secondarily provide pilot data on the added effect of hydrodissection.

Methods
Twenty CTS patients were randomized to an ultrasound-guided betamethasone injection with hydrodissection (5 mL) or without (2 mL). Patient tolerance was assessed directly after intervention and patient-reported outcome after 4 and 24 weeks. Intra-group data were compared using Wilcoxon Signed Rank and inter-group with Wilcoxon rank-sum tests.

Results
Tolerance and pain scores did not differ between the two groups. Symptom scores decreased in both groups, but to a lesser extent in the hydrodissection group with a mean difference of -0.8 versus -1.5 in the control group at four weeks ($p=0.02$). At 6 months, this difference was no longer present ($p=0.81$). No statistically significant differences were found between the hydrodissection and control groups in the function or pain scores at follow-up at either time point.

Conclusion
After injection, both symptomatic and functional scores improved, but the hydrodissected group did not show additional improvement. Data presented can be used to support larger studies to assess the value of hydrodissection in CTS management.

Introduction

Carpal tunnel syndrome (CTS) is a compression neuropathy affecting the median nerve with a life-time prevalence of roughly 8% in the adult population[1]. There is strong evidence to believe that one or multiple corticosteroid injection(s) can be used to effectively treat CTS symptoms in mild to moderate cases[2], but their added value remains controversial due to the temporary nature of the symptomatic relief[3]. This requires a significant percentage of CTS patients to either repeat the injection or convert to a surgical intervention[4,5]. Although research has been done comparing different types of injections, a current challenge with corticosteroid injections remains that their exact effect is unknown. Increased carpal tunnel pressure is often described in patients with CTS[6-11] as well as histological non-inflammatory fibrosis of the connective tissue inside the carpal tunnel[12-19]. Initially, using corticosteroids was thought to decrease intra carpal tunnel inflammation, however the fact that the injections only work for a limited time period indicates that they work at least on other levels as well[20].

In part due to the nerve swelling and/or the surrounding fibrosis, ultrasound (US) research has shown that both tendon and nerve transverse mobility is inhibited in CTS patients[21-24]. Normally, the median nerve would move in dorsal direction as a response to digital flexor loading, but in CTS patients this compensatory movement is limited allowing direct contact between the transverse ligament and the nerve. Possible fixation between the nerve and the surrounding structures could also lead to longitudinal shear stress and damage to the epineurium[25].

A relatively new technique to combat median nerve entrapment is (ultrasound-guided) hydrodissection[26-28]. Hydrodissection itself is not new and refers to using pressurized saline solutions during surgical procedures to create surgical planes and to realize adhesiolysis[29]. This technique differs from a regular US-guided injection in that now the volume of the injectate is used to create a fluid plane around the peripheral nerve and to use this blunt dissection to locally disrupt fibrotic tissue[30,31]. In the context of CTS, the interference of fibrotic connective tissue could allow the nerve to regain its physiological mobility. So far no comparisons between traditional CTS injections with and without hydrodissection have been published, nor data on patient satisfaction or treatment failure.

This study aims to provide preliminary data to estimate the effect of hydrodissection in a randomized double-blinded pilot. Foremost, this study is designed to test patient tolerance and secondarily, we aimed to study patient-reported clinical outcomes and treatment failure. Finally, nerve mobility will be assessed as a possible mechanistic explanation of hydrodissection.

Materials and Methods

Study design

This was a double-blinded, prospective, randomized parallel pilot study with a 1:1 allocation ratio. The study (CT.gov identifier NCT03427983) was approved by our Institutional Review Board (#17-009840). Written informed consent was obtained from all participants in accordance with the Declaration of Helsinki. A total of twenty patients (twenty hands) were recruited at a tertiary treatment center with a specialized hand clinic between May 2018 and March 2019. The CTS diagnosis and indication for injection were determined by the attending hand physicians based on symptoms at presentation, clinical tests and supporting electro-diagnostic tests. Only patients with a severity of moderate or less were included in this study[32]. Patients were also excluded if there was any indication of non-idiopathic CTS or a previous surgical intervention on the wrist or corticosteroid injection. A description of the inclusion and exclusion criteria has been added in Appendix A. In case of bilateral CTS only the hand with the most severe symptoms was included.

Study flow

The study flow is shown in Fig. 1. After recruitment, patients completed two site visits (baseline, 4 weeks post injection) and one telephone follow-up (24 weeks post injection). During the baseline assessment patients were asked to complete the patient-reported outcome measures and underwent a dynamic ultrasound assessment. Then, participants were randomized to either one of two research arms: (1) US in-plane injection with corticosteroid and hydrodissection or (2) US out-of-plane injection with corticosteroid without hydrodissection. Randomization was done using code words in a permuted block design (block size of 5) as designed by an external party using REDCap[33]. Participants were blinded to the allocation and the ultrasound screen was positioned away during the procedure. Study staff responsible for the outcome measures were blinded until the full analysis was established. In order to properly execute the intervention, the clinician performing the injection could not be blinded (JSB).

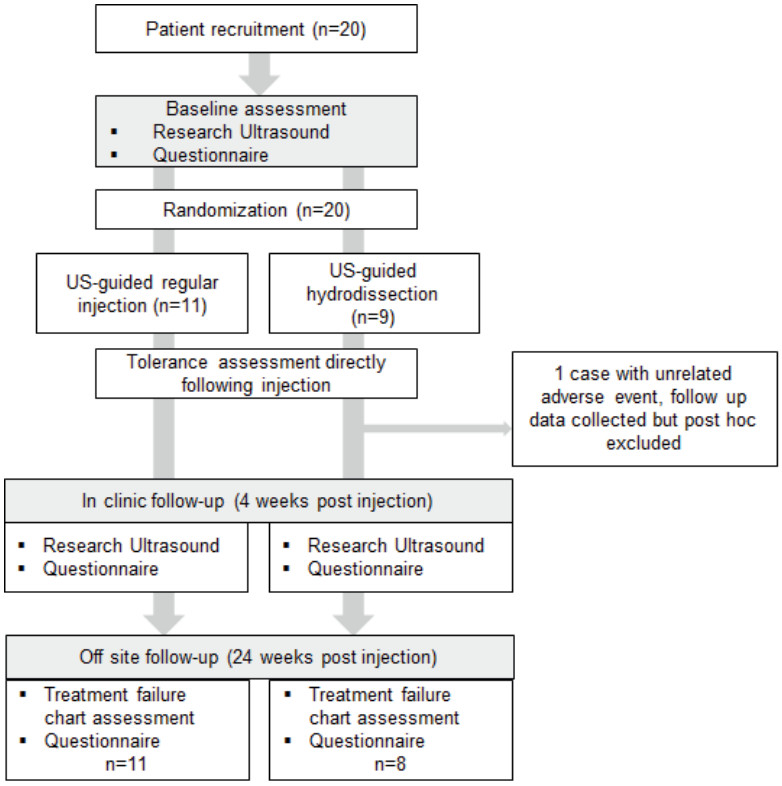


Figure 1 Study flow: Study flow from recruitment to final follow-up at 24 weeks post injection. Recruitment, randomization and injection were all done on the same day. All recruited patients stayed in the study until the final follow-up, but patient-reported data of 1 case in the hydrodissection group had to be excluded due to an unrelated event potentially affecting outcome.

Intervention

A SonoSite X-Porte ultrasound system (Sonosite, Bothell, WA, USA) with an L15-7io linear array transducer was used. The hydrodissection was accomplished with a total volume of 5 mL (1 mL betamethasone (6mg) and, 1 mL 1% lidocaine and 3 mL saline) based on a study demonstrating complete nerve hydrodissection throughout the carpal tunnel with this volume[34]. The control subjects received a total of 2 mL (1 mL betamethasone (6mg) and 1 mL 1% lidocaine). The ulnar in-plane approach has been described previously[30]. In short, under ultrasound guidance, injectate deposits are made lateral, palmar, and dorsal to the median nerve, creating a circumferential fluid plane along the length of the nerve (Fig. 2). The control injection followed the same trajectory except the injectate was placed as a singular deposit ulnar to the median nerve without targeted separation from the transverse ligament[35].

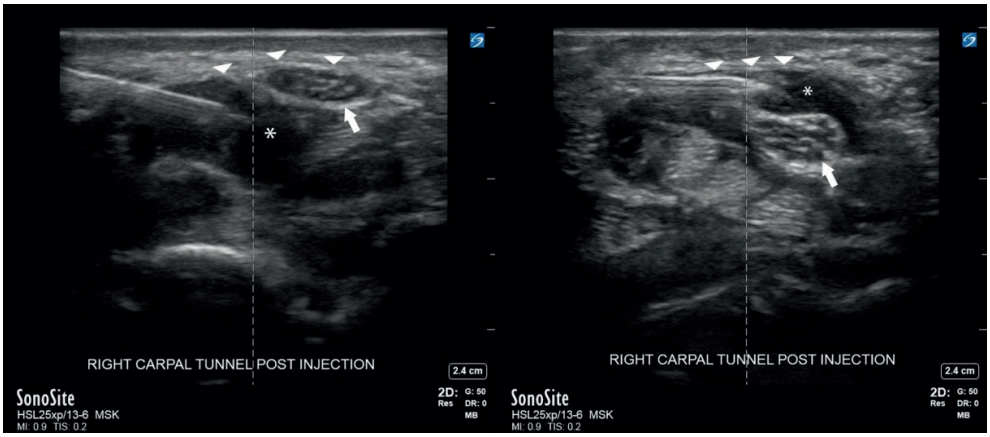


Figure 2 Ultrasound example: Transverse ultrasound images showing hydrodissection dorsal to (left) and volar to (right) the median nerve (arrow). The transverse carpal ligament can be seen superficial to the nerve (arrow heads). In the hydrodissected group, a total of 5 mL was used to separate the nerve from the ligament and the flexor tendons. Hydrodissected planes are indicated with *.

Outcomes

Primary outcome was patient satisfaction as determined by questions concerning the level of pain during the procedure and the likelihood of undergoing the same procedure again (scales 1-10; lower values equal better outcome). In addition, the survey included a multiple choice question on how their injection experience related to what they had expected (much less, somewhat less, about the same, somewhat more, much more comfortable than expected). As secondary outcomes, both patient-reported data as well as ultrasound data was collected. Patients completed the Boston questionnaire for carpal tunnel syndrome (BQCTS) which consists of two subscales emphasizing symptoms and function[36]. In addition, visual analogue scores (VAS) on pain and interference in day to day life (Likert scale 1-10) were completed. During the follow-up visits, patient-reported improvement and treatment failure, defined as a second injection or conversion to surgery, were completed and the latter was confirmed through medical chart review. If patients had a secondary intervention, patient-reported data at the follow-up time point(s) after the intervention date was excluded. Ultrasound data included both static (nerve cross-sectional area) and dynamic nerve characteristics which were all recorded at the proximal carpal tunnel inlet. Dynamic nerve movement was defined as the overall displacement during either four finger flexion (from neutral to maximum flexion) or wrist flexion (hand volar flexion with extended fingers). Images were corrected for probe and whole carpal tunnel movement. Nerve size was based on an average of two measurements. Ultrasound data were analyzed using specialized image processing software (Analyze, 12.1 Software, Biomedical Imaging Resource, Mayo Clinic, Rochester, MN).

Statistical Analyses

Analyses were done using JMP (Version 13. SAS Institute Inc., Cary, NC, 1989-2007.). Continuous data are presented as means including standard deviations. Categorical data is shown as count data and proportion. Continuous baseline characteristics were compared with two sample t-tests for normally distributed data and Wilcoxon Rank Sum tests for non-normally distributed data. Categorical data were tested using Chi-square or Fisher's exact test based on the distribution. Tolerance and pain differences between the groups were compared using a Wilcoxon Rank Sums tests. Clinical outcomes defined by the BQCTS, the VAS, and patient-reported improvement as well as the ultrasonographic data are (quasi)-quantitative and intended as. Sample size was based on expectations that potential tolerance differences would be identifiable while still providing enough data to support a power calculation and to identify trends regarding change in nerve dynamics. Preliminary analyses presented here were done using Wilcoxon rank-sum and Wilcoxon-signed rank tests with a preset type I error threshold of 0.05. For intra- and intergroup comparisons a post-hoc Bonferroni-correction was applied.

Results

Data quality

All twenty participants who enrolled in the study finished both follow-up time points. None reported intolerance for either of the injection types and no adverse events directly relatable to the intervention were reported. In one case, the randomization failed, leading to an uneven distribution over the groups. In another case, the patient complained of wrist and hand pain at the 4 week follow-up time point. This was attributed to an unrelated IV placement procedure, but results could not be used and were eliminated (hydrodissection group).

Baseline results

Demographics and disease characteristics are shown in Table 1. Eleven patients entered the control group, nine patients underwent hydrodissection. The hydrodissection group tended to be younger, mostly due to two patients aged 33 and 34, but did not differ significantly from the control group in age, sex, BMI, disease duration, dominance, or presence of bilateral symptoms. In both groups, the majority of patients had moderate CTS based on the electrophysiological severity (44% and 45% for the control and hydrodissection-group respectively). Baseline BQCTS and pain scores were comparable.

Table 1. Patient demographics and baseline characteristics

	Hydrodissection (n=9)	Control (n=11)	
Patient demographics			
Age, years (median, 25th-75th percentile [range])	47, 37-61 [33-68]	59, 55-66 [48-76]	p=0.09
Sex, female (n, %)	6 (67)	10 (91)	p=0.28
BMI (median, 25th-75th percentile [range])	32.3, 22.7-39.2 [21.6-51.5]	26.8, 21.1-35.1 [17.4-44.9]	p=0.54
Duration in months (median, 25th-75th percentile [range])	12, 6.5-14 [3-24]	12, 7-96 [5-180]	p=0.67
Dominance, right (n, %)	8 (90)	10 (91)	p=0.88
Dominant hand included, yes (n, %)	7 (78)	8 (73)	p=0.80
Presence of bilateral symptoms, yes (n, %)	7 (78)	8 (73)	p=0.80
Neuroconductive severity (n, %)			
Normal	3 (33)	2 (18)	p=1.00
Mild	2 (22)	3 (27)	
Moderate	4 (44)	5 (45)	
Missing	0 (0)	2 (18)	
PROM (mean ± SD [range])			
BQCTS-Symptom score	2.6 ± 1.0 [1.6-4.8]	2.8 ± 0.6 [1.8-3.6]	p=0.45
BQCTS-Function score	2.1 ± 0.9 [1.4-3.8]	2.1 ± 0.7 [1.0-3.5]	p=0.98
VAS – pain	4.0 ± 2.6 [1-8]	2.8 ± 2.3 [1-8]	p=0.31
VAS – interference	4.0 ± 2.5 [1-8]	3.2 ± 1.9 [1-7]	p=0.43
Ultrasound measures (mean ± SD [range])			
Nerve CSA, mm^2	11.7 ± 2.2 [9.6-15.9]	13.5 ± 2.7 [9.8-17.3]	p=0.13
Displacement with *finger* flexion, mm	2.2 ± 1.1 [0.9-4.2]	2.5 ± 1.9 [0.5-6.1]	p=0.67
Displacement with *hand* flexion, mm	3.8 ± 2.2 [1.3-8.2]	2.9 ± 1.4 [1.1-5.5]	p=0.27

SD: standard deviation, PROM: patient reported outcome measure, BQCTS: Boston questionnaire for carpal tunnel syndrome, VAS: Visual analogue scale, CSA: Cross sectional area. Statistical comparisons done using parametric and non-parametric tests based on data distribution.

Primary outcomes

Directly after the injection, pain scores were marginally higher in the hydrodissection group with a mean of 2.4±0.9 versus 1.9±1.2 in the control group, but this difference was not statistically significant (mean difference 0.5, p=0.19). All participants in the control group had selected the optimal score in reply to "Likelihood to undergo the procedure again", whereas one patient scored a "2" in the hydrodissection group (p=0.31). Expectations were met in equal fashions in both groups with 44% and 55% of participants indicating that the procedure was "much more comfortable than I had expected" for the hydrodissection and control groups respectively (p=0.90). In neither of the groups did any of the patients report a "(much) worse comfort than expected" result.

