End-to-Side Nerve Transfer: An Evaluation of Its Efficacy and Functional Impact

by

Simon Wu

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Centre of Neuroscience

University of Alberta

Abstract

Background

Peripheral nerve injury is common, effecting 3% of the population. While surgery can be effective in moderate cases, complete neurologic and functional recovery are often not possible in severe cases of proximal nerve injury. Poor outcomes are attributable to the long-distance nerves must regenerate to reach their targets. End-to-end (ETE) nerve transfer surgery can shorten the distance of regeneration by bridging a dispensable donor nerve to the end of the injured nerve that is closer to the denervated target. Unfortunately, these procedures involve cutting the injured nerve, preventing the possibility for native nerve regeneration, and making in unfeasible for incomplete injuries. Reverse end-to-side (RETS) nerve transfers is an increasingly utilized technique that involves connecting the donor nerve to the side of the injured nerve, which preserves the injured nerve continuity, and potentially allows for donor nerve (1) axonal crossover and the (2) babysitting effect. However, the source of regenerating nerve fibres in the RETS transfer has been inconsistent with some studies that show benefits and others that did not find efficacy in the surgery.

Objective

To evaluate the amount of (1) axonal crossover from the donor nerve in the RETS transfer using a novel electrophysiology technique. To evaluate the (2) babysitting effect by comparing the RETS transfer to a decompression surgery.

Aim 1 — A novel electrophysiological technique to quantify axonal crossover.

Seven Martin-Gruber anastomosis (MGA) and nine anterior interosseous nerve (AIN) to ulnar nerve ETE nerve transfer patients were recruited. Motor nerve conduction studies were performed, and the novel digital subtraction technique was compared against the collision technique

and innervation ratio method, previous techniques to measure crossover. The digital subtraction method was highly correlated with the collision technique and has several practical advantages. With the increasing use of nerve transfer surgery in severe high ulnar nerve injury, this could be a useful method to identify the presence of MGA prior to surgery and for evaluating nerve recovery following surgery.

Aim 2 — A prospective clinical trial comparing RETS with ETE and decompression surgery.

Sixty-two subjects (RETS=25 | ETE=16 | decompression=21) from four centres in Western Canada were enrolled. All subjects with severe ulnar nerve injury had nerve compression at the elbow except 10 in the ETE group had nerve laceration or traction injury. The novel digital subtraction technique was used to quantify the regeneration of AIN and ulnar nerve fibers while functional recovery was evaluated using key pinch and Semmes-Weinstein monofilaments. The subjects were followed post-surgically for 3 years. Post-surgically, no reinnervation from the AIN to the abductor digiti minimi muscles was seen in any of the RETS subjects.

Significance

While clinical translation of RETS has been increasing, the results from published clinical trials has been conflicting, in part because crossover regeneration from the donor nerve has never been measured. From applying the novel electrophysiological technique in the multicentre prospective study, we found there was no crossover regeneration in patients that underwent RETS compared to ETE nerve surgery. The extent of reinnervation from RETS surgery was also no different compared to decompression surgery alone. Based on these findings, the justification for the RETS surgical technique needs to be further evaluated.

Preface

This thesis is an original work by Simon Wu. The research project was developed with the guidance of Dr. K Ming Chan.

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I contributed to developing the novel method and co-created the manuscript. MWT Curran, A Hachisuka, and M Rajshekar contributed to manuscript creation. KM Chan was the senior author and was involved in conceptualizing the novel method, study design, data analysis, and manuscript creation.

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I contributed to the data analysis, tables, figures, and manuscript. MWT Curran and KM Chan, the senior author, were involved with study design, data analysis, and manuscript creation. JL Olson, MJ Morhart, and R Midha performed the nerve transfer surgeries. MJ Berger contributed to the data analysis and manuscript.

This is a paper-based dissertation. Chapters II and III are published articles in,

Muscle & Nerve and *Neurosurgery* in which I was the first and fourth author respectively.

" The people who are crazy enough to think they can change the world, are the ones who do. "

— *Steve Jobs (1997)*

Acknowledgements

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Abbreviations

2PD	Two-Point Discrimination
ADM	Abductor Digiti Minimi Muscle
AIN	Anterior Interosseous Nerve
CMAP	Compound Muscle Action Potential
CNS	Central Nervous System
CT	Computerized Tomography
ETE	End-to-End
ETS	End-to-Side
FDI	First Dorsal Interosseous Muscle
MGA	Martin-Gruber Anastomosis
MRI	Magnetic Resonance Imaging
MU	Motor Unit
MUNE	Motor Unit Number Estimation
NGF	Nerve Growth Factor
PNS	Peripheral Nervous System
RETS	Reverse End-to-Side
STS	Side-to-Side
SWMT	Semmes-Weinstein Monofilament Test

Chapter 1

Introduction

1.1 The Peripheral Nervous System

1.1.1 Anatomy of the Human Nervous System

The human nervous system is composed of the central nervous system (CNS), containing the brain and spinal cord, and the peripheral nervous system (PNS), all other nerves. The peripheral nervous system connects to the brain and spinal cord to the end target muscle and sensory organs throughout the body via a network of cranial nerves and spinal nerves, giving a total of 43 motor and sensory nerve pairs. The peripheral nervous system is divided into sensory and motor nerves. Sensory nerves carry afferent signals to the CNS, transmitting information from skin receptors, muscle, tendon, and joints. Motor nerves send efferent signals from the brain, carrying information to the end plates of skeletal muscle to enable muscular contraction. The motor division is divided into the somatic voluntary skeletal muscle system, and the autonomic involuntary smooth muscles of the heart, blood vessels, glands, and organs. The autonomic nervous system is further delineated by the sympathetic division of fight and flight, and the parasympathetic division of rest and digest.

1.1.2 The Neuron

The cell carrying electrical signals in the nervous system is known as the neuron. While there are different types of neurons which differ in morphology and function, anatomically, all neurons contain (1) a cell body, where the nucleus is, (2) dendrites, whose branching projections extent to detect stimuli from other neurons or the environment, (3) the axon, the longitudinal process which conducts electrical signals and transports neurotransmitters to the downstream synapse, and (4) nerve terminal, where neurotransmitters are released at the synapse with another nerve or target. The cell bodies of sensory neurons reside in the dorsal root ganglia which are nodules adjacent to the spinal cord. In contrast, the cell bodies of motor neurons exist in the ventral horn of the spinal cord.

1.1.3 The Neuroglia

Neuroglia are cell lines which support the function of neurons and are found in the CNS and PNS. Unique to the CNS are astrocyte glial cells which maintain and form the blood-brain barrier. The oligodendrocytes and microglia found in the CNS are equivalent to the Schwann cells and macrophages found in the PNS. Notably, Schwann cells in the PNS have a 1:1 ratio in myelinated axons, while oligodendrocytes in the CNS myelinate multiple neuronal axons.

1.1.4 Connective Tissue

In the PNS, axons of the large neurons are myelinated. In contrast, axons of the smaller neurons are either thinly myelinated or in the case of group c sensory nerves and sympathetic nerve fibres, small groups of axons are enwrapped by non-myelinating Schwann cells forming the Remak bundles. Myelin is a phospholipid membrane that wraps concentrically around axons to provide electrical insulation. The consecutive zones of myelin sheaths form Nodes of Ranvier in between each sheath. These nodes, which are electrically conductive, enable the rapid depolarization across the length of the axon, enabling faster rates of nerve conduction. Myelin is produced by Schwann cells in the peripheral nervous system and oligodendrocytes in the central nervous system. Bundles of connective tissue encompass and protect an axon and its myelin (see Figure 1.2). The endoneurium is a delicate layer of connective tissue which forms an endoneurial tube around each myelinated nerve fibre. A cluster of endoneurial tubes destined for the same end target are bundled together by a thicker perineurium tissue, leading to the formation of nerve fascicles. Nerve fascicles, arteries, and veins are protected by a fibrous epineurial tissue. Together, this forms the entity of a nerve trunk that can be purely sensory or contain mixed motor and sensory axon fibres.
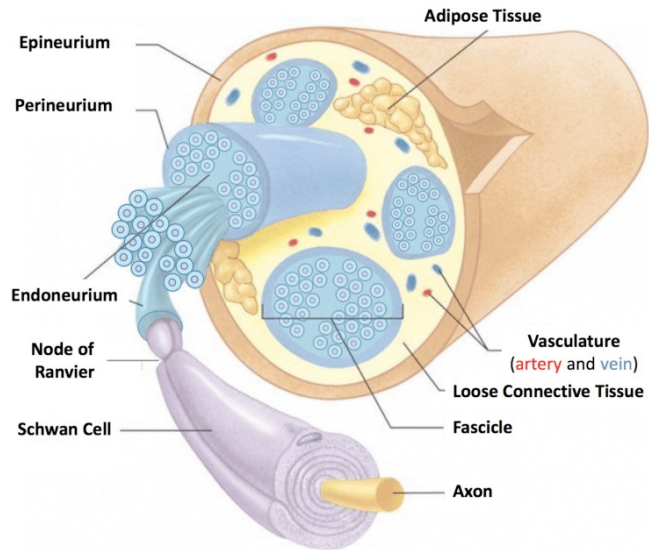
FIGURE 1.1 CROSS SECTION OF A PERIPHERAL NERVE.

Bundles of endoneurium-wrapped axons for a fascicle. Vasculature, adipose tissue, and mesenchymal cells lie between fascicles. [(Kuliasha et al., 2018) Solid-State, Actuators, and Microsystems Workshop.]