Secondary outcomes

None of the participants had had any additional treatment in the form of a second injection or conversion to surgery at the 4 week time point, however, 4 (20%) of the 20 patients did have a surgical release by the 6 month time point, two from each group. BQCTS symptom scores decreased in both groups, but to a lesser extent in the hydrodissection group with a mean difference of -0.8 (95%CI: -1.2; -0.4) versus -1.5 (-95%CI: 2.0; -1.1) in the control group at four weeks. This difference was statistically significant (p=0.02; Table 2), but after 6 months, this was no longer present (p=0.81). No statistically significant differences were found between the hydrodissection and control groups in the function or pain scores at follow-up at either time point. More patients from the control group responded with the most positive outcome in the patient-reported improvement (91% versus 50% in the hydrodissection group) at four weeks, but at 24 weeks, patients from the control group descended to lower scores (Appendix B). Over both time points, the proportional distribution showed no significant differences between the treatment groups (p=0.49). In terms of ultrasound parameters, nerve size decreased in both groups after 4 weeks. Nerve excursion marginally changed with finger flexion after injection, with an increase in the hydrodissection group and a decrease in the control, whereas conversely the hand flexion induced movement showed an increase in the control (+1.0 mm, p=0.32) and a decrease in the hydrodissected patients (-1.1 mm, p=0.25). No significant differences between the groups were found based on the mean differences (Table 3).

Table 2. Changes in Clinical Outcome at Follow-up

	Hydrodissection			Control			
	Mean ± SD	**Mean difference (95% CI)**	**Intra-group***	**Mean ± SD**	**Mean difference (95% CI)**	**Intra-group***	**Inter-group†**
BQCTS-Symptom score							
Baseline	2.6 ± 1.0			2.8 ± 0.6			
4 weeks	1.8 ± 1.1	-0.8 (-1.2; -0.4)	**p=0.008**	1.3 ± 0.3	-1.5 (-2.0; -1.1)	**p=0.001**	**p=0.02**
24 weeks‡	1.8 ± 0.4	-0.5 (-1.1; 0.1)	p=0.09	2.3 ± 0.7	-0.6 (-1.3; 0.1)	p=0.20	p=0.81
BQCTS-Function score							
Baseline	2.1 ± 0.9			2.1 ± 0.7			
4 weeks	1.5 ± 0.8	-0.7 (-1.2; -0.1)	**p=0.008**	1.3 ± 0.2	-0.8 (-1.3; -0.3)	**p=0.004**	p=0.59
24 weeks‡	1.4 ± 0.3	-0.5 (-1.4; 0.4)	p=0.31	1.6 ± 0.5	-0.4 (-0.9; 0.2)	p=0.19	p=0.86
VAS – pain							
Baseline	4.0 ± 2.6			2.8 ± 2.3			
4 weeks	2.4 ± 2.5	-1.9 (-3.9; 0.1)	p=0.03	1.2 ± 0.4	-1.6 (-3.0, -0.3)	**p=0.02**	p=0.90
24 weeks‡	1.8 ± 0.8	-1.7 (-4.0; 0.7)	p=0.13	2.6 ± 1.9	0 (-2.4; 2.4)	p=0.81	p=0.16
VAS – interference							
Baseline	4.2 ± 2.5			3.2 ± 1.9			
4 weeks	2.2 ± 1.8	-2.0 (-3.7; -0.3)	p=0.06	1.6 ± 1.0	-1.5 (-2.9; -0.2)	p=0.04	p=0.58
24 weeks‡	1.8 ± 1.2	-1.7 (-3.8; 0.5)	p=0.25	2.2 ± 1.1	-1.0 (-2.6; 0.6)	p=0.25	p=0.53

SD: standard deviation, PROM: patient reported outcome measure, BQCTS: Boston questionnaire for carpal tunnel syndrome, VAS: Visual analogue scale. CI: Confidence interval. Mean differences shown are in respect to baseline value.

* Wilcoxon Signed Rank tests with statistical significance reached at p<0.025

† Wilcoxon Rank Sum tests with statistical significance reached at p<0.025

‡ 24 Weeks follow-up data excludes those with additional treatment (n=4), so mean difference does not exactly match visible baseline-24wk follow-up difference.

Table 3. Changes in Ultrasound Parameters at Follow-up

	Hydrodissection			Control			
	Mean ± SD	**Mean difference (95% CI)**	**Intra-group***	**Mean ± SD**	**Mean difference (95% CI)**	**Intra-group***	**Inter-group†**
Nerve CSA, mm²							
Baseline	11.7 ± 2.2			13.5 ± 2.7			
4 weeks	10.9 ± 1.9	-0.8 (-2.1; 0.4)	p=0.25	12.9 ± 3.0	-0.6 (-1.7; 0.5)	p=0.10	p=0.82
Displacement with finger flexion, mm							
Baseline	2.2 ± 1.1			2.5 ± 1.9			
4 weeks	2.5 ± 1.4	0.2 (-1.1; 1.6)	p=0.91	2.3 ± 1.7	-0.2 (-1.1; 0.7)	p=0.70	p=1.00
Displacement with hand flexion, mm							
Baseline	3.8 ± 2.2			2.9 ± 1.4			
4 weeks	2.8 ± 1.4	-1.1 (-3.0; 0.8)	p=0.25	3.8 ± 2.3	1.0 (-0.6; 2.7)	p=0.32	p=0.15

SD: standard deviation, CSA: Cross sectional area. CI: Confidence interval. Mean differences shown are in respect to baseline value.
* Wilcoxon Signed Rank tests with statistical significance reached at p<0.025
† Wilcoxon Rank Sum tests with statistical significance reached at p<0.025

Discussion

In this blinded, randomized clinical pilot, we found that patient tolerance and satisfaction with hydrodissection were comparable to the lower volume control injection, suggesting that this technique can be used with comparable patient satisfaction in the treatment of mild to moderate CTS. However, based on this preliminary data, we found no evidence of a superior effect of a corticosteroid injection delivered with hydrodissection at 1 and 6 months follow-up.

One of the most interesting findings in this study was that the control group had small but statistically significantly better symptomatic relief after 4 weeks compared to the hydrodissected group. We had hypothesized that hydrodissection would show superior results based on findings from previous studies[34,37-39], but this group showed a less favorable clinical symptom score. The notion that increased injectate volumes add to the already increased pressure in the carpal tunnel seems unlikely, especially since as soon as 1 hour after the injection, the fluid has already been absorbed[38]. Potentially, during the hydrodissection procedure, by placing the fluid very close to the nerve, it may mildly and transiently irritate the nerve in some way. Armstrong et al. described using 2 mL of injectate with 6 mg of betamethasone in a placebo-controlled study and found a mean symptom difference of -0.8 which is about twice as low as the value of our control group and equal to our hydrodissection group[40]. Based on this comparison, and additional comparable studies[41,42], we ultimately concluded that our sample responded extremely well to the injection treatment in general and that therefore the hydrodissection is not significantly inferior but rather that there is no evidence implying superiority.

Pilot data presented here was intended to support power analyses for future studies; assuming an α of 0.05, a power of 0.80, and a clinically important difference[43] reached at a mean difference in symptom severity score of -1.0 at 24 weeks, a trial with an estimated minimum of 31 participants in each group would be necessary. In terms of generalizability, we would like to note that this study was conducted on a sample with only mild to moderate idiopathic CTS cases.

Previous studies looking at efficacy of hydrodissection either did not address CTS[44,45] or did not have a clinically relevant control group[35,46]. Malone et al. reported a case series of 44 wrists of CTS patients where they used a combination of US-guided hydrodissection with fenestration and showed that in the majority of the cases (28 wrists) all symptoms were resolved at their first follow-up (average of 8 months)[46]. Using a similar hydrodissection technique, another study focused on an elderly population comparing triamcinolone with a saline injection (total volume of injectate 3 mL in each group). They found overall results comparable to ours, namely significant within-group improvement but no differences between the groups and concluded that injections with either lidocaine and steroid or lidocaine alone may be efficacious for moderate CTS[47].

Wu et al. published two studies on the use of hydrodissection in carpal tunnel syndrome: the first compared 5 mL of 5% dextrose to a 5 mL saline injection to treat mild-moderate CTS, in which the dextrose showed superior results at 1, 3 and 6 months. Interestingly, the control group responded to treatment as well and they attributed that to a combination of hydrodissection, placebo effect, and spontaneous remission[38]. The follow-up study was a double-blinded study with a saline injection into the carpal tunnel versus saline in the subcutaneous tissue, effectively diminishing the placebo effect[37]. At 2-3 months follow-up, a significantly better symptom score was found in the intervention group, but this effect was lost after 6 months, as we found, and the differences were of questionable clinical value.

These studies, including ours, focused on patient-reported outcomes. Nerve size could potentially be a more objective physiological measure, with measurable changes at 1 week after the injection[48,49]. Although not significant, the nerve decreased an average of 0.7 mm^2 in our study, which is comparable to 0.9 mm^2 from Wu et al., but smaller than found by others[35,50]. Whether this difference is due to the type of corticosteroid or technique used is unclear, but the lack of differences between our groups implies that larger injection volumes with hydrodissection do not affect nerve size recovery, contrary to what Wu et al. found[37]. The goal of hydrodissection is to increase the mobility of the median nerve, due to the disruption of the fibrotic connective tissue, but this was not found in our limited data set. In our study, we included CTS patients irrespective of the extent of their nerve mobility; future studies could select only those with limited nerve mobility to assess the potential for hydrodissection.

Corticosteroid injections are common practice in the treatment of mild-moderate CTS, and although the necessity for ultrasound guidance is debatable[51], it is known that steroid injections are markedly effective in the short term as a treatment for CTS[3]. Based on the published studies and our data, it seems less likely that hydrodissection causes an additive effect exceeding the clinical results as known with regular injections. Since other studies have shown, though, that similar effects to steroids can be found by injections of lidocaine or dextrose, the question could arise as to whether simple hydrodissection, without any drug, might have a beneficial effect. We did not study this possibility, but note that, as a placebo effect seems very relevant in these studies, a well-controlled study in which one group receives an injection and the other hydrodissection with saline seems warranted to answer that question. This could be relevant to provide an option for patients sensitive to or contra-indicated for steroid injections.

Limitations of this pilot study include the underestimation of conversion to secondary treatment within the 6 month time frame. This led to the exclusion of a portion of patient-reported data at the final time point. It is also important to mention that although both the patient and the data analyst were blinded, the interventionalist was not, so we cannot rule that out as a potential source of bias. Incidentally, during a post-hoc data quality review, it was found that one patient received the wrong injection, leading to the

uneven distribution of patients in the two arms, as reported above. There was a minor technical difference in the injection approach between the two groups. This was done to decrease mechanical interference of the needle in surrounding tissue while maintaining the option to observe the sensitive structures i.e. the median and ulnar artery during the procedure. Finally, no parameters that directly measured connective tissue fibrosis and disturbance thereof or electrophysiological follow-up data were included in this study. We can therefore not include any conclusions on the effect of hydrodissection on the tissue or nerve function level.

In conclusion, this was a pilot study designed to assess patient tolerance and to provide preliminary data on the added value of hydrodissection as a technical addendum to a corticosteroid injection in the management of mild to moderate CTS. Our study showed that hydrodissection is well tolerated but that there is no indication for an additive clinical effect when used in conjunction with a corticosteroid. With the increased usage of ultrasound guidance for injections, additional studies will be needed to determine if hydrodissection is beneficial or not when compared to other injection methods used to treat CTS.

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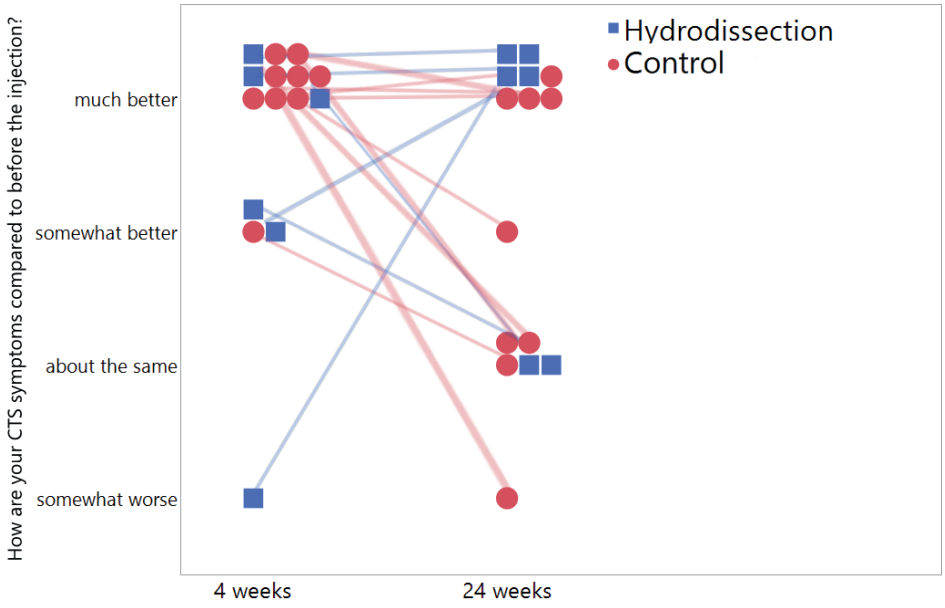
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Supplements

Appendix A: Study criteria

Inclusion Criteria	Exclusion Criteria
• Clinical CTS diagnosis with indication for steroid injection as determined by attending physician AND • adult men and women between age 21 and 80 AND • no more than moderate severity as indicated by electrophysiological test AND • symptoms of numbness or tingling for at least 4 weeks in the median nerve distribution area AND • classic or probable carpal tunnel syndrome on Katz-Stirrat hand diagram AND • ability to complete English-language questionnaires and clinical evaluations AND • is reachable by phone for the follow up contact.	• Previous CTR or other volar wrist surgery on the study hand OR known tumor/mass OR deformity of the study hand/wrist OR • previous history of steroid injection into carpal tunnel OR • currently taking a steroid medication either regularly or on an as needed basis OR • any of the following clinical diagnoses or conditions: Cervical radiculopathy; rheumatoid or other inflammatory arthritis, including gout; osteoarthritis in the wrist; renal failure; sarcoidosis; peripheral nerve disease; diabetes, thyroid disease or other metabolic disorder; pregnancy-induced CTS; amyloidosis; or major trauma (fractures or complete ligamentous tears) to the ipsilateral arm OR • prisoners, institutionalized individuals, or others who may be considered vulnerable populations, such as individuals with dementia. • Any of the exclusion factors on the contralateral hand were not considered reason for exclusion.



Appendix B: Categorical patient-reported improvement at 4 and 24 weeks for those who did not undergo a second injection or conversion to surgery. Each symbol represents 1 patient and lines connect data from individuals.

9

CHAPTER 9.

An incisionless ultrasound-guided carpal tunnel release technique

Schrier, V.J.M.M., Shin A.Y., Brault, J.S. (2020)

Techniques in Hand and Upper Extremity Surgery [epub ahead of print]

Abstract

Ultrasound guidance in operative treatment of carpal tunnel syndrome is gaining in popularity as it non-invasively provides the surgeon with a real-time high-resolution overview of anatomical structures. A new incisionless approach to achieve division of the transverse carpal ligament has been developed that combines ultrasound guidance with cannulated needled and a thread. Conceptually, an abrasive thread is looped percutaneously around the ligament while avoiding injury to neurovascular structures, the palmar aponeurosis, and skin.. The thread is positioned using two puncture sites and a contoured Tuohy needle under ultrasound visualization. With minimal injury to surrounding structures, this approach is designed to minimize recovery time and decrease pillar pain. This article will provide a step-by-step overview of the technique and includes a review of clinical outcomes published so far.

Introduction

Carpal tunnel syndrome (CTS) is a common peripheral compression neuropathy with a prevalence of 4% in the general population[1]. An estimated 71% of patients receive surgical intervention as their primary treatment[2] with an estimated overall cost over $2 billion annually[3]. The recovery time and the economic burden associated with CTS and its treatment is reflected in the days of work missed with a median of 30 days in 2017[4].

Multiple techniques have been described to decompress the median nerve with the common goal of dividing the transverse carpal ligament (TCL). The two most common surgical interventions are the open release (OCTR) and the endoscopic carpal tunnel release (ECTR). During OCTR, an incision of variable length is made over the proximal palm (and/or distal forearm), and the palmar fascia and TCL are divided under direct visualization. ECTR is accomplished with the use of an endoscope and specialized blades through one or two small incisions. Overall, there seems to be no clear superior approach in either functional or symptomatic outcomes. Endoscopic approaches are commonly associated with less post-operative (pillar and/or incisional-) pain[5-7] and a shorter return to work period[5,8,9], but also with increased risk for nerve injury[5-8]. Additional concerns with ECTR include the risk for incomplete releases, steep learning curve[10], and the higher costs mostly due to the required equipment[11]. A study on bilateral CTS patients who underwent both procedures resulted in clear patient preference for the endoscopic approach despite similar symptomatic improvements[12].