1.1.5 The Somatic Motor System

Upper motor neurons originate from the precentral gyrus, or primary motor cortex, of the brain. These neurons descend from the primary motor cortex through the internal capsule of the midbrain, to reach the medullary pyramids. At this location, motor nerve fibres cross to the contralateral side and descend into the white matter of the lateral corticospinal tract of the spinal cord. Motor neuron cell bodies of peripheral nerves arise from the grey matter of the spinal cord ventral horn. Peripheral nerves exit the spinal cord through the spinal roots where the terminal branches will synapse with the motor end plates in the muscle. Together, a single motor axon with the innervated muscle fibres forms a motor unit. Depending on the muscle, a single motor axon can innervate a few muscle fibres up to the thousands. The fewer muscle fibres innervated, the more precise the motion produced by that muscle contraction. In contrast, large motor units produce

powerful, gross movements. Groups of motor units can act in concert to produce muscle contraction and generate complex coordinated limb motion.

There are two types of motor neurons, α- and γ-motor neurons. The former is responsible for the initiation of muscle contraction through innervation of extrafusal muscle fibres in skeletal muscle. The latter innervates intrafusal skeletal muscle fibres and is not directly involved in muscle contraction, but rather regulates the activation of α-motor neurons that tunes the tension of intrafusal muscle fibres. The activation of γ-motor neurons will produce feedback loops to adjust the speed and magnitude of muscle contraction.

1.1.6 The Somatosensory System

The sensory system innervates a vast variety of organs enabling an abundance of conscious and unconscious sensory information. The somatosensory system represents the conscious portion of the senses. The regions of sensation across the body are anatomically specific, as represented by the homunculus in the primary sensory cortex with the lips and hands have the greatest afferent inputs. Somatic sense includes touch, pain, pressure, proprioception, temperature, and vibration. Many of these afferent signals use specific sensory receptors located at varying depths in the skin while senses such as pain, are detected by free nerve endings.

1.1.6A Receptor Types

Touch sensation is conveyed differently for different skin types. In non-glabrous, hairy skin, hair follicles act as the primary mechanoreceptor. In glabrous, non-hairy skin, four receptors are involved with conveying touch. (1) Merkel cells in the epidermis detect form and texture for tactile discriminations. (2) Meissner corpuscle in the dermis detect light touch and vibration. (3) Ruffini endings in the dermis are poorly understood but may play a role in detecting skin stretch. (4) Pacinian corpuscles in the subdermal tissue detect vibration.

The function and classification of these receptors is based on the (1) size of their receptor field and their (2) rate of sensory adaptation.

(1) Receptors located deeper in the skin have larger receptive fields. For this reason, Meissner corpuscles and Merkel cells have small receptive fields, while Ruffini endings and Pacinian corpuscles have large receptive field sizes.

(2) Meissner and Pacinian corpuscles are rapidly adapting receptors as they respond to any change in stimulus, while Merkel cell and Ruffini endings are slow to adapt.

The sensory signals generated from these receptors are transmitted to afferent fibres, up the dorsal column–medial lemniscus pathway and anterolateral system in the spinal cord, and into the postcentral gyrus (or primary somatosensory) cortex of the brain where the information is processed.
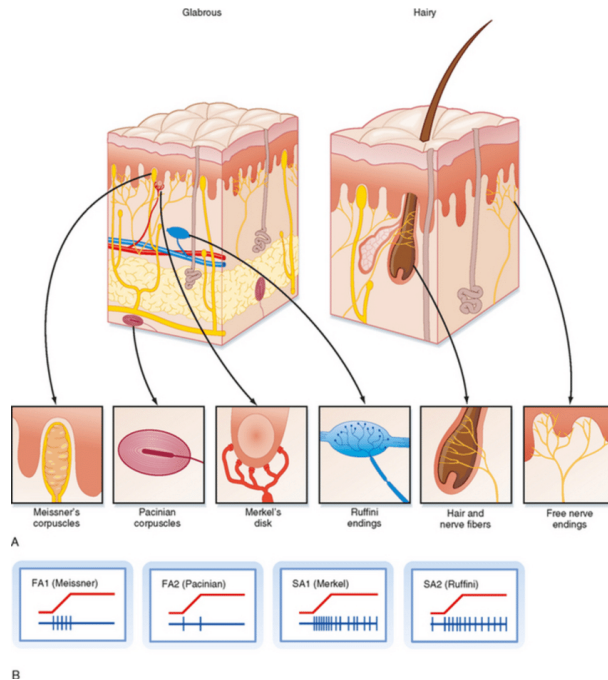
FIGURE 1.2 THE MECHANORECEPTORS IN THE SKIN.

Merkel cells, Meissner corpuscles, Ruffini's corpuscles and Pacinian corpuscles all transmit sensory information through Aβ figures. Free nerve endings transmit temperature and pain signals through Aδ and C fibres. [Purves (2008) Neuroscience.]

1.1.6.B Afferent Fibre Types

There are four types of axons that carry afferent sensory information from the PNS to the CNS. Axons from skin are classified as Aα, Aβ, Aδ, and C fibres while sensory axons from muscles are classified as: Group I, II, III, and IV fibres. Afferent axon classification and function are determined by their diameter and extent of myelination. Larger axons with greater myelination enable faster electrical conduction velocity.

(1) Aα (Group Ia & Ib) fibres carry proprioceptive information generated in muscles and joints. Group Ia fibres convey muscle length signals from muscle spindle fibres, while

Group Ib fibres convey muscle force signals from Golgi tendon organs. They have axon diameters of 13-20 µm and conduction velocities of 80-120 ms^{-1}.

(2) Aβ (Group II) fibres carry touch, pressure, and vibration information. They have axon diameters of 6-12 µm and conduction velocities of 35-75 ms^{-1}.

(3) Aδ (Group III) fibres carry temperature and pain information. Unlike the previous two afferent fibres, these fibres do not carry signals generated from receptors, rather, have free endings in the dermis. They have axon diameters of 1-5 µm and conduction velocities of 5-30 ms^{-1}.

(4) C (Group IV) fibres carry temperature, pain, and itch information. Like Aδ fibres, afferent signals are generated from free nerve endings, however, these nerve endings are found in the epidermis rather than the dermis. This is the only afferent fibres that is unmyelinated. They have axon diameters of 0.2-1.5 µm and the conduction velocities of 0.5-2 ms^{-1}.

An understanding of the somatic motor and somatosensory system enables the use of different electrodiagnostic methods and functional sensory testing. Based on conduction amplitudes, latency, and velocity, this testing can be employed to diagnose nerve injuries and follow the progress of recovery after a nerve injury.

1.2 Epidemiology of Peripheral Nerve Injuries

1.2.1 Peripheral Nerve Injury Prevalence

Peripheral nerves are fragile tissues that can be easily damaged. Damage to the peripheral nerve is known as peripheral neuropathy and can originate from numerous pathologies. Traumatic injury often leads to poor outcomes for patients due to the potential for incomplete recoveries. It is

often distressing and can be severely debilitating. In one of the largest studies of its kind, the incidence of peripheral nerve injuries presenting to a tertiary care centre in Ontario was identified in 2.8% of patients (Noble, Munro, Prasad, & Midha, 1998).

1.2.2 Classification of Peripheral Nerve Injury Severity

Peripheral neuropathy can occur through demyelinating processes where nerve signal transduction is affected because the myelin coating on axons deteriorates or fails to form properly. Neuropathy can also be caused by axon degeneration. Peripheral nerve injuries are commonly classified using the Sunderland classification system (Sunderland, 1951) (see Figure 1.1).

First degree is neuropraxia, a local reversible conduction block caused by damaged myelin.

Second degree is axonotmesis, a loss of axonal continuity within the nerve caused by axon injury.

Third degree are 2nd degree with endoneurium damage.

Fourth degree are 2nd degree with endoneurium and perineurium damage.

Fifth degree are 2nd degree with endoneurium, perineurium, and epineurium damage.

While the first-degree injuries are non-degenerative with potential for quick and full recovery, the latter stages are classified as axonotmesis (second to fourth degree) and lastly neurotmesis (fifth degree) the complete transection of the nerve. These nerve injuries are degenerative and require immediate surgical nerve repair. Extent of nerve damage is primarily diagnosed using electromyography and nerve conduction tests. Imaging techniques such as CT scan, MRI, and MRI neurography can also provide useful diagnostic information, especially for surgical planning.
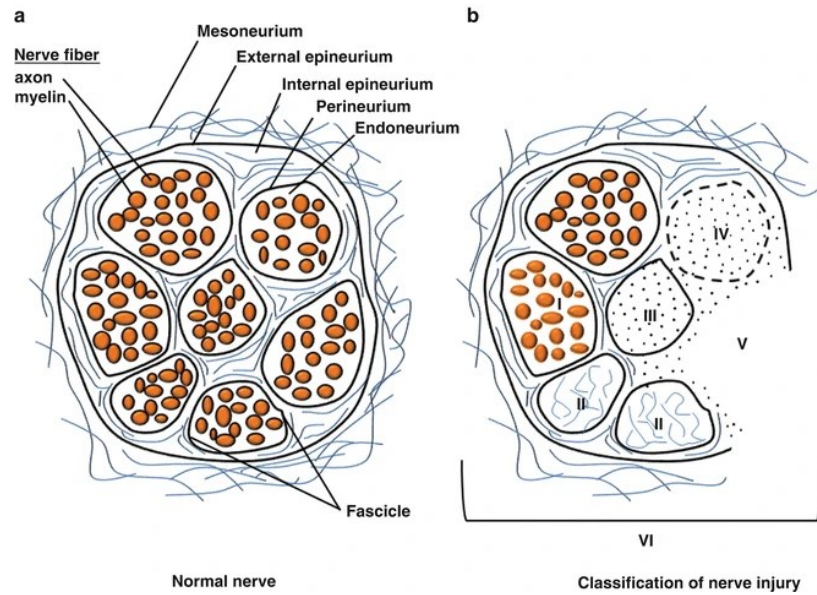
FIGURE 1.3 SUNDERLAND CLASSIFICATION FOR NERVE INJURY.

[(Snyder-Warwick, Yee, & Mackinnon, 2017) Esophageal and Gastric Disorders in Infancy and Childhood.]

1.2.3 Types of Peripheral Nerve Injuries

1.2.3A Compression nerve injuries

While the prevalence of compression neuropathies is not well understood, the annual incidence of common compressive neuropathies is estimated to be 491 cases per 100,000 new presentations in primary care (Latinovic, Gulliford, & Hughes, 2006). With carpal tunnel syndrome, compression of the median nerve at the wrist, being the most common with 281 per 100,000 cases. Other entrapment neuropathies include cubital tunnel syndrome (compression of the ulnar nerve at the elbow), radial tunnel syndrome (compression of the radial nerve in the forearm), meralgia paraesthetica (compression of the lateral femoral cutaneous nerve at the hip), tarsal tunnel syndrome (compression of the tibial nerve at the ankle), and Morton's metatarsalgia (compression of the interdigital nerves in the metatarsals). Compression neuropathies can cause motor and sensory

deficits that affect quality of life, and in severe cases, may require surgical intervention. Severe entrapment neuropathies cause demyelination and Wallerian degeneration. If surgically decompressed early, symptom and functional recovery outcomes can often be significantly improved (Matsis, Chou, & Clode, 2021).

1.2.3B Nerve Traction Injuries

Traction, also known as stretch injuries, is less common than compressive injuries but the consequences can be devastating especially in proximal nerves such as the brachial plexus. The brachial plexus is a network of nerves located in the shoulder, which originated from the C5 to T1 spinal segments, that innervate the shoulder girdle, arm, and hand. Obstetrical brachial plexus injuries (OBPI) are common, with an incidence of 1.6-2.6 out of every 1000 live births (Coroneos et al., 2017). These injuries occur during delivery as the infant's shoulders become impinged in the birth canal. Injury to the upper plexus from C5 to C6 is the most common and has the best prognosis with spontaneous recovery occurring in 80% of patients. Flail limb from injury to the entire brachial plexus occurs in 20% of OBPIs with poor outcomes without surgical management (Buterbaugh & Shah, 2016). Although children have a greater capacity for nerve regeneration, even with surgical intervention, functional deficits can persist (Ladak et al., 2013; Lin, Schwentker-Colizza, Curtis, & Clarke, 2009).

While traumatic brachial plexus injuries are not common in paediatric cases (Dorsi, Hsu, & Belzberg, 2010). In adult patients they are most commonly seen among young males following motor vehicle accidents or industrial trauma (Huckhagel, Nüchtern, Regelsberger, & Lefering, 2018). Patient outcomes after brachial plexus injury are often poor and require long primary hospital stay and prolonged inpatient rehabilitation.

1.2.3C Nerve Transection Injuries

Nerve transection can range from a partial axonotmesis, where some axons in a nerve are cut, to complete neurotmesis, where an entire nerve is cut. This type of nerve injury is common in traumatic peripheral nerve injuries, with an incidence of 43.8 nerve injuries per million (Karsy et al., 2019). The digital and ulnar nerves (18%) were the most frequently affected, followed by the radial (15%) and median (13%) with brachial plexus injuries identified in 15% of patients.

Digital nerve injuries, injuries to the fingers of the hand, are commonly caused by transection injuries, with an annual incidence of 6.2 per 100,000 (Thorsén, Rosberg, Steen Carlsson, & Dahlin, 2012). While sensory recovery following digital nerve laceration is variable in the literature, less than a quarter of patients will recover to normal sensory function (Dunlop, Wormald, & Jain, 2019). With most patients required to take leave from work with a median of 2 months, significant socioeconomic costs are associated with these injuries (Thorsén et al., 2012). Despite surgical nerve repair, outcomes still include cold insensitivity (2-53%), hyperesthesia (40-67%), and numbness (>75%).

1.3 Challenges of Peripheral Nerve Regeneration

1.3.1 Challenges of Peripheral Nerve Regeneration

For many decades, outcomes for patients with peripheral nerve injuries have not significantly improved despite advances in surgical technique (Lundborg, 2000). Even with timely repair, only ~10% of regenerating axons reach their targets, resulting in only partial functional recovery (Zochodne, 2012). Currently, there are no effective treatments that can reliably yield full functional recovery (Sabatier & English, 2015). This is because successful functional recovery is critically

dependent on the time it takes for regenerating motor and sensory axons to reach muscle and sensory receptors that are constrained by the following factors:

(a) Staggered axonal regeneration at the site of injury.

(b) Long distances between the injury site and distal targets.

© Slow innate speed of nerve regeneration of only 1mm/day.

(d) The declining regenerative capacity of the denervated distal stump.

1.3.1A Staggered Axonal Regeneration

Following axonal extension after nerve injury, Cajal recognized that rather than regenerating in a straight trajectory towards the target, axons traversing the site of injury took tortuous routes and sometimes retrograde spirals (Cajal, 1991). Initial axon regeneration was retarded in the first three days in a period denoted as staggered regeneration that have been confirmed by other investigators in the following decades. Indeed, over a four-week time period in a rodent model, axon regeneration across the site of surgical coaptation is staggered rather than as a unified front (Brushart et al., 2002). The end result of staggered regeneration is that it slows the rate of axon growth and prolongs the time end organs are denervated.

1.3.1B Regeneration Over Long Distances

The critical nerve gap is defined as the distance of a nerve gap where no natural recovery could occur without the addition of nerve grafting or bridging (Angius et al., 2012). For humans, the critical nerve gap is 4 cm, beyond which graft repair may not be efficacious (Schmidt & Leach, 2003). Proximal nerve injuries are particularly challenging because the long distance the regenerating proximal stump must regenerate to reach its end targets. For example, in high ulnar nerve injuries where the ulnar nerve may be injured at the elbow, the ulnar nerve would need to regenerate over 300 mm to innervate its end organs in the forearm and hand.

1.3.1C Slow Innate Rate of Regeneration

The innate rate of axon longitudinal growth beyond the injury site is slow at approximately 1 mm/day (Gutmann, Gutmann, Medawar, & Young, 1942). The limiting factor to the rate of axonal outgrowth is the rate that cytoskeletal proteins synthesized at the nerve cell body can be transported down the length of the axon to reach the growth cone (Cleveland & Hoffman, 1991). Consequently, proximal injuries are particularly debilitating because of the long distance of regeneration. This translates to potentially months or years for a regenerating nerve to reach its distal target. Unfortunately, functional restoration is often not possible.

Growth cones form with 50 to 100 axonal sprouts arising from the exposed node of Ranvier. Growth cone development is not dependent on the cell body, but rather on local factors that surround the site of axonal injury. In vitro studies have shown that isolated axons can continue to form growth cones as long as axonal transport is maintained (Bray, Thomas, & Shaw, 1978). However, axon elongation beyond the growth cones can only be achieved through the transportation of proteins synthesized from the cell body (Davis, Dou, DeWit, & Kater, 1992). While the development and fate of the growth cone is decided by the molecular environment at the site of injury, axonal regeneration is controlled by the cell body. If the environment distal to the nerve injury is not supportive, the growth cone grows in an undirected, spiral formation, sometimes yielding a neuroma (Sunderland, 1951). On the contrary, supportive environments encourage growth cones towards the distal nerve stump forming thin nerve fibres that grow distally. These are termed regenerating units that grow along the distal stump until they reach their end targets (Morris, Hudson, & Weddell, 1972). Subsequent pruning will occur when a single unit contacts its target. Axon diameters do not reach maturity until they form a functional target connection (T. Gordon & Stein, 1982). Once the growth cone reaches the distal stump, it stimulates SCs to proliferate and

myelinate the newly formed axons. The extent of myelination depends on the diameter of the regenerating axon (Hildebrand, Mustafa, & Waxman, 1986). Although SCs play a primary role in myelin formation, the initial myelin is produced with short internodal distances which result in low conduction speeds. With time, through myelin remodelling, the inter-nodal distances are lengthened so that normal conduction velocities can be achieved.

1.3.1D Declining Distal Regenerative Environment

Following nerve injury, the regenerative capacity of denervated distal targets progressively deteriorates with time. In chronic nerve injuries, this can further decrease the ability for motor neurons to regenerate by 66% and distal Schwann cells by 90% (Tessa Gordon, Sulaiman, & Boyd, 2003). Additionally, prolonged denervation caused by delayed nerve repair accounts for a 90% reduction in the number of functioning motor units.

It might be assumed that the poor regeneration associated with prolonged denervation is associated with the inability for chronically denervated muscle to accept reinnervation. However, regenerating axons can reinnervate three- to fivefold its original number of muscles, forming larger motor units than normal (S. Y. Fu & Gordon, 1995). Theoretically, only 30% of nerve fibres are necessary to maintain function, as the larger motor units can be recruited to generate five times the original force. Rather, prolonged denervation causes the continual deterioration of the intramuscular nerve sheaths. The deterioration of guiding nerve sheaths forces regenerating axons to extend outside the sheaths, resulting in axons failing to reach target and form functioning motor units. Thus, the primary reason for poor recovery after long-term denervation is the significant reduction in axons capable of successfully regenerating through the distal nerve stump. Moreover, chronically denervated muscle fibres undergo irreversible denervation atrophy. A possible reason muscle fibres fail to fully recover is due to the exhaustion of the satellite cell population.