Ultrasound (US) has been adopted to assist with visualization in several other minimally invasive surgical approaches to divide the TCL. Varied techniques include the use of, hook knives[13-15], trocar led blades[16,17], guided angled blades[18], meniscotomes[19], needles with retractable blades[20], saw blades[21-23], and fenestration techniques in combination with corticosteroid injection[24]. However, a recognized limition is the need for repetitive cutting motions to divide the TCL.

In 2015, Guo et al. described an US-guided method using an abrasive thread looped percutaneously around the TCL[25,26]. Pulling sequentially on the free ends of the thread cuts through the TCL similar to a Gigli saw cutting through bone. Patient-reported outcome measures were initially equal to existing surgical techniques[25]. After a revision of the protocol to reduce incomplete transection rates, scores have exceeded both open and endoscopic releases[26,27]. This article will describe this modified technique with slight alterations and practical suggestions for US guided incisionless thread carpal tunnel release (TCTR).

General Anatomy

The carpal tunnel is a tubular space on the palmar side of the wrist at the base of the hand. The space is hourglass-shaped and bordered dorsally by the carpal bones that form a backward convex arch. Volarly, the carpal tunnel is covered by the TCL; a tough fibrous ligament that covers the median nerve, all eight digit flexor tendons, and the flexor pollicis longus tendon.

The TCL is attached to the scaphoid and trapezium on the radial side and to the pisiform and hamate on the ulnar side. The volar carpal ligament separates the ulnar neurovascular bundle from the contents of the carpal tunnel.The TCL measures 31mm in length[28] and is approximately 2 to 4mm in thickness centrally. The distal end of the TCL is attached to the longitudinal fibers of the palmar aponeurosis; the palmar aponeurosis lies between the skin and the TCL. For the TCTR it is of great relevance to be familiar with the borders of the TCL since these will be the landmarks for the entry and exit points of the thread.

Anatomical landmarks using ultrasound

Knowledge of the anatomical structures identifiable with US is a prerequisite for the surgeon performing a US-guided TCTR. Ultrasound will help define the entry and exit locations of the cannulated needle and the anatomic structures at risk. From proximal to distal, the TCL can be recognized as a thin arch that lies superior to the median nerve in the transverse plane. The proximal edge of the ligament is positioned deep to the distal wrist crease. In the longitudinal plane, the TCL roughly resembles a disk; wider in the center and thinner at the edges. The superficial palmar aponeurosis is visible as a thin hyper echoic layer on top of the TCL. The distal border of the TCL covers the palmar fat pad for a distance of approximately 2 mm[29], creating an easily recognizable landmark with the US. This has been described as a "duck's beak"[13] (Fig. 1).

Distal to the "duck's beak" lies the superficial palmar arterial arch (SPA) and the sensory cutaneous branch of the median nerve. Identification of the SPA is imperitive as the first needle pass is between the arch and the distal end of the TCL. There has been a wide variation reported on the distance between the SPA and the distal edge of the TCL from 2-26 mm[30] with an incomplete SPA present in as much as 34% of cases[31].

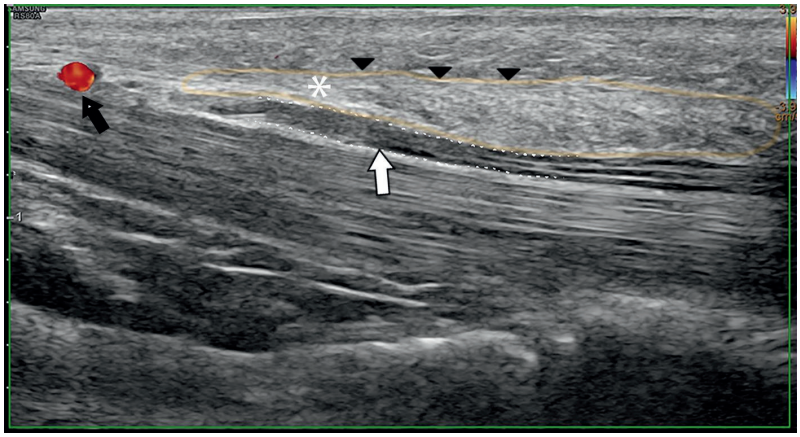


Fig. 1: Longitudinal view of the carpal tunnel at the start of the procedure. The transverse carpal ligament is visible (superficial border with black arrowheads, outline in orange), of which the distal end is also known as the duck's beak (asterisk). Just below the ligament, the median nerve (white arrow) is identifiable. The superficial palmar arterial arch (black arrow and Doppler signal) can be found distally.

After the visualization of the vascular structures at risk, the nerves including the recurrent branch of the median nerve and the common digital nerve to the third and fourth digit should be evaluated. The presence of Berrettini's branch is noted (a superficial communicating nerve branch between the ulnar and median nerves that can arise superficial or distal to the TCL[32]). Eventually, the thread will loop the TCL in the longitudinal plane between the median nerve and either the hook of the hamate or the ulnar vessels (whichever lies closer to the nerve). This area has been referred to as the "safe zone"[13,33] and is identified by a transverse US view (Fig. 2). A cadaveric study has shown that the "safe zone" is larger in men (9.9 mm) than in women (7.7 mm)[33].

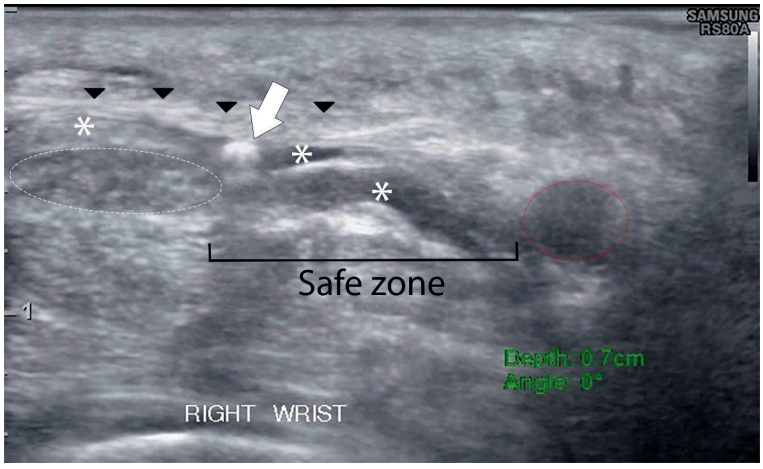


Fig. 2: Intraoperative transverse view of the proximal carpal tunnel. After the first needle advancement, the anticipated position of the thread is assessed (white arrow). The transverse carpal ligament (black arrowheads) is separated from the median nerve (interrupted white oval) by hydrodissection (asterisks) . The thread is located in the safe zone between the median nerve and the ulnar artery (continuous red oval).

Indications / Contraindications

The diagnosis of carpal tunnel syndrome is made by patient history and clinical examination findings, and can usually be confirmed by electrodiagnostic study.Typical symptoms include numbness and tingling in the distribution area of the median nerve, particularly at night. Clinical tests include Phalen's test and Tinel's sign and the presence of thenar atrophy but these are reported to have limited diagnostic sensitivity[34]. Conservative treatment measures are individualized and may include wrist splinting, activity modifications, and a steroid injection into the carpal tunnel.

The primary indication for TCTR is to treat CTS symptoms that are refractory to conservative care. In moderate-to-severe cases of CTS with thenar muscle denervation, the primary goal of TCTR is to deter progressive muscle atrophy and associated irreversible loss of thumb strength[35]. Contraindications include previous OCTR or trauma with scarring and/or altered anatomy. Relative contraindications to TCTR include aberrant anatomy identified by US or failure to safely position the cutting thread. In these cases, the surgeon must decide on either proceeding with US-guided TCTR or converting to an open procedure.

Technique

Step 1 Identification and anesthesia
The patient is placed in a supine position with the forearm in supination. No tourniquet is necessary. The procedure can be done with light sedation or with the patient wide awake. The hand is prepped and draped in a sterile operating room fashion. The pre-operative US assessment includes a review of the patient's anatomy and determination of the needle entry and exit points. First, with the probe placed transversely at the level of the distal wrist crease (carpal tunnel inlet), the median nerve and the ulnar neurovascular structures are identified and the "safe zone" is noted. . The probe is then advanced distally over the median nerve allowing identification of the recurrent motor branch and the common digital nerves. The probe is turned 90 degrees in a longitudinal direction and the superficial palmar arterial arch is located. If available, Doppler can be used to confirm the presence. As the probe is advanced distally, the TCL is identified with the more dorsally located palmar fat pad.

Based on the findings of the US, consideration of converting to an open procedure is made. Situations exist where an open procedure should be considered and include:
- Insufficient visualization of the palmar arch.
- Too narrow of a safe zone.
- Findings complicating the placement of the thread, including a persistent median artery, cysts, masses or other abnormal masses in the volar wrist.

Markings on the skin (Fig. 3) are made to indicatethe course of the median nerve,the ulnar vessels, the safe zone, the proximal and distal border of the TCL, and the SPA. It is essential to mark the planned needle entry and exit points. The *entry* point is located proximal to the SPA, in between the third and fourth rays. The *exit* point falls within a region 1 to 2 cm proximal to the distal wrist crease, in-line with the safe zone.

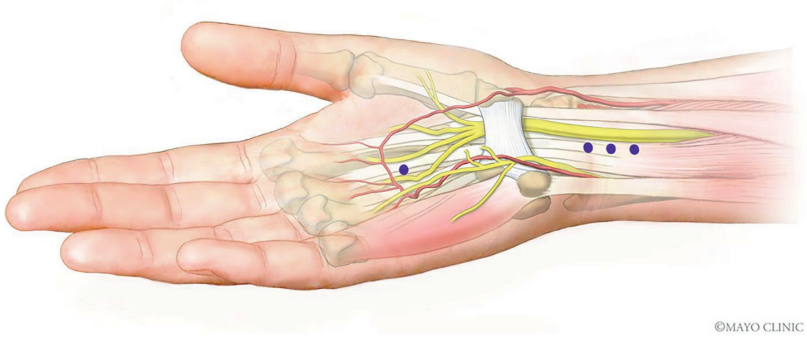


Fig. 3: Skin markings indicate the entry and exit points of the needle based on the position of the median nerve and the ulnar vessels. The distal site is the point of entry, while the exit should be aimed in the region between the two most promixal markings.

For local anesthesia, 0.5-1 cc of 1% lidocaine without epinephrine is infiltrated into skin and subcutaneous tissue with a 30-gauge needle at the identified entry point and the proximal marks. The wide-awake approach allows the surgeon to stay in communication with the patient, for notification of new paresthesias that may indicate contact of the needle with the median nerve. The patient will predictably experience numbness in the median nerve distribution during, or by the end of the procedure as the anesthetic is injected around the nerve for hydrodissection.

Step 2a Preparation for the first needle pass

The goal is to loop the thread around the TCL, staying as close to the ligament as possible. At our institution, a multiple filar stainless steel thread with an overall diameter of 0.22mm (0.0086") is used. The entire procedure is performed under US guidance with the non-dominant hand of the surgeon manipulating the probe. Initially, a straight 25g 1.5 inch needle on a 10 cc syringe with 0.5% lidocaine without epinephrine is placed thru the skin at the entry point directly over the superficial palmar arch and in line with the third webspace. Note that throughout the procedure, all syringes should be flushed with saline to prevent air in the US area since this will cause signal distortion. The needle tip is visualized by US and advanced palmar to the arch and through the palmar aponeurosis.

Hydrodissection is used to dissect connective tissue between the TCL and the median nerve. The landmark of the "duck's beak" is identified to locate the distal edge

of the TCL and its relation with the palmar fat pad. The process of hydrodissection and advancing the needle into the newly created space is repeated to place the needle between the median nerve and the TCL for a distance of approximately 0.5 cm. The injection needle is subsequently removed and re positioned at the exit point proximal to the wrist flexion crease. A small amount of 1% xylocaine without epinephrine is injected into skin and subcutaneous tissue at this site..

Step 2b First needle pass – advancement
An 18G – 4.75 inch epidural needle (Tuohy) is bent at a 30 degree angle 1 cm distal to the tip (bevel up) and at an angle of 20 to 30 degrees approximately 4 cm proximal to the tip (Suppl. I-A). The needle is attached to a 10 ml syringe syringe filled with 0.5% xylocaine without epinephrine to use for hydro dissection where necessary. The modified Tuohy needle is introduced through skin where the injection needle was placed.

This may require a gentle firm push as the needle is blunt. Under US guidance, the Tuohy needle tip is inserted under the "ducks beak" portion of the distal TCL following the path created by hydrodissection. The needle is advanced bevel up, over the superficial SPA and underneath the TCL. Once the tip of the needle is under the distal portion of the carpal tunnel, the position of the needle tip is verified with a transverse US view ensuring its placement between the median nerve and the TCL.. The needle is advanced through the hydrodissected plane, while visualizing the needle tip with a longitudinal US view. When the needle tip is at the proximal TCL border, it is directed 2.5 cm proximal to the wrist crease under the forearm aponeurosis. Longitudinal and transverse US images are obtained to verify accurate placement and if correct, the needle tip is pushed out the skin (Fig. 4A). The Tuohy needle penetration at the exit point can be supported by using a blunt instrument (e.g. hemostat or forceps) against the skin. If there is uncertainty about the needle's position in the safe zone, the needle should be withdrawn and repositioned. The syringe is removed and the cutting thread is inserted through the proximal end of the needle and retrieved at the distal end of the needle (Fig. 4B-C). The proximal end of the thread is secured and the Tuohy needle is removed. During the retraction of the needle, make sure to prevent the thread from being pulled back into the carpal tunnel. The position of the thread is rechecked by US in longitudinal and transverse views.

Step 3 Second needle pass
The same Tuohy needle is rebent with only a 20° bent at the tip and a 10° proximal bend (Suppl. I-B) and is introduced into the same entry point distally. It is advanced under US guidance using hydrodissection and is passed palmar to the TCL. The needle tip is directed above the "duck's beak" between the palmar aponeurosis and the TCL. Transverse US confirmation is performed at the TCL edges to ensure that the needle is positioned superficial to the TCL. Additionally, it is important that the second pass is made in the same longitudinal plane as the first pass to prevent unnecessary inclusion of soft tissue in the thread as verified by US. Two options now arise based on whether the surgeon wants to include the palmar aponeurosis: The needle should either stay *below*

the palmar aponeurosis if this should *not* be transected or *above* the palmar aponeurosis if this needs to be *included* in the transection. The needle should exit the same puncture hole where the thread exits to prevent unnecessary damage to skin and subcutaneous tissue. The proximal end of the cutting thread is placed through the needle and retrieved distally before or with extraction of the Tuohy needle. Both free ends of the cutting thread exit at the distal puncture hole (Fig. 4D-E).

Final verification of the thread placement is done under US. The acronym "TCTR" can be used as a mnemonic to make sure that the thread (T) is correctly placed in both planes, the circulation (C) is protected with the SPA excluded from the loop (both limbs of the thread should be superficial to the arch), the common digital nerve to the third (T) and fourth digit is excluded (if present, this includes Berritini's branch), and that the recurrent branch (R) of the median nerve lies outside the looped thread. Once all structures have been identified and cleared from the path of the thread, the thread ends are placed in a small tube/blunt angiocatheter. The tube will prevent the threads from diverging and lacerating the palmar skin. Each limb of the thread is the secured in a Luer lock tip handle (Fig. 4F).

Step 4 Division of the TCL
The wrist and fingers are gently actively extended by the patient and the Luer lock tip handles are alternatively pulled with about 2 kg (5 lbs) of force while making sure the small tube holding the threads remains close to the skin (Fig. 4G-H). The pull should be in line with the distal entry site to prevent iatrogenically enlarging the entry site and the surgeon's hands should be separated by about 15cm (5-7 inches). The US can be used to visualize the cutting, however, this will require an assistant.

Step 5 Finalization
After dividing the TCL, gauze is used to "roll out" the surgical area to diffuse the hydro dissection fluid. Subsequently, pressure is placed over the surgical site for 5 to 10 minutes to allow for hemostasis. The patient is directed to oppose the thumb to confirm recurrent branch intactness. Numbness in the median nerve distribution area is expected due to the anesthetic in the hydrodissection fluid. The insertion and exit points require no sutures but can be approximated with tape strips if necessary. A bulky dressing is then applied for approximately 10 minutes post-procedure. Instructions include maintaining this dressing for at least 4 hr. Light activity can be resumed as soon as the patient is comfortable, whereas heavy manual labor is discouraged for 2 to 3 weeks. For administrative purposes, Current Procedural Terminology codes 76942 and 64721 can be used (resp. "Ultrasonic guidance for needle placement, imaging supervision and interpretation" and "neuroplasty and/or transposition of the median nerve at the carpal tunnel and includes open release of the transverse carpal ligament").