1.3.2 Wallerian Degeneration

Upon a peripheral nerve fibre cut or crush, an active process of degeneration occurs to the axons distal to the injury (see Figure 1.4). Within 24 hours, these axons, which have been severed from their cell body, undergo granular axonal skeleton disintegration and anterograde myelin sheath degradation. This phenomenon is known as Wallerian degeneration, named after Augustus Volney Waller (Waller & Owen, 1850). Initial events involve Ca^{2+} accumulation within the neuron, leading to the dissolution of neurofilaments and axon breakdown. Schwann cells rapidly respond to axonal injury, taking on the major role of degrading their own myelin through the hydrolysis of phosphatidylcholine using Schwann cell phospholipases (Gaudet, Popovich, & Ramer, 2011). Additionally, phagocytosing extracellular myelin and attracting macrophages (Vargas & Barres, 2007). To recruit macrophages, Schwann cells release cytokines and chemokines. As the blood nerve barrier breaks down, the degenerating nerve produces macrophage chemotactic molecules aiding in their infiltration. In concert, macrophages and Schwann cells enable the rapid clearance of distal stump debris. Macrophages secrete interleukins and other cytokines aid in the dedifferentiation of Schwann cells to a proliferating Schwann cell. As neural debris is cleared, proliferating Schwann cells form endoneurial sheaths called the Bands of Büngner which guide proximal regenerating axons towards their targets. To attract new axonal sprouts growing from the proximal stump, Schwann cells also emit growth factors.
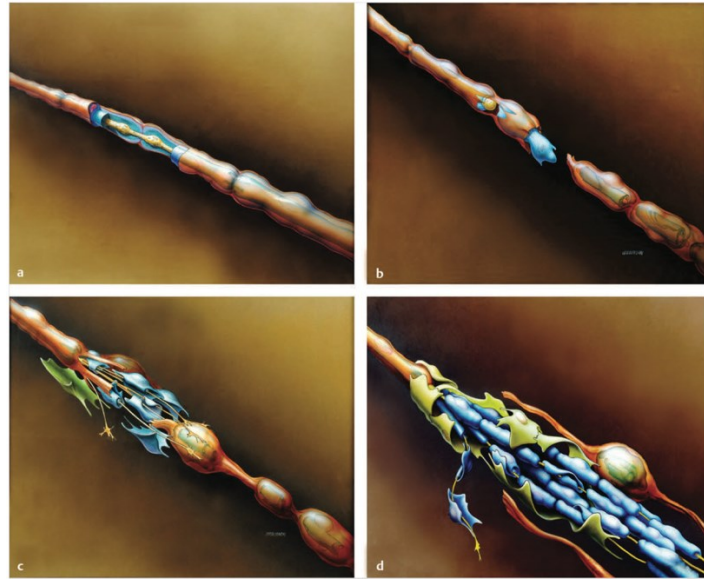
FIGURE 1.4 AXONAL INJURY AND REGENERATION.

Peripheral nerves can regenerate damaged axons. (a) A healthy myelinated axon. (b) Axons proximal to the nerve injury will retract to a node of Ranvier and neurons will change to a regenerative phenotype. Schwann cells distal to the injury become activated and switch into a phagocytic pro-regenerative phenotype. These activated Schwann cells and macrophages break down the distal stump in a process called Wallerian degeneration. (c) Regenerating axons sprout from the node of Ranvier. (d) As axons continue to regenerate distally, Schwann cells begin to myelinate portions of axons close to the site of injury. [(Mackinnon, 1988) Surgery of the Peripheral Nerve.]

1.3.3 Cellular Response to Nerve Injury

Following nerve injury, the neuron cell body undergoes chromatolysis, which are a series of structural changes that include nuclear eccentricity, nucleolar swelling, and the dissolution of Nissl bodies (Zochodne, 2008). The neuron may either undergo apoptosis or regenerate. Nonetheless, the underlying mechanisms which determine the neuron's ability to survive or undergo programmed cell death are not well understood. The neuron's response to survival and axonal regeneration is controlled by the cell body.

Injured neurons release cytokines that stimulate the inflammatory response. These cytokines act synergistically with other cytokines released by macrophages and non-neuronal cells and include interleukin-1 (IL-1), IL-2, IL-6, transforming growth factor-β (TGF-β), and interferon-gamma (IFN-γ).

Neurotrophic factors are a family of biomolecules, nearly all are peptides, involved in neuron survival and regeneration. These molecules can be released from the distal nerve stump, the proximal nerve, or surrounding glial cells. Neurotrophic factors upregulate the production of a variety of regeneration associated proteins, which include and are not limited to: tubulin, actin, calcitonin-gene related peptide (CGRP), and growth associated protein 43 (GAP 43) (Susan Y. Fu & Gordon, 1997).

Axon injury also upregulates signalling neuropeptides including CGRP in motor neurons and vasoactive intestinal peptide (VIP) in sensory neurons (Grafstein, 1975). CGRP is involved in sustaining the inflammatory response needed for regeneration while VIP increases blood supply to regenerating axons (Said & Mutt, 1970). CGRP and VIP may also be involved in positively regulating glial cell function by increasing cyclic adenosine monophosphate (cAMP), which potentiates the effects of mitogenic growth factors on SCs and epithelial cells (Cheng, Khan, & Mudge, 1995). These mitogenic growth factors include fibroblast growth factor (FGF), glial growth factor (GGF), and platelet derived growth factor (PDGF). They are released from injured axons, SCs, macrophages, and platelets.

1.3.4 Proximal Stump Degeneration

Like the distal stump, the proximal nerve to the injury site will also degenerate back to the first or second node of Ranvier, in a process called traumatic degeneration (Zochodne, 2008).

1.4 Nerve Transfer for Peripheral Nerve Repair

Optimal functional recovery following peripheral nerve injury hinges on decreasing the time between nerve injury and target end-organ reinnervation. Time which determines the degree of Wallerian degeneration and the severity of chronic end-organ irreversible atrophy. To mitigate the deleterious effects of prolonged target denervation, nerve transfer surgical techniques exist. The following sections describe these techniques on enhancing nerve regeneration and reinnervation outcomes, the underlying mechanisms of action, and its limitations.

1.4.1 History of Nerve Transfer Surgery

The surgical management of nerve injuries has been described since the 7th century in ancient Greece by Paul of Aegina. The first successful nerve regeneration after primary surgical repair was reported by Cruikshank at the end of the 18th century. With improved understanding of the pathology of nerve injury, nerve autograft techniques represent the modern era of nerve injury repair. Hanno Millesi's pioneering work in the 1960s using interfascicular nerve grafting to avoid tension at the repair and development of good microsurgical technique was a major milestone in nerve repair. With regards to timing of nerve repair, patient outcomes underscore the fact that outcomes are improved when repair is undertaken early. If a nerve can regenerate well on its own, typically Sunderland grade 1 and 2 injuries, the outcomes of spontaneous recovery are excellent. For this reason, surgeons often wait several months before surgically exploring the nerve injury. Nonetheless, the dogma of waiting many months is problematic so many nerve surgeons now operate within a few weeks from the time of injury. Depending on the severity of certain axonal injuries, damaged nerve segments may need to be removed, making primary repair not feasible. To bridge this gap, nerve conduit grafting can be used. Though the 'gold standard' for peripheral nerve

repair, the outcomes can be disappointing as the regenerating axons need to traverse two coaptation sites (see Figure 1.5).
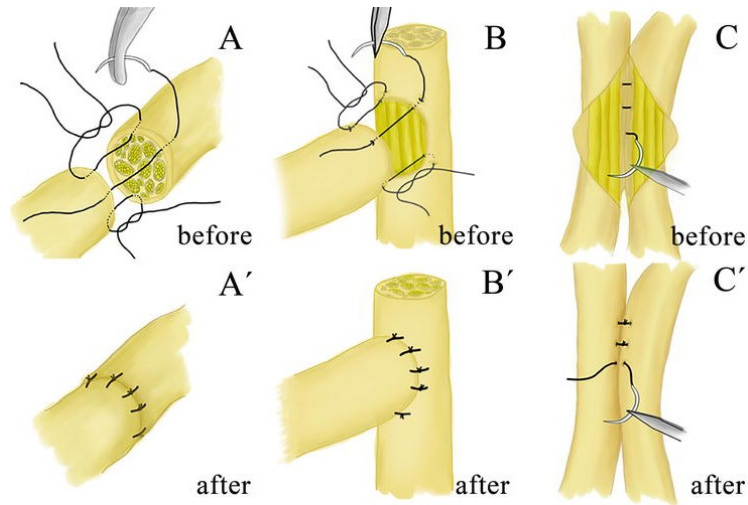
FIGURE 1.5 TYPES OF NERVE REPAIRS.

(a) Prior to invention nerve transfers, primary nerve repair and nerve grafting were the conventional treatment for severe nerve injuries. The cut end of a donor nerve is sutured to the cut end of an injured nerve in an end-to-end fashion. (b) In the end-to-side nerve transfer, the injured nerve is cut, and the distal end is sutured to the side of the donor nerve with either an epineurial or perineurial window. In the reverse end-to-side variation, the donor nerve is cut, and the proximal end is sutured to the side of the injured nerve with an epineurial or perineurial window. (c) The side-to-side nerve transfer involves exposing the nerve fascicles of the donor and injured nerves and coapting the sides of each nerve together. [Yang et al., (2019) Somatosensory and Motor Research.]

The modern nerve transfer era began arguably in the early 1990s with a series of papers that began to explore the possibility of neurotization in severe plexus injury using an extraplexal donor nerve such as the intercostals, the spinal accessory nerve, the phrenic nerve, and the medial pectoral nerve. In 1994, Oberlin published his now seminal paper detailing the transfer of an ulnar fascicle to

the biceps motor nerve for reanimation of elbow flexion. The success of this relatively simple procedure transformed the collective thinking that had previously regarded nerve transfers as a salvage-only procedure. Several caveats for success include choosing a donor nerve that has redundant function to other preserved nerves so that there is no significant downgrading of function in the patient. Interfascicular dissection so that single fascicles are chosen as donors again decreases the possibility of functional complications.