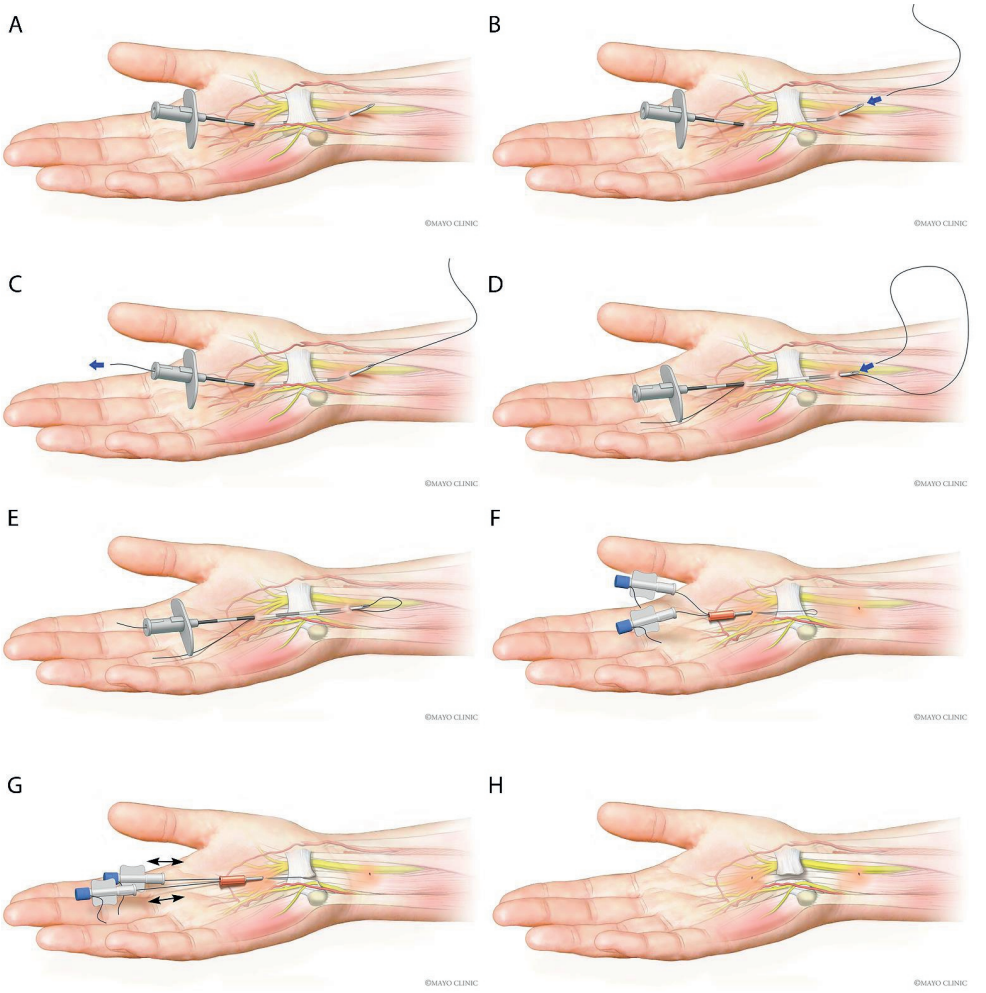


Fig. 4: (A) After hydrodissection, a Tuohy needle is used to complete the first needle pass under the transverse carpal ligament. (B,C) The thread is inserted in the needle, after which the latter can be removed. (D,E) After the second needle pass superficial to the ligament, the thread is looped around the transverse carpal ligament. (F) Two Luer lock tip handles can be used to secure the thread while a plastic tube prevents laceration at the skin site during transection. (G,H) The ligament is divided using a bimanual sawing motion.

Expected Outcomes

The TCTR procedure takes less than 20-30 minutes for an experienced surgeon new to this technique, but surgical times can be expected to decrease to <10 min[25]. Based on the experience of the authors, a learning curve of approximately 50 cases under supervision is to be expected for someone without previous MSK related US experience or 25 cases for someone with US experience.

So far, limited clinical data is available showing the clinical outcomes after TCTR. An initial case series of 16 patients showed a decrease in symptom severity score of 1.4±0.5 and a decrease in function score of 1.2±0.3 after 3 months with no adverse events[25]. In a larger sample (n=159 hands), Guo et al. showed similar scores at 3 months and showed that mean symptoms scores of 1.5±.42 were achievable within 1-4 weeks surpassing OCTR and ECTR in comparable literature[26]. These authors also reported a quick return to work, varying from 1 day to 2 weeks postoperatively. Both studies were without desirable control groups though and presented by the authors who designed the technique. A recent study from Burnham et al. in 20 moderate-severe CTS patients also showed no adverse events and significant symptomatic improvement within 1 week with comparable mean differences as those reported by Guo et al. A patient satisfaction rate of 90% was reported[27]. With a peak incidence of first CTR between 50-59 years for men and 40-49 years for women[36], the majority of patients are of working age, making a technique that allows a faster return to work a relevant one.

Complications

The original surgical procedure included an approach where the TCL looped around the distal edge, cutting the ligament in a proximal direction. This was related to a number of incomplete transections at the distal edge in cadaveric testing[37]. Consequently, we adapted an approach of looping the cutting thread around the proximal edge of the TCL and transecting the ligament in a distal direction. . Depending on the surgeon's expertise, conversion to an open approach is possible when visualization is inadequate and in cases of atypical anatomy. The real-time guidance of the US, and subsequently the proficiency in handling and interpreting the US, is essential for a successful outcome. Up until the transection of the ligament, the procedure is reversible or can be approached from another angle if the surgeon feels uncertain about the thread placement.

Conclusion

US-guided TCTR is a novel, minimally invasive technique that requires detailed anatomical knowledge and experience in musculoskeletal US imaging. Future independent studies with robust designs and adequate controls are necessary to provide more insights into the clinical outcome and added value of TCTR.

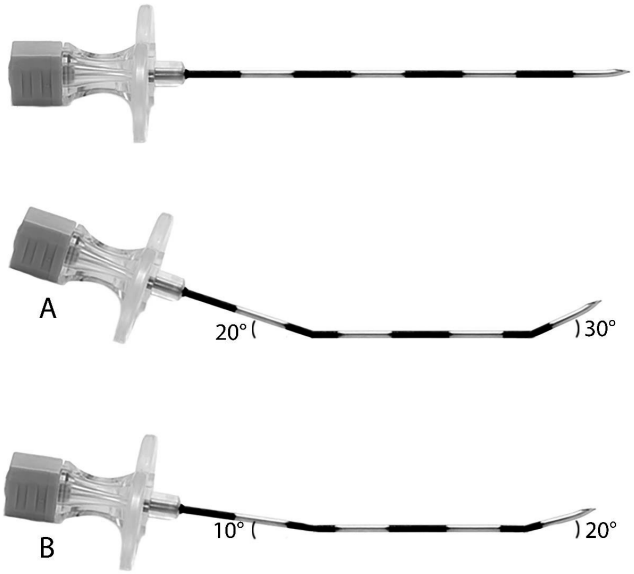
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Supplements



Supplement 1: Suggested angle configurations during the first deep (A) and second superficial (B) needle pass to assist with needle advancement.

10

CHAPTER 10.

Minimal clinically important difference is lower for carpal tunnel syndrome patients undergoing injection versus surgery – letter to the editor

Schrier, V.J.M.M., Gelfman, R., & Amadio, P.C. (2019).

Journal of Hand Surgery (European Volume), 45(1):90-92.

In a letter to the Editor

Introduction

In studies looking at carpal tunnel syndrome (CTS), one of the most commonly used patient-reported outcomes is the validated Boston carpal tunnel questionnaire (BCTQ)[1]. Whether the aim is to determine if a procedure is successful or not, or to show treatment superiority, the added clinical value needs to be addressed. Multiple studies have reported their minimal clinically important difference (MCID) for the BCTQ, which resulted in a range of cut-offs (0.16-1.45) rather than a consensus. De Kleermaeker et al. (2019) highlighted this topic and showed that a better distinction in clinical relevance is found when using a relative MCID, which is corrected for baseline scores[2]. This intuitively makes sense; patients with more severe symptoms are required, and able, to achieve a larger change in score to be equally satisfied as those with milder presentation. We would like to support and add to their findings by sharing the results from our dataset using similar methods.

Materials and Methods

BCTQ data was abstracted at baseline and at 1 and 3 month post intervention in a cohort of idiopathic CTS patients undergoing carpal tunnel release surgery (n=124). Participants also indicated their perception of treatment success on a 5-point scale at follow-up. For our analysis, only the optimal answer, a score of 5/5, was considered "success" (85% of patients). To quantify the discriminatory power of the BCTQ, a receiver-operator curve (ROC) was created including the area under the curve (AUC). Higher AUCs indicate that the independent variable, the (relative) change in BCTQ-score, is more likely to successfully distinguish between 'success' and 'not success' in the dichotomized outcome variable. The ROC allows us to find the optimal BCTQ cut-off value as defined by the highest overall combination of sensitivity and specificity rates.

Results and Discussion

We found an increase in AUC from 0.74 (95%CI: 0.69-0.78) to 0.82 (95%CI: 0.78-0.86) when using the relative MCID versus one based on baseline score alone (Figure 1(a)). The optimal cut-off point was a relative MCID of 0.41 with a sensitivity of 71% and a specificity of 84% which was similar to the results reported by De Kleermaeker et al. (2019). In addition, we looked at whether this was the same for patients undergoing injection, with the hypothesis that expectations might be lower when receiving a corticosteroid injection as compared with surgery. A total of 75 idiopathic CTS patients who underwent injection were analysed using the same method. A smaller percentage of patients (54%) reported the optimal intervention success score of 5/5 at follow-up as compared with the surgical group. Again, the relative MCID had a higher AUC (0.87, 95%CI: 0.83-0.92) as compared with the conventional MCID (0.83, 95%CI: 0.78-

0.88) (Fig. 1(b)). The symptom severity score yielded higher discriminatory potential than the function score in both the surgical and the injection group. Interestingly, the optimal relative MCID for injection patients was indeed lower than that of the surgical group at 0.30, which was associated with a sensitivity of 85% and a specificity of 77%. A similar trend was found with the functional score; a relative MCID of 0.41 was found for the surgical group and 0.27 for the injection group. This suggests that when using the BCTQ in a study that includes an injection group, the relative MCID of 0.30 should preferably be used. Assuming that there are additional confounders affecting success from the patient's perspective, including age, cultural background, workers' compensation, co-morbidities, as well as differing patient preferences and expectations[3], researchers can also consider creating an MCID using a subsample of the specific patient population under investigation. With patient-centred outcomes being increasingly seen as the main criteria of healthcare evaluation, we need to be aware that they provide merely quantification of the subjective rather than an objectification, as was previously noted[4]. In broader context, this study again underlines that when using patient-reported outcomes, a wider view on potential confounders affecting patient-defined success should be considered.

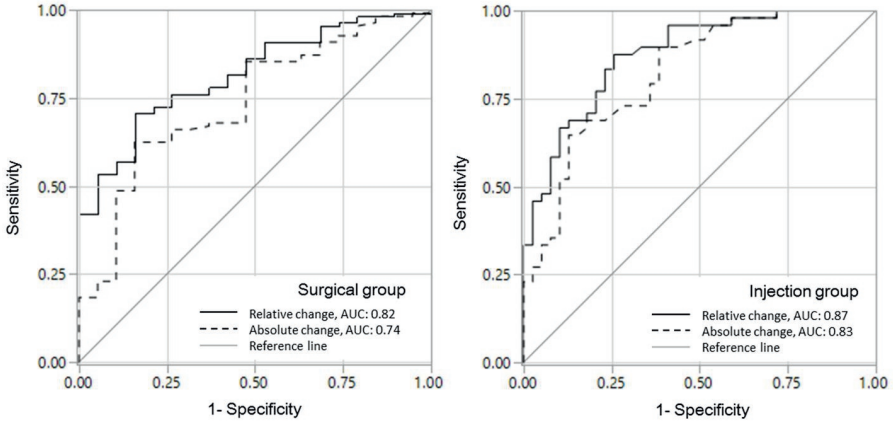


Fig. 1: ROC curves of the (relative) change in BCTQ symptom score after surgery (left) or injection (right). AUC: Area under the curve.

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11

CHAPTER 11.

Better patient-reported experiences with health care are associated with improved clinical outcome after carpal tunnel release surgery

Schrier, V. J.M.M., Poelstra, R., Selles, R.W., Slijper, H.P., Amadio, P.C., Hovius, S.E.R., & Porsius, J.T. (2019)

Plastic and reconstructive surgery, 143(6), 1677-1684.

Abstract

Background
In hand surgery, and specifically carpal tunnel syndrome (CTS), it is currently unknown if experiences with health care influence surgical outcome. In order to investigate whether there is an association between patient-reported health care experiences (PREMs) and symptom relief, data was gathered using a cohort of patients undergoing surgical treatment for CTS.

Methods
PREM and patient-reported outcome measures (PROM) were registered in a national database of sixteen hand surgery practices. PREM data was gathered at three months after surgery and included six subscales on different health care delivery aspects (provided information, communication, facility, operative care). PROM data was acquired before and three months after surgery with the Boston Carpal Tunnel Assessment Questionnaire (BCTAQ). The association was tested using linear regression analyses.

Results
A total of 1607 patients were included in the analysis. PREM scores were good to excellent with a median value between 8.0-8.5 on a 10-point scale. The regression analyses showed a significant ($p<0.001$) association with the BCTAQ for all individual PREM subscales. Biggest effects were found in physician communication and treatment information. PREMs accounted for more than 5% of the explained variance, with patient characteristics approximately explaining an additional 3%.

Conclusion
In this large dataset of CTS patient who underwent a surgical release, a significant impact of health care experiences on self-reported clinical outcome was found. This is relevant information, not only for directing care providers in improving health care experiences as a quality of health care measure but now also potentially to achieve better clinical outcome.

Introduction

Carpal tunnel syndrome (CTS) is the most common compression neuropathy of the upper extremity with an incidence of ~3.5 cases per 1000 person-years[1]. Surgical release of the entrapped median nerve under the carpal ligament is considered the most effective long-term method of treatment[2].

Many studies have focused on identifying predictors of success and failure after surgical treatment of CTS[3–5]. Success after surgery is often described in terms of outcomes directly related to the procedure itself, e.g. lack of complications, symptom relief, and regaining functionality of the hand. These parameters are part of an overall quality rating and can be assessed by the health care providers but can also be reported by patients themselves in patient-reported outcome measures (PROMs). The latter provides a helpful tool, not only for surgical outcome but also for assessing disease severity and disease progression in both a clinical as a research setting[6]. In the field of hand surgery, commonly used PROM tools include the Michigan Hand Questionnaire[7], the Disability of the Arm, Shoulder and Hand (DASH)[8] and the Boston Carpal Tunnel Assessment Questionnaire (BCTAQ)[9] with the latter specifically designed for patients with CTS.

Increasingly more attention has been directed not just towards measuring success post treatment, but also on the incorporation of the evaluation of the *health care process*, in order to provide a more complete measure of success[10]. Patient-reported experience measures (PREMs) are tools that help gain insight into the health care process from the patient's perspective[11] and can act as a robust, distinctive indicator of health quality[12]. So far, PREMS have not often been utilized to investigate the effect of health care experiences on clinical outcome in the field of hand surgery, but they have been used as possible predictors of success for several elective surgeries including knee and hip replacements and hernia repairs[10,13,14]. Also, a recently published study describes using PREMs to distillate points of attention in order to improve their patient's experience of elective surgery. They found e.g. that patients treated later in the day were significantly less likely to report a positive experience. Knowing, this, they could evaluate the effect of targeted improvements for this patient group (decrease waiting time and fasting, establish more realistic expectations) and reported on a 30% increase in positive experiences[15]. This study illustrates how PREMs can be used in a study design for both identification of problems and assessment of the effect. The question remains however, if changes based on PREM scores only improve satisfaction rates or also clinical outcomes.

Since carpal tunnel release surgery is a one of the most commonly performed orthopedic hand treatments with an annual case load of ~4-500.000 in the United States alone[16], it provides an interesting patient population for further investigation. In order to assess the possible association between PREM and PROMs in patients with carpal tunnel syndrome, patients who underwent a carpal tunnel release surgery were selected from a large database with routinely-collected outcome measures. Using a pre-operative PROM

and a post-operative PREM and PROM, the association between a higher PREM score and the difference in clinical outcome will be reported, hypothesizing a positive effect.

Materials and Methods

Study design

This is a cohort study using prospectively acquired data. As part of a routine outcome measurement, all patients who underwent CTR as part of their CTS treatment were asked to participate. Written informed consents were obtained for all participants, in compliance with the Declaration of Helsinki[17]. Patients were included between November 2011 and September 2017 and data was collected in a consortium of sixteen national hand surgery practice sites. Study data were collected and managed using GemsTracker electronic data capture tools[18]. GemsTracker (GEneric Medical Survey Tracker) is a secure web-based application for distribution of questionnaires and forms during medical research and quality registrations. Data extraction was done in in December 2017. CTS diagnosis was made based on symptoms of numbness or tingling in the distribution area of the median nerve and nocturnal aggravation in combination with clinical examination including Phalen and Tinel's tests. To ensure complete data, a selection was made based on PROM baseline and PREM and PROM post-operative availability. If patients were included twice, for example due to a repeat surgery or bilateral CTS, only the chronological first data set was used. Patients were invited to complete an emailed PROM questionnaire prior to surgery and both a PROM and PREM questionnaire three months post-surgery. Two reminders were emailed to non-responders. Additionally, data on age, sex, body mass index, alcohol, smoking, dominance, occupational intensity, duration of symptoms was collected.

Patient-Reported measures

A Dutch version of the Boston Carpal Tunnel Assessment Questionnaire (BCTAQ)[9] was used as PROM. This questionnaire consists of two parts: a symptom severity scale (SSS) and a functional status scale (FSS). The SSS indicates how severe the symptoms are, as experienced by the patient and the FSS indicates how much interference the patient experiences with daily activities. Each score can range from 1 (no symptoms) to 5 (most severe) with the SSS consisting of eleven and the FSS of eight questions.

The PREM consisted of a questionnaire commonly used in private practice clinics throughout the Netherlands. Questions target the patient's experiences with associated health staff and the medical facility at various time points during their treatment. By means of an exploratory factor analysis, as described in a recent study[19], six subscales were identified to assess experiences across different domains: physician communication and competence, peri-operative care, post-operative care, general information, treatment information, and quality of facilities (Appendix A). For each item, patients were asked to give a score in line with the Dutch Academic grading system where a value of '1' represents a very poor result, a value '10' indicates an excellent result and '6' indicates

the lowest passing grade. This method of outcome was chosen because of the high degree of familiarity of the Dutch population with this particular grading system. If a question did not apply to a patient, for example if they did not use the website, there was a possibility to answer accordingly. For questions concerning follow up care it is relevant to know that in the included hand clinics, it is standard protocol for all patients to be offered a follow up by a hand therapist. Internal consistency in our sample, estimated using Cronbach's α, was: physician communication and competence 0.96; peri-operative care 0.52; post-operative care 0.91; general information 0.92; treatment information 0.88; quality of facilities 0.87.

Analyses

All analyses were performed using R statistical computing[20]. PREM subscale scores were calculated by averaging the scores given to the corresponding questions per subscale. PROM results were also calculated by averaging the separate scores for both subscales and the difference between the pre- and post-surgery questionnaires was used as the dependent variable. Significance testing for pre- and post- surgery PROM scores was done using a paired T-test for normally distributed data and a Wilcoxon signed rank test for non-normally distributed data. Distribution of the data was evaluated visually using histograms and QQ norm plots. An effect size was calculated to compare the before and after surgery PROM scores by using Cohen's d for a matched sample comparison (dividing the t-statistic by the square root of n)[21]. To assess the association between increased PREM scores and PROM change scores, linear regression analyses were used. In these models, the beta-coefficients represent the change in overall SSS/FSS score associated with 1 absolute point increase in PREM subscale. Multivariable regression models were used to adjust for potential confounding of various patient and disease characteristics including age, sex, body mass index, smoking, alcohol, occupational intensity, surgery on dominant hand, first time surgery and duration of disease. About 18% of patients had missing data on BMI, alcohol and smoking so the adjusted model was done on a sample size of 1311. An additional comparative analysis on demographics was done to compare the final dataset to those who missed either their follow-up PROM or the PREM. Significant differences were found in age and the amount of positive responders on smoking and alcohol usage, but these were attributed to the large size of the sample and regarded as not clinically relevant. All subsequent analyses were done with the complete data set. To determine to which extent the variation in treatment outcome between patients could be explained by the experience with health care delivery, all six PREM-subscales were introduced simultaneously in the same model as independent variables. Significance was set at an alpha lower than 0.05.

Results

Complete data sets on a total of 1607 patients were included (Fig. 1). Table 1 lists the patient demographics. The sample included more women than men (71.6%), and the majority of cases involved the dominant hand (58.9%). Duration of symptoms varied with a median value of 12 months (IQR: 30).

Absolute scores for both the PROMs and the PREM are shown in Table 2. Both scales of the BCTAQ were normally distributed at baseline with a normally distributed difference in means pre- and post-surgery. The average PROM scores decreased with 1.13 for the SSS and 0.72 for the FSS, showing a significant decrease three months after CTR ($p<0.001$) and effect sizes of 1.50 and 0.94 respectively. PREM scores indicated a generally positive experience, ranging between median values of 8-8.5 over the six different subscales. Different subscales showed between 0-18% missing data, the main reason for which was that some of the specific topics (like information provided via a website/ brochure) in the PREM were not applicable to specific patients.

The unadjusted regression analyses for both subscales of the PROM showed significant ($p<0.001$) negative associations with all individual PREM subscales (Table 3), indicating that better PREMS score are associated with better (that is, lower) PROM (BCTAQ) change scores. Values ranged from -0.08 to -0.17 in PROM point difference as a result of a one point increase on the PREM subscales. The strongest associations were found in the 'physician communication and competence', 'post-operative care', and 'treatment information' subscales. Explained variance in PROM score, considering all PREM subscales, was 5.5% for the SSS and 5.3% for the FSS. In the adjusted, multivariable regression, patient characteristics added to the explained variance to a total of 8.9% for the SSS and 9.8% for the FSS.

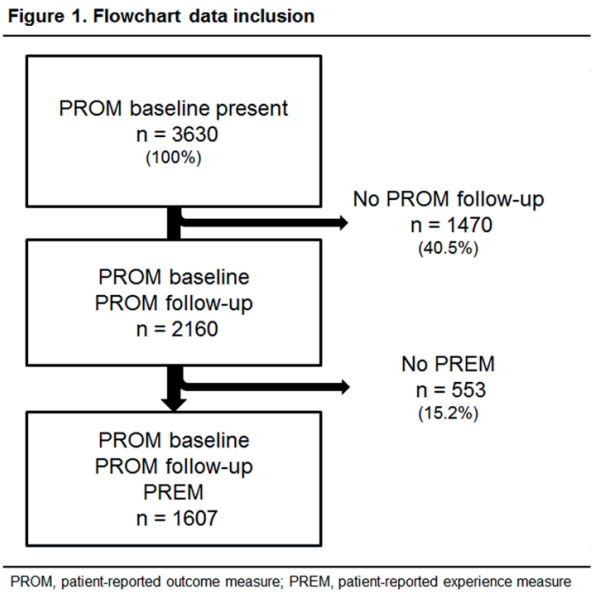


Figure 1: Flowchart data inclusion

Table 1. Patient characteristics (n=1607)

Age, years (mean, SD)	54.2 ± 12.6
Sex (female)	71.6%
First CTR	89.0%
BMI, kg/m^2 (mean, SD)	27.4 ± 5.0
Smoking (yes)	18.0%
Alcohol (yes)	56.2%
Occupational intensity	
No occupation: Unemployed/ retired	38.0%
Light (e.g. office work)	23.3%
Medium (e.g. cleaning)	24.3%
Heavy (e.g. construction work)	14.4%
Surgery on dominant hand	58.9%
Duration of symptoms, months (median (IQR))	12 (30)

Table 2. Patient-Reported Measures scores

	Pre-operative	Post-operative	
PROM scores (mean, SD)			**Effect size**
Symptom Severity Score	2.85 (±0.64)	1.72 (±0.61)*	1.50
Functional Status Scale	2.47 (±0.75)	1.75 (± 0.68)*	0.94
PREM scores (median (IQR))			
Physician communication & competence (n=1607)		8.2 (1.3)	
Peri-operative care (n=1529)		8.5 (1.0)	
Post-operative care (n=1325)		8.0 (1.0)	
General information (n=1320)		8.0 (1.0)	
Treatment information (n=1587)		8.0 (1.3)	
Quality of facilities (n=1594)		8.2 (1.2)	

*Difference in mean score pre- versus three months post-operative, p<0.001.
PROM, Patient-Reported Outcome Measure; PREM, Patient- Reported Experience Measure; SD, standard deviation; IQR, Inter Quartile Range. Sample sizes in the PREM scores differ based on applicability of the question to the individual patient.

Table 3. Summary of regression analyses for PREM variables and the prediction of PROM change scores

	Change in Δ PROM subscale score (95% CI)			
	Symptom Severity		Functional Status	
	Unadjusted	Adjusted*	Unadjusted	Adjusted*
Physician communication & competence	-0.16 (-0.19, -0.12)	-0.15 (-0.19, -0.12)	-0.15 (-0.18, -0.11)	-0.15 (-0.19, -0.11)
Peri-operative care	-0.11 (-0.14, -0.07)	-0.10 (-0.14, -0.06)	-0.09 (-0.12, -0.05)	-0.10 (-0.14, -0.06)
Post-operative care	-0.15 (-0.18, -0.11)	-0.14 (-0.18, -0.10)	-0.15 (-0.19, -0.11)	-0.15 (-0.19, -0.11)
General information	-0.10 (-0.14, -0.06)	-0.08 (-0.13, -0.03)	-0.09 (-0.13, -0.05)	-0.09 (-0.13, -0.04)
Treatment information	-0.16 (-0.20, -0.13)	-0.16 (-0.19, -0.12)	-0.17 (-0.20, -0.13)	-0.17 (-0.20, -0.13)
Quality of facilities	-0.14 (-0.18, -0.09)	-0.13 (-0.18, -0.08)	-0.12 (-0.16, -0.07)	-0.12 (-0.17, -0.07)
Explained variance (%)**	5.5	8.9	5.3	9.8

*Adjusted for age, sex, body mass index, smoking, alcohol, occupational intensity, surgery on dominant hand, first time surgery, duration of disease. Sample size for adjusted regression n=1311.
**For all six PREM-subscales combined
All beta-coefficients reach significance with p< 0.001.
PROM, patient-reported outcome measure; PREM, patient-reported experience measure; CI, confidence interval

Discussion

In this large dataset involving CTS patients who underwent a surgical release procedure, we found that better health care experiences are associated with better clinical outcome in self-reported post-surgical questionnaires. The largest effects were found in the experiences with physician communication & competence, post-operative care and treatment information. Altogether, the PREMs constituted more than 5% of the explained variance in PROM score change, where patient characteristics explained an additional 3.5-4.5%.

Both our effect size as well as the PREM-PROM association has been in line with what has been described before[22]. In a study by Black et al., they found an overall weak association on both the Oxford Hip Score and the Oxford Knee Score after hip or knee replacement as a response to change in overall PREM scores[10]. More specifically, their biggest effect was also found in the doctor's trust and communication section, but like in our study, data did not allow for any conclusions on (the direction of) causality.

The influence of patient-physician communication has been described in other articles as well: On satisfaction, good communicative skills have a stronger effect than, for example, time spent with the patient as was found in patients from hand and orthopedic surgeon populations[23–26]. Satisfaction is thus linked more to perceived quality of the conversation than the quantity. Additionally, a recent meta-analysis has found that patients report more beneficial health behavior, less symptoms and higher quality of life scores when they reported more trust in their health care professional[27]. Improving physician empathy with e.g. communication training has been shown to lower depression[28], reduce pain levels[29], and improve medication adherence[30]. Another meta-analysis identified thirteen articles describing randomized controlled trials that empirically tested methods of improving the patient-clinician relationship, focusing on trust and empathy. After improving the patient-clinician relationship, a small but significant effect was found on outcomes in a variety of pathologies[31] indicating the possible direction and translatability of studies performed concerning this topic.

Another PREM category that stood out in our results was treatment information. The relatively higher scores in this subcategory indicate that taking away uncertainties about the surgical process and providing information on the recovery period will help in achieving better PROMS. A previous survey study showed that it is mostly the surgical outcome that determines the success in the eyes of the patient, so achieving a realistic expectation could improve clinical outcome[32].

The main strengths of the present study are the size of the data set, the robust implementation in standardized clinical care and the emphasis on patient-reported data. The internal consistency was excellent for all subcategories except peri-operative care. Since the PREM questions were selected on applicability from a larger set of questions, only two questions remained in this category which in part explains the lower consistency. This study focused on using PROMs as the main outcome parameter after surgery rather than the perhaps more classic objective measurements like grip strength or the need for a second operation. Since patients and clinicians do not appear to have a universal

agreement as to what determines success after surgery[33,34], objective measures might not adequately reflect success of a surgical procedure. As mentioned before, modern medicine is shifting more towards a personalized approach with an emphasis on patient feedback which includes having a better understanding of patient experiences and how possible effects on clinical outcome fits in that model.

Intrinsic to the design of the acquisition method used to create the database, there are limitations to the generalizability of our results. The explained variance and increase in PROM scores in this study were tested by means of a multiple regression, meaning that, although an effect was found, causality is not proven. It might be that our findings indicate that better post-operative outcomes lead to a more positive memory of health care experiences. Secondly, potential confounders like psychological state or recollection bias of the patient were not taken into account in this analysis. Future studies should address this using more extensive measurement tools, acquired at multiple time points including a pre-surgical evaluation. Also, information was gathered based on patients who visited a specialized care center outside of an academic/regular hospital setting, implying that there could be a selection bias with a relatively low occurrence of non-typical CTS presentations. The structural validity of the PREM questionnaire used in this study was determined using a factor analysis, but it is important to note that no test-retest reliability, responsiveness or cross-cultural validity validation was done. Additionally, in the current sample, detailed information on other (relevant) musculoskeletal diseases, medical history and occupational intensity was not available. In standard practice, occurrences of corticosteroid injections and other conservative therapies, which might have played a role in our clinical outcome, are very low due to reserved usage as described in guideline published by the Dutch Association of Neurology[35]. Nonetheless, any interference of other hand related pathologies on the BCTAQ scores needs be taken into consideration. This applies mostly to the functional scale as the symptom severity scale uses questions specifically designed for the clinical CTS presentation.

Conclusion

In conclusion, this study demonstrates that experiences as reported by CTR patients on health care delivery are associated with their treatment outcomes. Although it is important in its own right to assess and improve health care experiences of these and others patients, our study provides an additional reason to further explore interventions aimed at improving the context in which health care is provided. These future studies would also support uncovering the presence of causality and potential directionality of the relationship between PREMs and PROMs.

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Supplement

Appendix A: PREM questionnaire*

Physician communication & competence	Your grade:										
What do you think of the personal contact between you and the doctor?	1	2	3	4	5	6	7	8	9	10	NA
How well did the doctor listen to your input?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the way the doctor treated you (politeness, etc.)?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the time the doctor has taken for you?	1	2	3	4	5	6	7	8	9	10	NA
Did the doctor explain things in an understandable fashion?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the doctor's medical expertise?	1	2	3	4	5	6	7	8	9	10	NA
Peri-operative care											
How do you feel about the anesthesia used during your surgery (local, regional or otherwise)?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the guidance/care provided by the nursing staff?	1	2	3	4	5	6	7	8	9	10	NA
Post-operative care											
What do you think of the hand therapist's guidance before and after treatment?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the information that the hand therapist has provided you?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the alignment in communication between the hand therapist and the doctor?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the aftercare provided by the clinic (recovery period, controls, medication, emergency)?	1	2	3	4	5	6	7	8	9	10	NA
General information											
What do you think about the quality of the information brochure?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the information on the website?	1	2	3	4	5	6	7	8	9	10	NA
Treatment information											
What do you think about the information provided to you prior to your treatment?	1	2	3	4	5	6	7	8	9	10	NA
Do you feel like you have been well informed about the results, alternatives and risks of the treatment?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of information about the aftercare (checks, emergencies, etc.) after your treatment?	1	2	3	4	5	6	7	8	9	10	NA
Quality of facility											
What do you think of the telephone accessibility of the clinic?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the telephone assistance?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the in the clinic cleanliness?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the clinic's accessibility and parking availability?	1	2	3	4	5	6	7	8	9	10	NA
Has Xpert Clinic performed your treatment safely?	1	2	3	4	5	6	7	8	9	10	NA
What do you think of the way you were received at the clinic (hospitable, friendly, etc.)?	1	2	3	4	5	6	7	8	9	10	NA

*Questions translated from original Dutch questionnaire, PREM: patient-reported outcome measure; NA: not applicable

12

CHAPTER 12.