1.4.2 Advantages of the End-to-End Nerve Transfer

Improved outcomes from nerve repair came as a paradigm shift with the advent of the end-to-end (ETE) nerve transfer technique. This technique involves repairing the distal end of an injured denervated nerve element by connecting an anatomically adjacent proximal end of a donor nerve with redundant intrinsic function. Any peripheral nerve injury where there is complete nerve damage, will require axonal regeneration and reinnervation, and is therefore a chronic nerve injury. Because the rate of nerve regeneration is 1 mm/day and the distance between the site of injury and end organs is lengthy, it renders the distal nerve zone, muscle end organs, and sensory receptors to be chronically denervated. By performing these ETE nerve transfers as far distally from the original nerve injury site and close to the end organ, one can circumvent the long regeneration distances and uncertain outcomes that plague proximal nerve injuries, and theoretically innervate end targets faster.

ETE nerve transfers perform better than repairs requiring interpositional grafts (Chan, Olson, Morhart, Lin, & Guilfoyle, 2014; Sallam, El-Deeb, & Imam, 2017; Wang et al., 2013). For example, compared to grafting techniques, ETE nerve transfers provide superior and reliable recovery of elbow flexion and shoulder abduction in the context of upper plexus injuries which involve multiple roots (Socolovsky, Martins, Di Masi, & Siqueira, 2012; Zarina S. Ali et al., 2015).

Indeed, the efficacy of ETE nerve transfers has been systematically analyzed several times (Garg, Merrell, Hillstrom, & Wolfe, 2011; Yang, Chang, & Chung, 2012).

In contrast to primary nerve repairs, the success of nerve transfers is also influenced by cortical plasticity, which has been robustly demonstrated in nerve transfer rehabilitation (Anastakis, Malessy, Chen, Davis, & Mikulis, 2008; Mohanty, Bhat, & Devi, 2015; Wang et al., 2013). This is evident from the choice of using synergistic donor/recipient combinations for successful nerve transfers as this would minimize the challenge of cortical re-education. In the Oberlin's nerve transfer, a single mixed branch of the proximal ulnar nerve is transferred to the end of the biceps motor branch to restore elbow flexion following brachial plexus injury (Oberlin et al., 1994). This is an example of a synergistic donor/recipient nerve combination because the ulnar donor nerve innervates digit flexion, which is a movement pattern that is associated with elbow flexion. Another use case for nerve transfers is in traction injuries to the superficial portion of the common peroneal nerve at the knee, which leads to tibialis anterior muscle denervation and results in foot drop, the inability for a patient to dorsiflex their foot. In these cases, branches of the tibial nerve that innervate plantar flexion muscles, are transferred in an antagonistic donor/recipient nerve combination to the end of the common peroneal nerve (Nath, Lyons, & Paizi, 2008). While this surgery can restore function, the degree of improvement has been variable, which can be attributable to the cortical plasticity and nervous system learning needed to orchestrate coordinated movement of antagonistic peripheral nerves (Bao, Wei, Zhu, & Zheng, 2022).

Other factors which influence the success of nerve transfers are donor size matching with the recipient nerve, and the preoperative electromyography activity from the donor nerve. Because a redundant nerve is often selected as the donor nerve to minimize additional loss of function, the diameter of this nerve is often small. This leads to a donor nerve that has less axons than the injured

nerve. While this might intuitively lead to poor functional recovery due to the mismatch in axon count, it is well known that as the terminal axon sprouts are capable of expanding their innervating territory. To compensate for the decrease in motor unit number, the motor unit size can dramatically increase by almost an order of magnitude (Luff, Hatcher, & Torkko, 1988), leading to greater motor unit force generation (T. Gordon & Tyreman, 2010). The optimal donor-to-recipient nerve axon count ratio in the Oberlin transfer seems to be greater than 0.7:1 (Schreiber et al., 2015). For over 15 years, nerve transfers have been the go-to reconstructive technique to reinnervate, denervated end organs by coapting a nearby healthy dispensable nerve.

1.4.3 Shortcomings of ETE Nerve Transfers

The superior outcomes compared to primary repair and the potential cost benefits compared to grafting have solidified distal nerve transfers as the standard of treatment for over a decade. However, there are reservations to the ETE nerve transfers. For example, the complete transection of damaged nerves leads to an absence of option for intrinsic regeneration of the injured nerve. This is an important consideration in incomplete nerve injuries, where once could not justify transecting an incompletely injured nerve when there are still substantial healthy axons that persist.

1.5 End-to-side Nerve Transfer for Peripheral Nerve Repair

In the conventional end-to-side (ETS) nerve transfer, the injured nerve is cut, and the distal end is connected to the side of a donor nerve, preserving donor nerve continuity. The clinical outcomes of this transfer are often worse than a direct end-to-end nerve transfer repair (Xie et al., 2021). In contrast, reverse end-to-side (RETS) nerve transfers may be attractive for incomplete nerve injuries as instead, the donor nerve is cut and coapted to the side of the injured nerve using an epineurial or perineurial window (see Figure 1.6). This technique has been of particular interest over

the past decade for its applications in the forearm to restore ulnar function through an anterior interosseous nerve to ulnar nerve reverse end-to-side nerve transfer. Finally, there is the side-to-side (STS) transfer where the donor and injured nerves are coapted either with regular intervals of interpositional grafting or opening an epineurial window along the length of the nerve and directly sutured together.
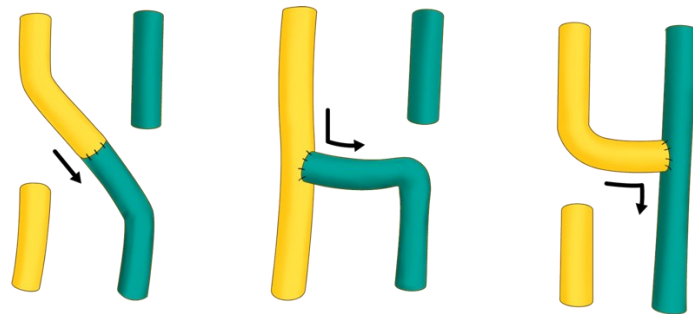
FIGURE 1.6 ILLUSTRATION OF ETE, ETS, AND RETS NERVE TRANSFERS.

(a) The cut end of a donor nerve is sutured to the cut end of an injured nerve in an end-to-end fashion. This nerve transfer does not allow any native axonal regeneration. (b) In the end-to-side nerve transfer, the injured nerve is cut, and the distal end is sutured to the side of the donor nerve with either an epineurial or perineurial window. This nerve transfer does not allow any native axonal regeneration, however, preserves the continuity of the donor nerve. (c) In the reverse end-to-side variation, the donor nerve is cut, and the proximal end is sutured to the side of the injured nerve with an epineurial or perineurial window. This transfer allows native axons in the injured nerve to regenerate alongside donor axons. [Illustrated by Bonnie Wang.]

1.5.1 The End-to-Side Nerve Transfer

1.5.1A History

ETS nerve transfers were first described in the late 19th century by Balance and Kennedy, who coapted the distal end of a cut facial nerve to the side of the spinal accessory nerve to treat facial paralysis. However, the resulting denervation of the muscles innervated by the donor nerve as well as the functional impairment in the association between facial, shoulder, and tongue movements lead to the abandonment of this type of nerve transfer (Babcock, 1927). It was not until a century later, when this technique was revisited by Viterbo et al, who for the first time, performed the ETS transfer without injury to the donor nerve, proposing that an epineurial or perineurial window through the connective tissue was not necessary (Fausto Viterbo, Trindade, Hoshino, & Mazzoni Neto, 1992). Through a series of rodent experimental studies, it was proposed that lateral axonal sprouting through the endoneurium, perineurium, and epineurium barriers occurred from the intact donor nerve and innervated a recipient nerve (Cederna, Kalliainen, Urbanchek, M. Rovak, & Kuzon, 2001; Lohman, Bullock, McNaughton, & Siemionow, 1997; Lundborg, Zhao, Kanje, Danielsen, & Kerns, 1994; Matsuda et al., 2015; Rovak, Cederna, & Kuzon Jr, 2001).

1.5.1B Current Applications

The use of ETS transfers to restore motor recovery in brachial plexus injuries has been reported by multiple groups. Results have been varied though most were poor (Pienaar, Swan, De Jager, & Solomons, 2004). Grossman et al. demonstrated significant improvements in shoulder function using ETS transfers to repair upper brachial plexus injuries, however, the patient demographic were infants who possess a greater potential for nerve regeneration (Grossman et al., 2004). In contrast, Ferraresi et al noted disappointing outcomes in all subjects, failing to recover past M1 function (palpable twitch), subsequently abandoning this technique at their institution (Ferraresi

et al., 2002). In a higher-powered retrospective study with a variety of brachial plexus and upper extremity injury, similarly poor outcomes were demonstrated (Pienaar et al., 2004). At the final endpoint, not only was there no evidence of motor recovery, two patients also suffered inadvertent donor nerve morbidity, prompting Pienaar and colleagues to abandon this technique as well. While there is some evidence for the use of ETS transfers for peripheral motor repair in a variety of injury models (Amr & Moharram, 2005; Amr, Moharram, & Abdel-Meguid, 2006; Franciosi, Modestti, & Mueller, 1998; Mennen, 1998, 2003; Mennen, van der Westhuizen, & Eggers, 2003), their account of success is benchmarked to the nerve graft, which may have inferior outcomes to nerve transfers depending on the paradigm.

1.5.1C Facial Applications

Compared to the poor outcomes in brachial plexus motor recovery, the applications of ETS in facial reanimation have been more convincing (Okochi et al., 2016; Sforza et al., 2015; Su et al., 2020; Ueda et al., 2007; F. Viterbo, 1993; Fausto Viterbo, Romão, Brock, & Joethy, 2014). An important point of consideration is that most of these studies had used ETS nerve transfers for the babysitting effect while the cross-face nerve graft was reinnervating.

1.5.1D Sensory Recovery

There is also evidence for the efficacious use of ETS for sensory recovery (Artiaco, Tos, Conforti, Geuna, & Battiston, 2010; Leechavengvongs et al., 2011; Puonti, Jääskeläinen, Hallikainen, & Partanen, 2011, 2017; Rapp, Lallemand, Ehrler, Buch, & Foucher, 1999; Voche & Ouattara, 2005). A potential explanation for the improved sensory outcomes can be attributed to the greater number of sensory axons available, which can outnumber motor axons nine to one (Gesslbauer et al., 2017). Additionally, sensory nerve transfers do not appear to have the inherent time limitation that the motor nerve transfers have, secondary to terminal Schwann cell senescence (Wagstaff et al.,

2021). Better sensory nerve regenerative capabilities cannot be explained by the difference in rate of regeneration, as sensory and motor myelinated axons regenerate with similar rates following peripheral nerve injury (Moldovan, Sørensen, & Krarup, 2006).

1.5.1E Limitations of ETS

Experimental models have had varied results in functional differences between ETE or ETS nerve transfers, with some demonstrating not difference (Fausto Viterbo, Amr, Stipp, & Reis, 2009; Fausto Viterbo et al., 2017), and others showing superior outcomes from ETE compared to ETS transfers (Jaeger et al., 2011; Liao, Chen, Wang, & Tseng, 2009). The only advantage the ETS transfer has over the ETE transfer, is the preservation of donor nerve continuity. Nevertheless, because the injured recipient nerve is completely cut in both techniques, they share a common limitation, which is the loss of potential intrinsic axon regeneration that might have occurred. While this limitation may not be applicable for injuries of Sunderland grade V or higher, where complete nerve injury has already occurred, for Sunderland grade IV and lower, complete transection of a partially injured nerve would not be feasible. The most widely accepted mechanism of nerve regeneration from the ETS nerve transfer is collateral sprouting of axons from the proximal Ranvier nodes of the donor nerve, however, histologic evidence is lacking (Beris & Lykissas, 2009). This is a process where axons from an intact donor nerve will laterally sprout and enter the neural tubes of the injured nerve. While sensory nerves may collaterally sprout, motor axons may only regenerate in response to deliberate injury (Pannucci, Myckatyn, Mackinnon, & Hayashi, 2007). However, there is a major concern that even if axons could collaterally sprout, how might they be able to traverse the endoneurium, perineurium, epineurium of the donor nerve, and the coaptation site which is prone to staggered regeneration. Hayashi et al and others have demonstrated the efficacy of the transfer is a function of the degree injury induced to the donor nerve, suggesting axonal cross-over may be

sourced from terminal sprouting of intentionally injured donor axons, rather than collaterally sprout from intact donor axons (Hayashi et al., 2008). In summary, the current literature indicates that the success of an ETS transfer is premised on performing a partial neurectomy to the donor nerve, which in effect, means a properly done ETS repair is a partial ETE transfer.

1.5.2 The Reverse End-to-Side Nerve Transfer

For this reason, the reverse end-to-side (RETS) nerve transfers may be an attractive option for incomplete nerve injuries. In this nerve transfer, the donor nerve is cut rather than the injured nerve, and coapted to the side of the intact injured nerve using an epineurial or perineurial window. This strategy allows the infiltration of donor axons into the injured nerve and thus increases the number of regenerating axons that can reach the target, without sacrificing any natively regenerating axons. For this reason, this technique has been of particular interest over the past decade for its applications in the forearm to restore ulnar function through an anterior interosseous nerve to ulnar nerve RETS transfer in treat high ulnar injures (Barbour, Yee, Kahn, & Mackinnon, 2012; Davidge, Yee, Moore, & Mackinnon, 2015; Dengler et al., 2020; Farber et al., 2013; Head, Zhang, Hicks, Wolff, & Boyd, 2020; Kale et al., 2011; Koriem, El-Mahy, Atiyya, & Diab, 2020; McLeod, Peters, Quaife, Clark, & Giuffre, 2020; Xie et al., 2021).

The RETS technique was first described in an experimental rodent model by Isaacs et al., with the intention to test a potentially useful nerve reconstructive strategy (J. Isaacs, Allen, Chen, & Nunley, 2005). Their preliminary work showed that donor nerve axons can invade an intact, regenerating injured nerve to achieve functional recovery in rats (J. E. Isaacs, Cheatham, Gagnon, Razavi, & McDowell, 2008). Using confocal imaging, the early RETS results were supported by

studies done by Mackinnon et al., which showed axonal infiltration across the RETS coaptation site in rodents (Kale et al., 2011).

1.5.3 Current Understanding of the Mechanism of Action

While the exact molecular mechanism of action by which RETS nerve transfer augments regeneration is not well studied or understood, there are two fundamental concepts that the RETS transfer is predicated on. Firstly is the babysitting effect, which, through an unknown mechanism, can preserve end organs and augment the regeneration of the native nerve fibres (Barbour et al., 2012; Farber et al., 2013; Kale et al., 2011). Secondly is axonal crossover from the cut donor nerve to the intact injured nerve.

1.5.3A The Babysitting Effect

Although the exact process is not well understood, the "babysitting" effect is often thought of as a muscle or sensory preservation phenomenon that is provided by donor nerve axons. While not fully elucidated, the preservation of end organs involves keeping terminal Schwann cells viable until intrinsic axons regenerate (Olawale A. R. Sulaiman & Tessa Gordon, 2018). The babysitting effect is particularly relevant in motor nerve repair where the window prior to complete end-organ atrophy is 18 – 24 months. It is thought that by providing an ample supply of new axons from a donor nerve, axons can temporarily innervate and preserve the end-organs, which leads to hyper-reinnervation. Though an individual regenerating axon can, and typically does, innervate multiple muscle fibres, a single muscle fibre cannot be innervated by more than one axon (J. Isaacs, 2021). For that reason, it is believed that eventual pruning occurs as the regeneration of injured axons reach the end-organs (Mackinnon, 2013).

Like the effects of muscular atrophy, nerves also undergo progressive degeneration caused by chronic denervation. This phenomenon is initiated as Schwann cells shift from a pro-

degenerative state to a senescent state that no longer supports axonal growth (Höke, Gordon, Zochodne, & Sulaiman, 2002; You, Petrov, Chung, & Gordon, 1997). However, alongside end-organ preservation, the baby-sitting effect also stimulates the presence of a neurotrophic environment provided by the donor neurons and terminal Schwann cells, which switch from a "transmitting mode", to a "growth mode". While both mechanisms have been postulated, it is not entirely clear which are the predominant mechanisms involved. Indeed, Nadi et al's work in rodents demonstrated that the majority of reinnervating axons came from the donor nerve, which competed with native axons, rather than augmenting them (Nadi et al., 2018).

1.5.3B Axonal Crossover

Axonal crossover in RETS transfers has been consistently demonstrated in experimental models. By cutting the donor nerve, the proximal stump is now an exposed face of terminal axon sprouts. It is believed that these axon sprouts traverse the coaptation site, and can potentially penetrate the epineurium, perineurium, and endoneurium. Nonetheless, many surgeons will remove the epineurial and perineurial connective tissues to ease donor axon infiltration. Although axonal crossover is clinically well-recognized in ETE transfers, it has not been clearly demonstrated in human RETS transfer.

1.5.4 Gaps in RETS Nerve Transfer

The success of this technique in experimental models has led to rapid translation from the bench side to the clinic (Barbour et al., 2012; Davidge et al., 2015; Dengler et al., 2020; Farber et al., 2013; Koriem et al., 2020; McLeod et al., 2020; Xie et al., 2021). However, major gaps persist regarding the overall presence of a babysitting effect and whether axonal crossover from the donor nerve is possible. Early studies have shown that patients that have undergone the RETS procedure have experienced functional improvements. However, due to the lack of a control group, and the

indirect nature of functional outcomes, it is unclear whether the improvements are due to axonal crossover, or regeneration of intrinsic injured nerve axons. To objectively test for the presence of a babysitting effect, a RETS procedure would need to be compared with similar patients with no nerve transfer surgery. While experimental studies suggest an equivalence in outcomes when comparing RETS and ETE transfers, intuitively, RETS may be less efficient at maximizing donor axonal infiltration, in the context of the complex anatomy of human nerves (J. Isaacs, 2021). To test for the presence of axonal crossover, the RETS transfer would need to be compared with an ETE transfer group. For both comparisons, adequate randomization, blinding, and patients with similar injury aetiology and baselines would be necessary.