General discussion

This final Chapter will be used to reflect on the three main aims of the thesis including a discussion of the results of the research presented. The three headers correspond directly to the Thesis sections. **Part I** describes SSCT function and structural development in order to describe its role in CTS development. **Part II** describes recording these biomechanical changes to assess disease severity and prediction of surgical treatment response. Since it remains challenging to predict how CTS patients will respond to treatment, and with the prevalence of CTS not likely to decrease in the years to come[1], identifying prognostic parameters aids in the decrease of treatment failures, improving patient satisfaction, and lower healthcare costs[2]. Then, in **Part III**, new applications of ultrasound in carpal tunnel release surgery and injection will be discussed. Visualization of the anatomical structures at risk as provided by ultrasound guidance during an intervention can lead to less intra-operative and post-operative complications and opens doors for treatment improvement. Finally, we change perspectives from enhancing the technical aspects of healthcare to understanding the impact of healthcare experiences as seen by the patient and close with a final remark.

Pathophysiology of CTS and connective tissue (aim 1)

Carpal tunnel syndrome refers to the symptoms that arise due to compression of the median nerve at the level of the carpal tunnel. Increased intra-carpal tunnel pressure is known as one of the hallmarks of CTS pathology and a decrease in this pressure can be measured directly after carpal tunnel release surgery[3]. The cause of the pressure increase seems to be multifactorial but is associated with repetitive, high force and high velocity finger and hand movement[4,5]. In **Chapter 2** we combined the results of published work looking at these epidemiologic findings from a biomechanical perspective. We describe that the subsynovial connective tissue surrounding the tendons and the nerve has unique features that are currently not known to be found elsewhere in the body. The layered structure, composed mostly of proteoglycans and collagen types I, III, and VI[6], carries blood supply to the tendons, but also seems to protect the nerve from excessive longitudinal gliding by absorbing the majority of the shear stress during finger flexion and extension[7]. Histological samples have shown fibrotic changes in this tissue in CTS patients as well as tearing of the connections between the tendon and the directly adjacent SSCT. Cadaveric studies, animal models, and ultrasonographic evidence has led to a proposed model in which initial damage to the SSCT leads to an overall decrease in its capacity to function as a force buffer with neuro-ischemia, edema and the initiation of a self-sustaining loop of increased gliding resistance as results. Questions remain, however, on why some people go on to develop CTS while others seemingly at risk do not. In order to see whether the characteristic structural layout of the SSCT is a product of mechanical adaptation (much like the dynamics in bone formation in response to force loading) or whether the SSCT as seen in adults is already present during the fetal stage, we described the chronological SSCT development using a rabbit model in **Chapter 3**. Using scanning electron microscopy, we showed that the SSCT is formed at an early fetal stage as a single solid layer between the tendons that does not specialize in individual

horizontal sheets until weeks after birth. Although we were limited by the number of tissue samples, the results matched the ambulatory development of the rabbit kit with loading of their front limbs. The morphological development fits the idea that there is a dynamic adaption of the SSCT to loading. In the context of CTS, this warrants the question of whether those who go on to develop symptoms have an inadequate adaptive response, emphasizing the role of activity adaptations and prevention of excessive force loading. Another question is why the adaptive response is inadequate in the first place. The fact that CTS is a disease mostly seen at a later age could represent the accumulated effect of, amongst others, long-term flexor tendon loading and the inability to distribute the forces adequately or, conversely, altered cellular response due to senescence or other factors. Nonetheless, idiopathic CTS does also present in younger people, so they might be a good subpopulation to investigate the adaptive potential of SSCT.

Where Chapter 2 emphasizes the potential role of the SSCT in CTS development by combining observational and experimental studies within one framework, Chapter 3 presents new information detailing the onset of the multilayered architecture until after birth. Cellular responses to mechanical forces have been reported in bones and tendons outside the carpal tunnel[8,9], but now we have evidence that this dynamic response is also present in the connective tissue. Combining this with the proposed model in Chapter 2, repetitive damage to the SSCT and subsequent fibrosis[10] is thus more likely the result of a failed adaptive response, emphasizing the role of activity modification for those with new-onset symptoms and mild presentation. In addition, the model is supported by recent studies showing the potential for new therapeutics that target SSCT fibroblasts by suppressing fibrotic genes and SSCT cell proliferation[11].

It is important to note that although the SSCT might play a relevant role in CTS development, there are other known (anatomic) factors that are of importance. For example, CTS patients tend to have smaller carpal tunnels [12], in which the structures do not necessarily follow the same ratios (resulting in relatively large areas occupied by the flexor tendons and the median nerve)[13]. It has also been found that in some patients the lumbrical muscles infiltrate the carpal tunnel during a grip of the hand (as might frequently occur in laborers), causing decreased carpal tunnel volume and an increased risk for CTS development. Finally, CTS is also often described as a cyclic ischemia-reperfusion injury process[14]; increased carpal tunnel pressure causes the capillaries in the SSCT to collapse[15] (intermittently) and, as a result, CTS patients have measurable differences in intra-neural blood flow[16].

Future research looking at the pathophysiology of CTS will have to combine not only the role of the SSCT with disease, but also incorporate structural development. For the development of new treatment strategies, CTS will have to be looked at from a cellular, tissue, and an anatomical level.

(Dynamic) ultrasound in the clinical evaluation of a CTS patient (aim 2)

Previous publications have shown diminished nerve displacement in patients with CTS[17]. Our goal was to build on this finding by testing whether similar measures of nerve and SSCT mobility would hold prognostic value.

Longitudinal mobility

Non-invasive measurement of the SSCT has been done before using both commercial tracking software as well as specifically-designed algorithms. So far, published work on speckle tracking has been prone to overestimation due to the incorporated measurement of perceived motion from static structures[18]. Recently, an improved version of our speckle tracking algorithm has been shown to more accurately measure tendon displacement[19]. In **Chapter 4**, we have shown that the clinical application (represented by repeated measurements and multiple interpreters) of the speckle tracking provides reliable results, in both tendons as well as SSCT, in both control participants as well as CTS patients. Tendon tracking showed higher repeatability values than the SSCT, which we can in part explain by the difference in size and the higher likelihood of the SSCT to fall outside of the preset and stationary region of interest. Nonetheless, the repeatability in CTS patients of the shear index, a relative measure of excursion between tendon and SSCT, was excellent with an ICC of 0.82 (95% CI: 0.61-0.92). This finding allows us to apply this method of ultrasound image processing on clinical data to test for an association with disease severity and clinical outcome, which we did in **Chapter 5**. In this Chapter, we included a surgical cohort of 125 patients, 90 of whom had analyzable SSCT data. The average shear index at baseline was 79%, indicating that for every centimeter the tendon moves, the SSCT moves only 0.2 mm. When relating this to disease severity, we found decreased SSCT motion, but also a considerable variation between subjects, implying that it will be challenging to use this parameter for patient-based assessment. A similar study looking at the relative motion of the nerve to the tendon found decreased nerve motion with increased disease severity but found no correlation when measuring SSCT[20]. We then tested the US measurements against clinical outcome at 1 and 3 months, but no statistically or clinically relevant associations were found.

Two main challenges limit the measurement and validation of longitudinal SSCT motion with ultrasound. A first challenge is that the SSCT is approximately 0.7mm wide, so measurement of the SSCT requires a high spatial resolution; secondly, rather than a homogenous layer, it shows visco-elastic properties with layers closest to the tendon responding differently from those further away[21]. Any assumptions on SSCT response and expected trajectory, therefore, need to account for both a spatial and temporal correction. These challenges do not only defy the measurement itself, as showcased by the fact that 28% of our surgical cohort had unanalyzable US images (mostly due to out of plane motion), but also the design of an adequate validation test. Different versions of the speckle tracking algorithm have been validated for tendon tracking,[18,19] but authors cautioned for parameter alteration when applying the speckle tracking to

SSCT movement. Other SSCT validation tests were done with relatively large metallic markers or were done intra-operatively[22-24]. However, both examples are likely to disrupt the SSCT infrastructure and thus its mechanical character. With the ever-continuing development and increased affordability of high-performance ultrasound machines, the limitations posed by resolutions are most likely of a temporary nature. A validation study involving a simplistic representation of the SSCT and its movement using phantom materials is a low-cost approach to establishing error margins of the speckle tracking performance.

Transverse mobility

Using the same cohort, we also looked at an easier approach to quantify nerve mobility, namely the transverse excursion of the median nerve. In **Chapter 6**, we showed that this can be reliably measured between raters, but that for non-static parameters, there is a considerable test-retest variance (ICC of ~0.60 during wrist flexion). This was also reflected in the coordinate plot in which all individual nerve end positions were represented, spread out over an almost 180° dorsal field. After carpal tunnel release surgery, the mean nerve displacement during wrist flexion increased significantly in a dorsal (i.e., more normal) direction, but when tested in a linear regression model, no prognostic value was found for nerve mobility. In contrast, a larger nerve area was associated with better functional clinical outcome. We also used a non-linear approach by looking at extreme responders (either extremely positive or extremely negative clinical outcomes) but neither showed an identifiable movement pattern that differed from the rest of the sample.

Nerve Elasticity

In the final chapter of this section, we investigated the sensitivity of a novel ultrasound technique that quantifies tissue elasticity. Ischemia, neuro-edema, and fibrosis occur in the connective tissue sections within the epineurium. After longer periods of exposure, this promotes fibroplasia, causing replacement with scar-like tissue. These biophysical changes are reflected in the increase in nerve stiffness. As shown in the results in **Chapter 7**, shear wave elastography is capable of adequately reflecting changes in nerve elasticity without being significantly influenced by surrounding high stiffness tissues or artifacts caused by the anisotropic nature of the nerve. Part of the study was designed to provide practical suggestions for a measurement protocol. In the literature, both transverse as well as longitudinal measurement approaches have been reported25, but based on our results, longitudinally acquired elasticity results are significantly higher compared to the transversely acquired data. Therefore, we have reason to warrant direct comparisons between studies and to recommend using the longitudinal approach for future designs. By confirming that SWE can detect changes in elasticity characteristics of the nerve under increasing tensile loads, our study also emphasizes the possibility of applying SWE for the evaluation of patients who are immobilized or are undergoing nerve gliding and stretching exercises.

Parallel to the identification of more details relevant to CTS pathology, new US techniques are being developed which can help these new facets relevant to disease progression. An exciting new approach for example is to look at shear wave elastography to function as a proxy for overall carpal tunnel pressure[26,27], which exemplifies a combination of a historically known disease feature with new techniques.

Conclusion to AIM 2

To summarize, we looked at two methods to quantify nerve and SSCT mobility in CTS patients, with both showing limited potential to provide additional support in the decision for surgical treatment. Although we *can* measure these parameters reliably, our data suggest that at this point we *should not* use them in daily clinical practice in patients who are otherwise indicated for surgery, since they provide no additional information to predict outcome. The lack of association between US and clinical outcome was present in both planes (transverse and longitudinal). Important limitations of the study included that firstly, there was a relatively high success rate after surgery, which prevented testing for factors causing poor outcome. Secondly, there was a wide range of the inter-subject US results in both the nerve and SSCT excursions which limited testing for poor outcome. Nonetheless, our study results were derived from a clearly-defined cohort, preceded by adequate power analysis, specifically targeting idiopathic CTS patients. Similar research done before on the altered biomechanical response of carpal tunnel structures was mostly with small sample sizes[28-30], with the nerve motion calculated relative to other moving structures[20], or without clinical result as outcome.

Opportunities for **future research** directions could include a phantom-based validation of the SSCT using the speckle tracking techniques as presented here. A finite element model of the SSCT has already been proposed[31,32], so a combination of the experimental validation with computational modeling seems within reach. Potentially, biomechanical changes in the SSCT can then also be included in the model and phantom. Also, future studies could focus on the inclusion of patients receiving conservative or injection-based interventions. This population is likely to present a wider spread of outcomes, making statistical identification of those likely to respond more feasible.

Although the use of shear wave elastography is still in its explorative phase, there is evidence to suggest that we will be able to use elasticity as a new measure for CTS severity[33]. Additionally, with advancing machine learning tools and subsequent improvement in data processing, associations hidden from the investigator's eye might be unveiled when combining multiple US measures and patient characteristics. With morphological features as the main US parameters of interest, automated segmentation and real-time assessment in three dimensions will be interesting paths to take. The full potential of US as an addendum to the diagnostic workup including post-treatment evaluation and relation with symptomology will have to be topic of future research.

Improving treatment and the assessment of success (aim 3)

Improving surgical and injection-based intervention

There is a wide variation in the type and volume of corticosteroid injection in the clinical treatment of CTS. In **Chapter 8**, we presented preliminary data on the added effect of US-guided hydrodissection. Working with the hypothesis that hydrodissection would be less patient-friendly due to the increased volume, we found that there was no clinically relevant difference in patient satisfaction between the two groups. However, at 1 and 6 month follow-up, data showed no differences in clinical outcomes. The added value of hydrodissection has been described[34,35], but so far no robust studies with clinically relevant control groups have shown superiority over regular (US-guided) injections, making it too early to draw any conclusions on the usefulness of hydrodissection. Our study provides data for a power analysis but based on the results we are inclined to advise a study that addresses whether hydrodissection alone would have an effect equal to an active injection. For clinical practices, where the added costs that come with US-guided interventions are not cost-effective in standard CTS care, the patient group who would benefit most from this study might be those contra-indicated for a corticosteroid injection. The corticosteroid injection has a proven added role in CTS management, but with approximately 2/3 of injection patients requiring secondary treatment[36], the superior clinical outcomes[37] and decreased costs[38] with US-guidance, identifying the right patient for this approach is also warranted.

US-guidance is not only useful for injections but also provides new options for surgical releases. Incomplete ligament transection is one of the most common complications of carpal tunnel release surgery, causing persistence or recurrence of symptoms. US provides a fast and easy way to evaluate ligament division reliably, even in an office setting[39-41] and has associated with a steep learning curve[42]. **Chapter 9** introduced a novel carpal tunnel release technique that uses an abrasive thread as the divisive tool. This work is mostly a resource for those interested in learning the technique as clinical studies are required to confirm the preliminary positive outcomes since these were mostly presented by the designers of the technique. Clinical studies should include a relevant clinical outcome including patient satisfaction but also the evaluation of the learning process for the surgeon not necessarily familiar with US. So far, studies indicated a significant decrease in the often debilitating post-operative rehabilitation program for patients as well as the chance of pillar pain[43,44]. With the main incidence of CTR cases burdening patients within working age, any alteration that allows for faster return to work seems to be a relevant one.

Factors involved in the measurement of success

Any study that assesses effectivity needs a relevant outcome. This can be done in the form of quantifiable measurements (grip strength, sensitivity, etc.), but currently patients are increasingly being asked to complete patient-reported outcome measurements at different time points. With a known poor correlation between physician's and patient's interpretation of success[45], understanding adaptable factors in healthcare that a patient

(consciously or subconsciously) deems important is essential. **Chapter 10** showed that the minimal clinically important difference (MCID) of the most commonly used patient-reported outcome in CTS is dependent on what type of treatment they receive, irrespective of the severity of their symptoms at baseline. Researchers implementing this questionnaire should take that into consideration in the design and interpretation of their study. Consequently, it also introduces the question what other factors are relevant to reach the "minimal" point of patient satisfaction. It was not until recently that a study came out that the MCID should be individualized based on baseline severity[46], but if factors like time point of survey, type of physician, and the presence of ultrasound-guidance during an injection also influence the MCID, then it might not be much different from a regular multivariate model that predicts the answer to the question "Are you satisfied?". The added value of the MCID as an evaluation construct has been questioned by others as well[47-49]. Then, in **Chapter 11**, we looked at a large cohort of Dutch surgical CTR patients and found that patient-reported experiences significantly affect surgical outcome. More specifically, the biggest effects were found for the patient-rated experience concerning physician communication and the quality of treatment information. The healthcare experience accounted for 5% of the explained variance in outcome score with patient characteristics providing an additional 3.5-4.5%. This implies that besides hypothesized alterations to improve surgical technique, research focus should also include experiences before the intervention. Although causality is not proven, this study showed that for CTS patients subjective experience measures are reflected in what we consider being objective surgical outcomes.