1.6 Thesis Objectives

For successful peripheral nerve regeneration and functional recovery to occur, there are significant nerve physiological challenges to overcome. ETE nerve transfers have presented a feasible methodology to tackle the long-distance of nerve fibre regeneration associated with proximal injuries, as well as offering a solution to rescue the rapidly degenerating distal nerve stump. Nonetheless, ETE nerve transfers are not a feasible intervention for patients with incomplete nerve injuries, because it entails completely wiping out the opportunity for intrinsic injured nerve axons from regenerating. In the RETS nerve transfer, the donor nerve is cut, and the proximal end is coapted to the side of the structurally preserved injured nerve. This theoretically allows for both donor nerve and native nerve axonal regeneration. Based on early experimental models, the RETS transfer presents a promising surgical solution to treat incomplete injuries. Indeed, this enthusiasm has led to the rapid translation of RETS procedures into clinical use across North America and in Europe. While donor axons have been shown to regenerate across the epineural and perineurial

barriers and down the distal stump of injured nerves in ETS nerve transfers in small animal models, there is a question whether this also occurs in humans. Furthermore, there are concerns for the lack of basic science understanding of the mechanism behind ETS/RETS/STS transfers. Despite these uncertainties, RETS transfers have been translated to clinical use. However, it cannot be assumed that the same results can be achieved in humans, rather, it requires close evaluation. Although there is a body of literature on RETS transfers, the records primarily consist of retrospective, non-randomized, and non-controlled studies.

The objective of this thesis is to systematically evaluate whether reverse end-to-side nerve transfers are an efficacious surgical strategy to treat incomplete proximal nerve injuries.

I)   Create a novel technique to objectively quantify the degree of donor to recipient nerve cross-over regeneration.

II)   Apply this novel technique as a primary outcome measure in a prospective clinical trial comparing reverse end-to-side to gold standard end-to-end and decompression surgical techniques.

This is a paper-based dissertation. The following two chapters are published articles in, *Muscle & Nerve* and *Neurosurgery* in which I was the first and fourth author respectively.

Chapter 2

A new method to quantify innervation of the ulnar intrinsic hand muscles by the anterior interosseous nerve in Martin-Gruber anastomosis and nerve transfer surgery.

Simon Wu BSc[1] | Matthew W. T. Curran MD, MSc[2] | Akiko Hachisuka MD, PhD[3] | Mithun Rajshekar PhD[1] | K. Ming Chan MD[1]

[1] Division of Physical Medicine and Rehabilitation, University of Alberta, Edmonton, Alberta, Canada

[2] Division of Plastic Surgery, University of Alberta, Edmonton, Alberta, Canada

[3] Department of Physical Medicine and Rehabilitation, University of Occupational and Environmental Health, Kitakyushu, Japan

2.1 Abstract

Introduction/Aims:

It is important to quantify the amount of crossover innervation from the anterior interosseous nerve (AIN) through Martin-Gruber anastomosis (MGA) particularly in patients with high ulnar nerve injury who undergo nerve transfer surgery. The objective of this study is to describe a novel electrophysiological method for quantifying innervation from the AIN that can be done using conventional nerve conduction study setup and commonly available software for analysis.

Methods:

Seven subjects with MGA and 9 patients who had undergone AIN to ulnar nerve transfer underwent conventional motor nerve conduction studies. Recording was done over the hypothenar and first dorsal interosseous muscles while stimulating the median and ulnar nerves at the wrist and elbow. Datapoint-by-datapoint subtraction of the CMAPs evoked at the elbow and wrist was performed after they had been onset-aligned. The results were compared to the collision technique and innervation ratio method.

Result:

Results from the digital subtraction method were highly correlated with the collision technique (r=0.96, p<0.05). In contrast, its correlation with the innervation ratio method is substantially lower.

Discussion:

In comparison to previously described techniques, the digital subtraction method has a number of practical advantages. It uses conventional nerve conduction study setup, and the added step of digital alignment and subtraction can be done through commonly available software. With the increasing use of nerve transfer surgery in severe high ulnar nerve injury, this could be a useful

method to identify the presence of MGA prior to surgery and for evaluating nerve recovery following surgery.

2.2 Introduction

Martin-Gruber anastomosis is a common anatomic variant that conveys motor axons from the anterior interosseous nerve to the ulnar nerve most frequently in the proximal forearm. Although it has been reported in 20% of the general population (Roy et al., 2016), the size of the anastomosis and the intrinsic hand muscles that receive the motor nerve fibres are highly variable, even between the two sides in the same individual (Martin, Schauer, Czyrny, & Ablove, 2019). In patients with high ulnar nerve injury such as cubital tunnel syndrome, the presence of Martin-Gruber anastomosis could have major functional implications. Therefore, methods that can accurately quantify the amount of motor innervation from the anterior interosseous nerve are needed. Although a qualitative impression of the contribution from the anterior interosseous nerve can be gleaned through visual inspection of the compound muscle action potentials (CMAP), this can be highly inaccurate especially in the first interosseous muscle where the CMAP is highly contaminated through volume conduction by the median innervated thenar muscles that lie in close proximity. Unfortunately, currently available methods including the innervation ratio and collision technique have substantial limitations (Amoiridis & Vlachonikolis, 2003; Kimura, Murphy, & Varda, 1976; Uchida & Sugioka, 1992). These limitations are further magnified by the increasingly common use of nerve transfer surgery for patients with high ulnar nerve injury (Haase & C Chung, 2002; Novak & Mackinnon, 2002; Schenck et al., 2015). To circumvent the challenge of the long distance of regeneration to reach the hand, a branch of the anterior interosseous nerve to the pronator quadratus muscle in the distal forearm can be used to reinnervate the denervated ulnar intrinsic hand

muscles (Schenck et al., 2015). The goal of this study is to describe a novel method that can be used to quantify motor innervation of intrinsic hand muscles by the anterior interosseous nerve in Martin Gruber anastomosis as well as in patients with high ulnar nerve injury following end-to-end nerve transfer where the donor and recipient nerves are both cut and directly coapted. Although an alternative method termed reverse end-to-side nerve transfer, in which the cut end of the anterior interosseous nerve is coapted to the side of the ulnar nerve, is also used, it will not be included in this paper. This is because the ulnar nerve trunk is left intact in the latter situation, and therefore has no impact on Martin-Gruber anastomosis.

2.3 Methods

2.3.1 Subjects

To establish the utilities of the digital subtraction technique, two groups of subjects were recruited for quantification of anterior interosseous nerve fibers innervating ulnar innervated intrinsic hand muscles: i) Individuals with Martin-Gruber anastomosis; ii) patients with severe high ulnar nerve injury (absent or nearly absent ulnar CMAP) who had undergone anterior interosseous nerve to ulnar end-to-end nerve transfer surgery. The project was approved by the institutional research ethics board at the University of Alberta and all subjects provided informed consent.

2.3.2 Study Protocols

2.3.2A Nerve conduction study of the intrinsic hand muscles to quantify contributions from the anterior interosseous nerve.

All studies were performed using a Nicolet Viking Select EMG machine (Natus Inc, Middleton, WI, USA) or an Advantage EMG machine (Neurosoft, Sterling, VA, USA). With the subjects lying supine and the elbow flexed at 90 degrees, recordings were done using surface

electrodes measuring 2.5 x 1.5 cm (Kendall 5500 tab electrodes, Mansfield, MA, USA) placed over the motor point of the hypothenar and first dorsal interosseous muscles where the maximum CMAP with the highest amplitude and sharpest rise time were obtained. A ground electrode was placed between the stimulation sites at the wrist and the recording electrode. The bandwidth of EMG recording was 5–1000 Hz at a sampling frequency of 10 kHz (Kattla & Lowery, 2010). A bipolar hand-held stimulator was used to deliver single stimuli with a pulse width of 100 $\mu\mu$s at an intensity 10 % above the maximum to ensure supramaximal stimulation. The ulnar and median nerves were stimulated at the wrist and elbow (the experimental set up and typical test results are illustrated in Fig 2.1).

2.3.2B Quantitative Analysis of Waveforms Using Digital Subtraction

The CMAPs collected from stimulation of the median and ulnar nerves at the wrist and elbow were digitized and analyzed using a custom program written in MATLAB software (MathWorks Inc, Natick, MA, USA). The CMAPs evoked by stimulating the median and ulnar nerves at the elbow were onset aligned with the CMAP evoked at the wrist from the respective nerve, followed by datapoint-by-datapoint subtraction of the two waveforms. This same process was done with the ulnar nerve CMAPs evoked at the wrist and elbow as illustrated in Fig. 1. Further technical details of the script compiled in MATLAB are described in the supplementary video. This free-standing program can be run on a personal computer without the need for purchasing any proprietary software.
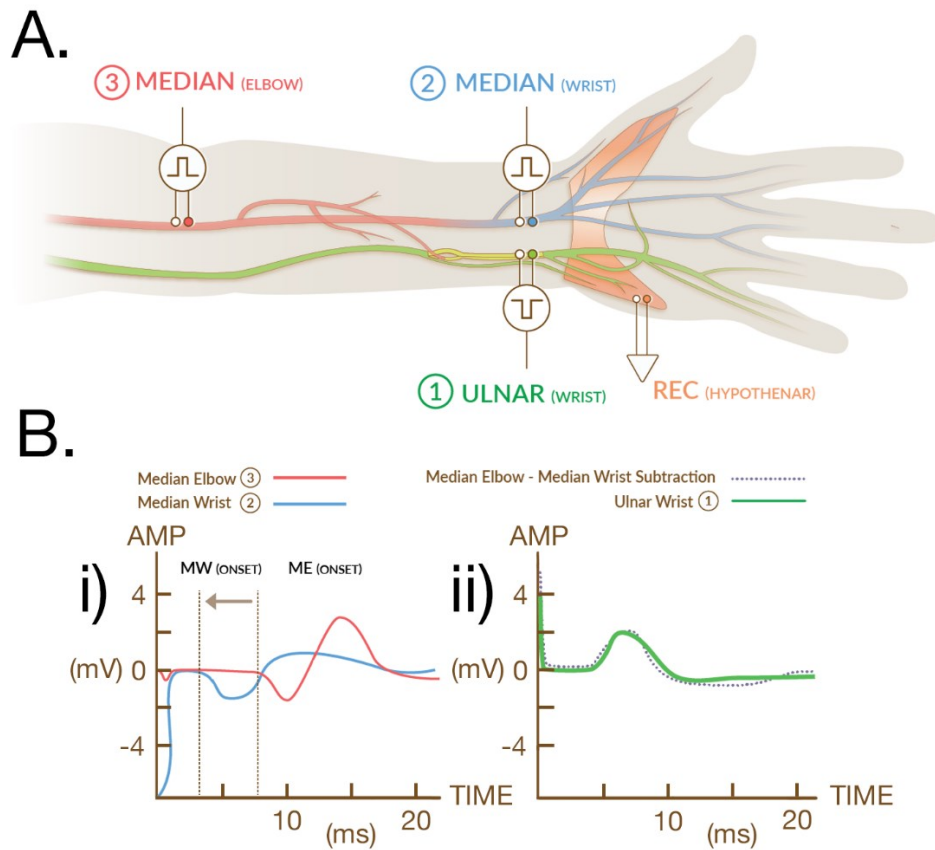
FIGURE 2.1 DATAPOINT-BY-DATAPOINT DIGITAL SUBTRACTION TECHNIQUE.
(a) *Depiction of the sites of stimulation on the median and ulnar nerves while recording over the hypothenar muscles.*