Future studies: The small but significant difference in MCID threshold between CTS patients who received an injection and those who underwent surgery implies that the "one fits for all" single MCID value that is usually applied in CTS research is not suitable. Perhaps by finding these confounders, instead of weakening the (assessment of) patient-reported outcomes, we could see this as an opportunity to gain insight into potential improvement to the patient-oriented evaluation. Just like we found that *type of treatment* and healthcare *experience* play a clinically significant role in treatment outcome, we need to be critical of what effect sizes we expect to gain from changing interventions and be open to shifting our efforts to a more holistic view in which we incorporate everything in the healthcare process of the CTS patient. For future studies within the field of hand surgery, emphasis could be placed on the incorporation of social science perspectives including evaluation of physician empathy and communication skills.

Final remarks

To me, the last part of the thesis emphasizes how important it is to be part of a shared-decision making process in which clear communication on expectations and limitations of current treatment options for CTS are essential. This loops back to the start of the Discussion where we started with microscopic changes in fibril composition in the SSCT, all in the quest of finding puzzle pieces to help create a more rigid and tailored prognostic model. As a final comment at the end of this thesis, I believe that although ultrasound has yet to be proven to be that puzzle piece in its assessment of carpal tunnel dynamics in surgical patients, similar research questions using injection patients remain to be studied in the other arm of the DUCATS study. Future successes will be reached by translating new biological and epidemiological findings in CTS etiology to improved evaluation techniques and by keeping an open attitude towards looking into all patient care elements from start to finish.

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13

CHAPTER 13.

Summary

Carpal tunnel syndrome (CTS) is the most common peripheral neuropathy. It involves increased intra-carpal tunnel pressure and subsequent neuropathy of the median nerve distal to the carpal tunnel. CTS is hallmarked by tingling and numbness of predominantly the thumb, index, and long finger, but can progress to a more severe stage involving muscle weakness and irreversible nerve damage. Decisions concerning the type of treatment are made based mostly on disease severity, but currently, in part due to the complexity of the pathophysiology, there are limited treatment predictors available to help guide this decision. In this dissertation, we looked into CTS by exploring the role of connective tissue in idiopathic CTS, by describing the role of ultrasound in the prediction of clinical outcome, and by assessing potential targets for improving treatment.

Part I CTS pathophysiology and the role of connective tissue

Increased carpal tunnel pressure is a hallmark of CTS, but in the majority of cases, the cause of this increase is unclear. In the first part of this thesis, we described the complexity of CTS pathophysiology and the association with high-force and high-frequent finger motions. One area of research has indicated a prominent role for the subsynovial connective tissue (SSCT) in the carpal tunnel. The review in **Chapter 2** details the overall mechanical and anatomical changes of the median nerve as well as the SSCT. CTS patients show increased fibrosis in the SSCT, making it less flexible and more prone to additional damage with prolonged shear stress during finger and hand motion. We describe a hypothetical model in which initial damage to the SSCT leads to a self-sustaining feedback loop of continuing and increasing damage and fibrosis. Although it is a hypothetical model, this can function as the starting point to increase our understanding of CTS pathology and support the development of targeted therapies, for example to prevent additional SSCT fibrosis.

In **Chapter 3**, we build on this by assessing the chronological development of the SSCT in a rabbit model. Using scanning electron microscopy, we observed and non-quantitatively compared the architecture from fetal to young adults. We found that the SSCT starts out as a solid and dense structure surrounding the flexor tendons, but after a few weeks after birth splits into a multilayered system interconnected by collagen fibrils. Although a causal relation could not be tested, this change coincided with the young rabbits showing weight-bearing movements and could thus imply an adaptive response to loading. This finding could be of clinical relevance considering the prevention of further SSCT damage, as seen in CTS patients, by stimulating the adaptive response (e.g., by splinting).

Part II (Dynamic) Ultrasound in the clinical assessment of the CTS patient

In the second part of this thesis, the central topic was the added value of ultrasound (US) in CTS patient care. The static measurement of the cross-sectional area of the nerve has been the primary topic of studies published so far, but increased interest has also developed for dynamic assessments. Both in transverse as well as longitudinal planes, the median nerve, the flexor tendons, and the SSCT can be visualized using standard brightness-mode ultrasound. CTS patients show decreased nerve motion compared to controls, and dynamic assessment might thus hold valuable information for the prognosis.

Recently, a speckle tracking algorithm, enhanced with a singular value decomposition filter to reduce noise from background signals, was validated to track flexor tendon and SSCT motion with finger flexion and extension. **Chapter 4** assessed the reliability of these measurements in twenty-two CTS patients as well as sixteen healthy controls. Intra- and inter-class correlation coefficients for both groups were good to excellent. Higher reliability was found for tendon tracking compared to the SSCT. We concluded that speckle tracking with SVD can reliably analyze longitudinal excursion of these carpal tunnel structures.

Directly following these findings, speckle tracking was applied to a clinical CTS cohort of 90 patients in **Chapter 5** to test the association between relative SSCT movement (shear index) and CTS severity. An increase in shear index was found (equals a decrease in SSCT movement) with increasing CTS severity as quantified by neuroconductive tests. There was no association present when categorizing severity with patient-reported symptom scores or with clinical outcome after surgery. Limitations of this study included the overall good response to surgery, complicating discrimination between responders and non-responders.

The measurements described in chapters 4 and 5 were based on the (relative) movement in the longitudinal plane. More commonly, US acquisitions are done in the transverse plane since this is more practical and allows for the quantification of the cross-sectional area of the nerve. It is known that nerve area decreases after intervention, but currently, less is known about the changes in nerve dynamics or the correlation with clinical outcome. In **Chapter 6** we describe a prospectively followed CTS cohort of 85 patients. After surgery, the median nerve decreased significantly in size and moved more in dorsal direction. Although a larger nerve area at baseline was significantly associated with better functional scores, this was not found for symptomatic relief. Additionally, none of the dynamic baseline parameters showed an association with clinical outcome.

As the final chapter in part II, we presented a novel US technique in **Chapter 7**. This study was designed to assess the usability of shearwave elastography in the carpal tunnel context. Elastography was compared to a classic lab-based indentation test while measuring under incremental longitudinal nerve stress, with different probe orientations and positions. Compared to the indentation test, elastography showed a statistically comparable increasing pattern of elasticity. No significant interference of the osseous

structures was found. However, we did find a lower elasticity when measuring the nerve in the transverse plane compared to a longitudinal assessment. This is relevant since in clinical practice and some of the studies published so far, measurements were done only in the transverse plane.

Part III Improving CTS treatment outcome

The current guidelines recommend treating CTS with either splinting, a local steroid injection, and/or surgical intervention based on the clinical presentation sometimes in conjunction with electrophysiological results. Surgery is seen as the only definitive solution since studies have shown that injections in the majority of patients only provide temporary relief. The usage and role in CTS treatment of injections, therefore, remains a topic of debate. To improve clinical outcome after injection, the procedure is now also done under ultrasound guidance. Hydrodissection is an injection technique in which, while ultrasound-guided, anatomical planes are created by targeting the injection between the nerve and the transverse carpal ligament and around the nerve. Some studies indicate a beneficial effect of this technique, but so far it has not yet been compared with standard clinical practice. In **Chapter 8** we used a double-blinded, randomized pilot study to show equal satisfaction rates for those with and without hydrodissection therapies. Both groups showed symptomatic and functional improvement after one and six months, but in the preliminary data no clinical superiority was found.

In **Chapter 9** we discussed a new surgical approach as an alternative to the classic transverse carpal ligament release. We describe an US-guided carpal tunnel release in which an abrasive thread is looped around transverse carpal ligament thereby allowing the surgeon to divide the ligament without the open approach. The main advantages include minimal damage to surrounding tissue and short recovery periods while the required expertise with the US forms the main challenge. Future studies are necessary to fully assess the added clinical value.

Improved clinical outcome can also be approached by studying pre-therapeutic patient satisfaction. In **Chapter 10**, we describe a CTS database study in which we show that higher experience scores were associated with better symptom and function scores post-surgery. Despite the lack of a causal relation, this does indicate that there is an interplay between patient experience and what we are currently using as outcomes to determine the success of a surgery.

Finally in **Chapter 11**, we again looked at patient-reported outcomes, but this time we focused on the question of whether type of treatment (i.e., surgery or injection) would alter the minimum score necessary for patients to be satisfied after treatment. Even when corrected for baseline severity, patients who underwent injection required less improvement to reach the minimal clinically important difference threshold than those who underwent surgery. These results are directly relevant for researchers designing future studies with these instruments and, on a larger scale, the results infer that patient-reported outcomes are prone to external factors unknown to us.

14

CHAPTER 14.

Nederlandse samenvatting

Carpaletunnelsyndroom (CTS) is de meest voorkomende perifere neuropathie. Een karakteristieke bevinding bij CTS is verhoogde intra-carpaletunnel druk en daaropvolgende dysfunctie van de nervus medianus distaal van het polsgewricht. Typische symptomen bij presentatie zijn tintelingen en verlies van gevoel in met name de duim, wijs-, en middelvinger, maar ziekteprogressie kan ook leiden tot irreversibele zenuwschade en krachtverlies. Op basis van de ernst van de neuropathie wordt bepaald welke behandeling het beste is, maar de exacte pathofysiologie is complex en tot op heden is onduidelijkheid welke factoren een voorspellende waarde hebben over hoe het ziektebeeld na interventie zal verlopen. In dit proefschrift hebben we onderzocht wat de rollen zijn van het bindweefsel bij idiopathische CTS, hoe echografie kan helpen in het voorspellen van klinische uitkomst, en hoe we huidige behandelingen kunnen verbeteren.

Deel I Pathofysiologie van CTS en de rol van het bindweefsel

Een van de meest prominente kenmerken van CTS is de toename in intra-carpale druk, maar in de meeste gevallen is de oorzaak hiervan niet bekend. In het eerste deel is beschreven dat het onduidelijk is hoe CTS exact ontstaat maar dat het vaak in verband wordt gebracht met frequente hand- en vingerbewegingen. In de laatste decennia is er veel onderzoek geweest naar CTS en een van de etiologische modellen toont het bindweefsel in de carpale tunnel, het subsynoviale bindweefsel (SSCT), als prominente speler.

Het **tweede hoofdstuk** is een samenvatting over wat we weten omtrent de veranderingen in de mechanische reactie van zowel de nervus medianus als het omringende SSCT in de vorm van een review. CTS patiënten vertonen meer fibrose in het SSCT waardoor het minder meebeweegt tijdens flexie- en extensiebewegingen en onderhevig is aan additionele schade door blijvende shear stress. In dit artikel presenteerden we een hypothetisch model waarbij schade aan het SSCT aan de basis ligt van een zichzelf in standhoudende en versterkende pathologische feedbackloop. Het beter doorgronden van dit model kan helpen met de ontwikkeling van specifiek gerichte therapieën die bijvoorbeeld fibrosering van het SSCT tegengaan.

Het **derde hoofdstuk** bouwt hierop voort door te kijken naar de chronologische ontwikkeling van het SSCT in een diermodel. Middels elektronenmicroscopie hebben we kwalitatief de architectuur van de SSCT van prenatale tot jongvolwassene konijnen vergeleken. Hier zagen we dat de SSCT initieel een meer solide en dense structuur heeft maar een aantal weken na geboorte zich splitst tot een meerlagig platensysteem, verbonden door collageenfibrillen. Deze verandering vond plaats na fenotypische krachtdragende bewegingen en lijkt daarmee een proces van aanpassing (‘adaptive response’). Klinisch kan dit relevant zijn vanuit de gedachte dat CTS patiënten, waarbij schade aan het SSCT een rol speelt, meer baat zouden hebben bij een therapie die tijdelijk het aanpassingsvermogen van het SSCT kan stimuleren (zoals spalktherapie) dan aan een aanpak waarbij de fibrosering wordt tegengegaan.

Deel II (Dynamische) Echografie in de klinische evaluatie van de CTS patiënt

In het tweede deel stond de toegevoegde waarde van echografie voor CTS patiënten centraal. Tot nu toe heeft de statische meting van de zenuwgrootte de meeste aandacht in de literatuur gekregen, maar er is meer interesse voor de toegevoegde waarde van dynamische metingen. In zowel transverse als longitudinale vlakken kunnen de nervus, buigpezen, alsook het bindweefsel gemeten worden met echografie.

Met de recente verbetering van een speckle tracking-gebaseerde methodiek om relatief kleine structuren te volgen op bewegende echobeelden, zijn veranderingen in de dynamiek van de SSCT nu nauwkeuriger meetbaar. **Hoofdstuk 4** beschrijft deze eerste metingen in tweeëntwintig CTS patiënten en zestien controle personen en had als voornaamste doel om de herhaalbaarheid van de methode vast te leggen. De gevonden intraclass correlatiecoëfficiënten waren goed tot uitstekend in beide groepen. Tracking van de pezen was meer betrouwbaar dan de tracking van het SSCT.

Daaropvolgend kon in **hoofdstuk 5** de speckle tracking toegepast worden in een klinisch CTS cohort van negentig patiënten om te kijken naar een mogelijke associatie tussen relatieve SSCT beweging (shear index) en CTS ernst. Met toenemende elektrofysiologische zenuwschade werd er een grotere shear index gevonden (minder SSCT beweging). Er was echter geen associatie met baseline symptoomernst of uitkomst na chirurgie. De grote spreiding in shear index waardes en de algemeen positieve uitkomst van de operatie zouden hierbij een limiterende factor kunnen zijn geweest.

De metingen beschreven in hoofdstuk 4 en 5 waren gebaseerd op de (relatieve) beweging in het longitudinale vlak. Doorgaans zijn echografische metingen in het transverse vlak makkelijker uit te voeren en daarmee gangbaarder; deze opnames worden ook gebruikt bij het meten van de oppervlaktegrootte van de nervus medianus. Studies hebben laten zien dat de zenuwgrootte significant vermindert na invasieve behandelingen, maar er is minder bekend over de veranderingen in zenuwbeweging. In **hoofdstuk 6** beschreven we een prospectieve studie van vijfentachtig CTS patiënten. Na chirurgie nam de nervus medianus significant af in grootte en was er een toename in dorsale beweging. Een grotere zenuw voor operatie was geassocieerd met een grotere verbetering in functionele score, maar er was geen verband met symptoomscore. Tevens was er geen relevante associatie tussen zenuwbeweging en klinische uitkomst.

Als afsluitend onderzoek in deel II hebben we in **hoofdstuk 7** een nieuwe echotechniek getest. In dit hoofdstuk werd gekeken naar de haalbaarheid van het gebruik van shearwave elastografie in de carpale tunnel door deze echotechniek. Er werd vergeleken met een klassieke indentatiemethode onder toenemende longitudinale zenuw stress, met verschillende echokoporiëntaties en -posities. Elastografie liet een statistisch vergelijkbaar toename patroon zien met indentatie. Verstoring van het signaal door ossale structuren bleek minimaal. Er werd een lagere elasticiteit gevonden wanneer de acquisitie in het transversale vlak plaatsvond, vergeleken met longitudinale meting. Dit is relevant omdat doorgaans nervus metingen op de eerste methode worden gedaan.

Deel III Verbeteren van behandeling uitkomst

De huidige richtlijnen bevelen drie type behandelingen voor CTS aan: spalken, een lokale steroïde injectie, of een chirurgische interventie. De keuze van behandeling ligt bij de arts op basis van klinische presentatie met of zonder de resultaten van aanvullend onderzoek in de vorm van elektrofysiologische testen van de nervus en musculatuur. Over het algemeen wordt aangenomen dat enkel een operatie een definitieve oplossing is en dat injectie in de meerderheid van de mensen enkel tijdelijk werkt. De toegevoegde waarde van injecties is daarom een bekend onderwerp van discussie. Corticosteroïd injecties kunnen nu ook onder echobegeleiding worden gedaan. Hydrodissectie is een injectietechniek waarbij, onder echografische begeleiding, vloeistof wordt gebruikt om ruimtes tussen de nervus en het transverse carpale ligament dan wel het SSCT te creëren. Tot nu toe was dit nog niet als toevoeging op de huidige standaard zorg in de vorm van een corticosteroïd injectie getest. In hoofdstuk 8 beschreven we een dubbelblinde, gerandomiseerde pilot studie waar we lieten zien dat patiënten met of zonder hydrodissectie geen andere tevredenheid of pijnscores vertoonden. Beide groepen hadden symptomatische en functionele verbeteringen na één en zes maanden, maar zonder evidente klinische meerwaarde.

Hoofdstuk 9 beschreef een nieuwe chirurgische variant voor het doornemen van het carpale ligament. Een echografisch begeleide operatietechniek werd beschreven waarbij een draad middels een voernaald rondom het transverse carpale ligament gebracht wordt. De draad heeft een schurend effect wanneer er kracht op uitgevoerd wordt en kan zo door manipulatie van de chirurg het ligament doornemen. Voordelen zijn de minimale schade die peri-operatief gemaakt wordt en de zichtbaarheid die komt met constant echografische begeleiding. Toekomstige studies moeten de toegevoegde waarde voor de patiënt kwantificeren.

Naast verbeteringen aan de interventietechnieken zelf, is het ook van waarde om te onderzoeken wat we rondom de zorg van de CTS patiënt kunnen verbeteren. Patiënttevredenheid als uitgangspunt werd in **Hoofdstuk 10** beschreven middels een databasestudie. Hierin vonden we dat drie maanden na de ingreep hogere tevredenheid met arts-patiënt communicatie en informatie over de behandeling significant geassocieerd waren met een betere chirurgische uitkomst. Hoewel deze studie, door de methodologische opzet, geen causaliteit kon aantonen, geeft het wel aan dat patiëntervaringen niet losstaan van de uitkomsten van de behandeling.

In **hoofdstuk 11** hebben we ten slotte wederom gekeken naar uitkomsten gebaseerd op patiëntenrapportage, maar in deze studie wilden we toetsen of CTS patiënten die een injectie krijgen een andere symptoomscore moeten hebben om dezelfde tevredenheid te halen als zij die een chirurgische ingreep ondergaan. Zelfs na baseline correctie vonden we dat injectiepatiënten inderdaad minder verbetering in score nodig hebben om een minimaal klinisch relevant verschil te bereiken. Dit is relevant voor wetenschappers die studies op basis van deze uitkomsttools ontwerpen en, op grotere schaal, om aan te tonen dat patiënt gerapporteerde uitkomsten onderhevig zijn aan factoren die eerder niet bekend waren.

A

APPENDICES

List of publications

Schrier, V. J., Evers, S., Geske, J. R., Kremers, W. K., Villarraga, H. R., Kakar, S., ... & Amadio, P. C. (2019). Median Nerve Transverse Mobility and Outcome after Carpal Tunnel Release. Ultrasound in medicine & biology, 45(11), 2887-2897.

Schrier, V. J., Evers, S., Bosch, J. G., Selles, R. W., & Amadio, P. C. (2019). Reliability of ultrasound speckle tracking with singular value decomposition for quantifying displacement in the carpal tunnel. Journal of Biomechanics, 85, 141-147.

Schrier, V. J., Gelfman, R., & Amadio, P. C. (2019). Minimal clinically important difference is lower for carpal tunnel syndrome patients undergoing injection versus surgery. Journal of Hand Surgery (European Volume), 45(1):90-92.

Schrier, V. J., Lin, J., Gregory, A., Thoreson, A. R., Alizad, A., Amadio, P. C., & Fatemi, M. (2020). Shear Wave Elastography of the Median Nerve: A Mechanical Study. Muscle & Nerve [epub ahead of print]

Schrier, V. J., Poelstra, R., Selles, R. W., Slijper, H. P., Amadio, P. C., Hovius, S. E., & Porsius, J. T. (2019). Better patient-reported experiences with health care are associated with improved clinical outcome after carpal tunnel release surgery. Plastic and reconstructive surgery, 143(6), 1677-1684.

Schrier, V. J., Vrieze, A., & Amadio, P. C. (2020). Subsynovial connective tissue development in the rabbit carpal tunnel. Veterinary Medicine and Science [epub ahead of print].

Schrier, V. J., Brault, J. S., & Amadio, P. C. (2020). Ultrasound-Guided Hydrodissection With Corticosteroid Injection in the Treatment of Carpal Tunnel Syndrome: A Pilot Study. Journal of Ultrasound in Medicine.

Festen-Schrier, V. J. M. M., & Amadio, P. C. (2018). The biomechanics of subsynovial connective tissue in health and its role in carpal tunnel syndrome. Journal of Electromyography and Kinesiology, 38, 232-239.

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Browne*, J. L., **Schrier*, V. J.**, Grobbee, D. E., Peters, S. A., & Klipstein-Grobusch, K. (2015). HIV, antiretroviral therapy, and hypertensive disorders in pregnancy: a systematic review and meta-analysis. Journal of Acquired Immune Deficiency Syndromes, 70(1), 91-98. *Shared first author.

Soepnel, L. M., Norris, S. A., **Schrier, V. J.**, Browne, J. L., Rijken, M. J., Gray, G., & Klipstein-Grobusch, K. (2017). The association between HIV, antiretroviral therapy, and gestational diabetes mellitus. Aids, 31(1), 113-125.

Portfolio

PhD student: V.J.M.M. Schrier
PhD period: 2016-2020
Promotor: Prof.dr. S.E.R. Hovius
Co-promotor: Dr. R.W. Selles

1. PhD Training	Year	Workload
Courses and Training		
Human Subject Protection training program, *Mayo Clinic*	2017	0.7 ECTS
Microsurgery course, *Mayo Clinic skills lab*	2017	1.4 ECTS
BME 6302 Ultrasound tutorial, *Mayo Graduate School of Biomed Sciences*	2017	2 ECTS
CTSC 5020 Regulatory issues in clinical research, *Mayo Graduate School of Biomed Sciences*	2017	1 ECTS
CTSC 5600 Statistics in research and translational research, *Mayo Graduate School of Biomed Sciences*	2018	2 ECTS
CTSC 5601acc Utilizing statistics in clinical research, *Mayo Graduate School of Biomed Sciences*	2018	1 ECTS
Summer school Advances in Hand Research, *Erasmus MC*	2018	1.4 ECTS
CORE 6000 Responsible conduct of research, *Mayo Graduate School of Biomed Sciences*	2019	1 ECTS
Hands-on Ultrasound for MSK application, *American Society for Surgery of the Hand*	2019	0.3 ECTS
Online courses		
Introduction in programming in Matlab, *VanderBilt University*	2017	2 ECTS
Programming with R, *Datacamp*	2018	0.5 ECTS

2. Conferences and Presentations	Year	Workload
Attendance research and hand surgery grand rounds, Mayo Clinic	2016-2020	2 ECTS
Podium presentations		
Ultrasound Research Center Annual Symposium, Mayo Clinic *Shearwave elastography and median nerve stiffness, a mechanical Study*	2018	15 hours
Patrick J. Kelly research day, Mayo Clinic *Median nerve mobility in carpal tunnel syndrome: changes and prediction after surgery*	2018	15 hours
Convocation of thanks anatomy department, Mayo Clinic *How the donor program shaped my research on carpal tunnel syndrome*	2019	5 hours
Invited by the Federation of the European Societies for Surgery of the Hand, annual meeting, Basel *Minimal clinically important difference is lower for carpal tunnel syndrome patients undergoing injection versus surgery*	2020	-
Poster presentations		
Orthopedic Research Society annual meeting, San Diego *Ultrasound-guided Hydrodissection Decreases Gliding Resistance of the Median Neve within the Carpal Tunnel.*	2017	10 hours
American Association Clinical Anatomists annual meeting, Minneapolis *The Biomechanics of Subsynovial Connective Tissue in Health and Its Role in Carpal Tunnel Syndrome*	2017	10 hours
Orthopedic Research Society annual meeting, New Orleans *Reliability of Ultrasound Speckle Tracking with Singular Value Decomposition for Quantification of Structure Displacement in the Carpal Tunnel*	2018	10 hours

Gordon Research Conference, biomechanics meeting, New Hampshire *Ultrasound Shear Wave Elastography validation in the Carpal Tunnel*	2018	10 hours
American Association for Hand Surgery annual meeting, Boston *Better Patient-Reported Experiences with Health Care are associated with improved Clinical Outcome after Carpal Tunnel Release Surgery*	2018	10 hours
Orthopedic Research Society annual meeting, Austin *Median Nerve Mobility after Carpal Tunnel Release and the Relation with Patient-Reported Outcome*	2019	10 hours
American Society for Surgery of the Hand annual meeting, Las Vegas *Ultrasound Changes in Connective Tissue Motion and Carpal Tunnel Syndrome Severity*	2019	10 hours
Orthopedic Research Society annual meeting, Phoenix *Ultrasound Shear Wave Elastography as a New Imaging Technique for Carpal Tunnel Syndrome: A Mechanical Study*	2020	10 hours

3. Professional Development	Year	Workload
Teaching & additional tasks		
Supervision of minor students, Erasmus MC *Diagnostic value of ultrasound elastography in carpal tunnel syndrome: A systematic review.*	2017	15 hours
Supervision of medical students, Mayo Clinic o *Shearwave elastography validation project* o *Oophorectomy as a risk factor for carpal tunnel syndrome*	2018-2019	60 hours
Creator and organizer of monthly research meeting "Fellow Huddle" for the Tendon and Soft Tissue Biology lab.	2018-2020	15 hours
Peer reviewed >20 articles for several medical journals	2017-2020	40 hours

Medical improvisation		
Teacher in monthly classes and workshops	2017-2020	5 hours
Oral presentations o Professionalism & Values seminar series: Mayo Clinic Values and Applied Improvisation o Musculoskeletal Department noon conference o Education & Technology Forum o St Catherine's University, Symposium for Art and Health partnerships in St. Paul	2018-2019	30 hours
Completed course "Medical Improv for healthcare professionals", Northwestern University Chicago	2019	40 hours

Curriculum Vitae

Verena Schrier was born in Rotterdam, the Netherlands on August 30th 1989. She grew up with her two younger siblings in Capelle a/d Ijssel but returned later to Rotterdam to eventually graduate from Emmauscollege in 2007. She started a bachelor in Biomedical Sciences at Utrecht University, which she completed in 2010. Then, after a year of research as a master's student in the field of experimental cardiology, she decided that bench life alone was not going to be enough, so she applied and got accepted at Utrecht's Selective Medical Master (SUMMA); this four-year program allowed her to obtain her medical degree while still being actively involved in research. Under the supervision of Dr. Joyce Brown and Prof. Klipstein-Grobusch she worked on several projects concerning maternal health in middle and lower income countries and the added value of ultrasonography in the assessment of cardiovascular risk in HIV-exposed infants. To complete these global projects, Verena traveled to Korle Bu Teaching Hospital in Ghana, to Australia to train in Melbourne's Royal Children's Hospital, and finally to Johannesburg in South-Africa to the University of Witwatersrand between 2014-2015.

She graduated from SUMMA fall of 2015 and with the desire to work in the surgical field as well as to gain experience outside of the hospital, she stayed local for a year working in Amsterdam for a company focused on using medical videos to train surgeons (inter) nationally. Looking to move on to new challenges while continuing clinical research, she was presented with an opportunity to once again work abroad on ultrasonography in 2016; She obtained a PhD position under the supervision of prof.dr. S.E.R. Hovius and prof. P.C. Amadio through a collaboration between Mayo Clinic Rochester, Minnesota and Erasmus Medical Center. Verena and Raymond packed their bags, got married, and moved to the United States of America, where they both worked in the Orthopedics department. During her traineeship, she focused on medical imaging in carpal tunnel syndrome and realized that a career not in surgery but radiology would be the best fit for her. After returning to the Netherlands in September 2019, she was accepted to start her radiology residency at the Radboud Medical Center in Nijmegen where she is currently enjoying her training (prof. dr. Prokop).

Dankwoord

"I wish there was a way to know you're in the good old days before you've actually left them."

- The Office S09E25

Met beide voeten weer terug in Nederland is het afronden van het promotietraject ten dele een aaneenschakeling van herinneringen geweest aan de prachtige tijd in Rochester. Bij deze is hier de kans om iedereen te bedanken die meegeschreven, gedacht, of anderszins geholpen heeft aan de thesis voor u. Zonder jullie steun was het niet gelukt.

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Jon, thanks for the fish and mushrooms. You showed me that you can be an adult without having to be mature. Love to Marie and Daniel.

Dear Dr. Fatemi, thank you for investing all that time in me as your solo student during the Ultrasound Tutorial course in 2017. Thank you and Dr. Alizad for welcoming us into your home and for showing me how cool radiology is and that jokes around a campfire can be first-class entertainment.

Stefanie, Julia, and Mohammad, I find myself between you three in the DUCATS legacy and I am incredibly grateful for all your help in the writing of the articles. All three of you guys are incredibly clever and efficient people and you made my work that much easier.

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Lieve nieuwe collega's in het Radboud. Ik kan zelf nog steeds niet helemaal geloven dat ik onderdeel van jullie groep mag zijn en ik weet zeker dat in de jaren te komen ik een enorme collectie aan gênante foto's van jullie ga opbouwen, dus pas op!

Alle vrienden thuis bedankt voor het bewijzen dat uit het oog niet uit het hart hoeft te betekenen. Elise, dankzij jou hebben we een dijk van een afsluiter aan het VS hoofdstuk kunnen geven ("kijk maar even naar een filmpje met Corgi kontjes"). Marit, vanaf nu zal ik weer bij alle grote momenten zijn, dankjewel voor alle fijne skype calls.

Lieve Eef, vanaf SUMMA coschappen trouw samen. Je bent m'n reisbuddy ("ja, laten we de woestijn in gaan"), m'n carriere coach ("ik zou je prof heel snel bellen"), en mijn ik-bel-je-zo-maar-even-op vriendinnetje ("laten we een cactus halen"). Onze levens lopen vrijwel parallel en ik wacht nog op de dag dat ik jou kan steunen zoals jij mij hebt gesteund de laatste jaren. Paranimf voor 2020, maatje voor het leven.

Ad en Suze, jullie waren de eerste die de grote overtocht maakten en met ons het plezier van Chicago deelden (6 uur met ons in een auto is een applausje waard).

Angeline en Peter, dankjulliewel voor de tijd die jullie me gunden met Raymond zo ver weg. Het was een grote reis over de plas en het feit dat jullie meteen de eerste avond met onze voltallige collega-groep naar een honkbalwedstrijd zijn gegaan doe ik mijn petje voor af!

Lieve opa en oma v. Woerkom, een van de moeilijkste dingen om thuis te laten was dat we nu niet meer snel even konden langskomen of babbelen. Goede nieuws is wel dat ik nu vouwkundig ben.

Marissa en Freek, dankjewel voor het staande houden van het thuisfront. We zullen snel na de COVID-19 misère een kopje koffie doen in het nieuwe huis.

Beata en Niels, wat een ontzettend leuke verrassing dat jullie langs kwamen! Beetje spikeball, beetje dobberen, en beetje spelletjes doen was precies wat ik nodig had in die chaotische eindfase.

Mama en papa. Wat een achtbaan moet dat zijn geweest; van "hoera, we gaan trouwen!..." naar "…en over 3 maanden vertrekken we naar de VS". Toen werd het ook nog eens een jaar langer dan gepland én moesten jullie langskomen. Een tocht over de grote oceaan, dat was een avontuur voor iedereen. Ik heb er in ieder geval toffe herinneringen aan overgehouden (en zorg dat er nu altijd koffiemelk is), want jullie waren werkelijk overal voor in! Nu blijven we in Nederland, dichtbij elkaar…tenminste…als de auto het doet… Dankjulliewel voor de onvoorwaardelijke liefde en steun. Ik weet dat jullie altijd voor me klaarstaan.

Raymond, wederhelft in het leven en paranimf voor nu. Niemand heeft zo veel rollen in mijn leven als jij. Je hebt naast me gewerkt in Mayo en inhoudelijk steun geboden als ik voor de zoveelste keer vastzat met Matlab. Met elk congres was je mee om van de extra tijd ook vrije tijd te maken. Je was m'n vaste badmintonpartner en de co-regisseur van de improgroep. Onze dagen liepen geheel parallel, met elkaar, door elkaar. Dankjewel dat je met beide benen in het avontuur bent gestapt. Samen maken we er nog meer. I love you and I like you.