(b) *A representative example of the CMAPs evoked by stimulating the median nerve at the wrist and elbow in a subject who had anterior interosseous to ulnar nerve transfer. The digitally subtracted waveform (dotted line in ii) after onset alignment (i) is similar to the CMAP evoked through stimulating the ulnar nerve at the wrist (solid line in ii).*

2.3.2C Innervation Ratio Technique

Using the same data set, the 'innervation ratio' (IR) technique described by Uchida et al. was used to calculate the ratio between the median elbow and median wrist CMAP recorded from the hypothenar muscles (Uchida & Sugioka, 1992).

$$IR(\%) = \frac{Median\ CMAP_{Elbow}(mW)}{Median\ CMAP_{Wrist}(mV)} \times 100\%$$

2.3.2D The Collision Technique

This technique described by Kimura and Murphy involves stimulating the median nerve at the wrist followed by stimulation at the elbow with a delay of approximately 4 ms (Kimura et al., 1976). This interval is deliberately chosen to be long enough to provide temporal separation of the wrist and elbow evoked waveforms to minimize overlap but yet close enough that the orthodromically propagated volley from the elbow along the median nerve could be collided out by the antidromic volley from the wrist. With the median nerve contribution eliminated, the net waveform represents only the contribution from the anterior interosseous nerve through the anastomosis. The exact stimulation delay between the wrist and elbow stimulations was fine tuned in each individual to ensure complete collision and that the onset of the CMAP from elbow stimulation was optimized for clear identification.

2.3.3 Statistical Analysis

The baseline patient demographics are reported as mean $\pm$ SD while descriptive statistics for the electrophysiologic data are reported as median (range). As the goal is to demonstrate the clinical utility of the digital subtraction technique, this study was not powered to test the efficacy of the nerve transfer procedure. The correlation coefficients between the innervation ratio, amplitude of the CMAP obtained using the collision technique versus amplitude of the CMAP obtained using the digital subtraction technique were calculated. Based on the results of Shapiro-Wilk test, the digital subtraction, collision, and innervation ratio data were significantly skewed. Therefore, non-parametric analysis methods were used. Paired Wilcoxon rank sign test was used to compare differences in the CMAP amplitudes obtained between the digital subtraction method and the

collision technique. Mann-Whitney U test was used to compare differences between the nerve transfer and Martin-Gruber anastomosis subjects. A p-value for type I error of <0.05 was deemed statistically significant. The statistical program Stata 14 (StataCorp, College Station, TX, USA) was used.

2.4 Results

2.4.1 Subjects

Participants consisted of 7 subjects with Martin-Gruber anastomosis and 9 with severe high ulnar nerve injury. The patient demographic and physical characteristics are shown in Table 2.1. The average time from surgery for the ulnar nerve injury patients was 4±2 years.

TABLE 2.1 BASELINE PATIENT DEMOGRAPHICS FOR 13 PATIENTS.

Characteristics	MGA group (n = 7)	Nerve transfer group (n = 9)
Gender (F:M)	2:5	1:8
Age ± SD (y)	66 ± 8	43 ± 22
Dominant limb (L:R)	1:6[a]	2:7
Dominant limb affected (L:R)	2:5	5:4
Follow-up (y ± SD)		4 ± 2
Comorbidities	Type II diabetes (n = 2) Carpal tunnel syndrome (n = 1)	Type II diabetes (n = 1) Traumatic brain injury (n = 1)

[a]One patient was ambidextrous.

2.4.2 Digital Subtraction Method

The findings from a typical patient with severe high ulnar nerve injury who had undergone anterior interosseous nerve to ulnar nerve transfer three years earlier are shown in Fig 2.2. The resultant post-subtracted median waveform revealed substantial reinnervation to the hypothenar and first dorsal interosseous muscles.

The average amount of crossover innervation from the anterior interosseous nerve to the hypothenar muscles as represented by the CMAP negative peak amplitude was 1.99 (0.37-3.51) mV. Compared to subjects with Martin-Gruber anastomosis, crossover innervation from the anterior interosseous nerve in was significantly greater in the nerve transfer group (see Table 2.2). In the first dorsal interosseous muscle, the average amount of crossover innervation from the anterior interosseous nerve was 2.21 (1.18-3.68) mV. The difference between the nerve transfer group compared to the Martin-Gruber anastomosis group was not statistically different. In subjects with Martin-Gruber anastomosis, the amount of crossover innervation from the anterior interosseous nerve to the first dorsal interosseous muscle was significantly greater than in the hypothenar muscles. Details of the CMAP negative peak amplitude and negative phase duration in both groups are summarized in Table 2.3.
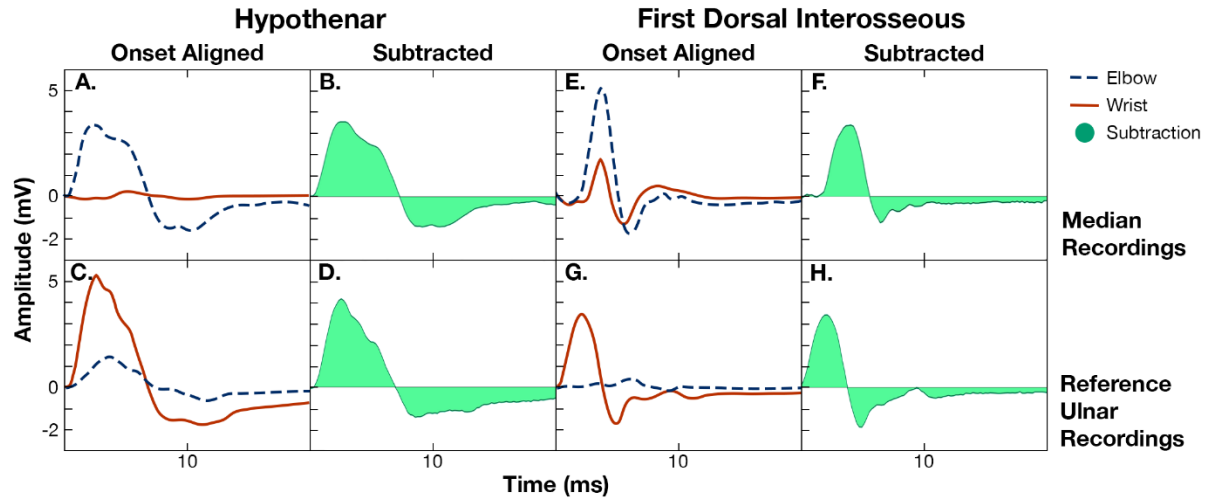
FIGURE 2.2 REPRESENTATIVE RECORDINGS OF THE HYPOTHENAR AND FIRST DORSAL INTEROSSEOUS MUSCLES FROM A PATIENT WITH HIGH ULNAR NERVE INJURY 3 YEARS AFTER NERVE TRANSFER SURGERY.

The onset aligned median waveforms evoked at the wrist and elbow are shown in panel A for the hypothenar muscles and in panel E for the first dorsal interosseous muscles. The resultant digitally subtracted waveforms (B and F) are very similar to those obtained by stimulating the ulnar nerve (D and H).

2.4.3 The Innervation Ratio Technique

In the hypothenar muscles, the innervation ratio in the Martin-Gruber anastomosis group was significantly lower than the nerve transfer group. In contrast, in the first dorsal interosseous muscle, there was no significance between two groups.

2.4.4 The Collision Technique

The CMAP amplitude recorded from the hypothenar muscles in the Martin-Gruber anastomosis group was significantly lower compared to the nerve transfer patients. Similar to results found using the digital subtraction method and innervation ratio, there was no significant difference in the first dorsal interosseous muscle between the Martin-Gruber subjects and the nerve transfer group.

2.4.5 Comparisons between the three methods of quantification

Results obtained using the digit subtraction method and collision technique are highly correlated with a correlation coefficient of 0.96 ($p<0.05$). The CMAP waveforms obtained using these techniques are shown in Fig 2.3. The close resemblance of these waveforms can be readily appreciated from a first dorsal interosseous recording in an ulnar injury patient who had a nerve transfer in Fig 2.3 (i) and another recording over the hypothenar muscles in a subject with Martin-Gruber anastomosis in Fig 23(ii). Variability of the amount of crossover reinnervation from the anterior interosseous nerve to the hypothenar and first dorsal interosseous muscles in the nerve transfer patients is shown in Fig 2.4. In contrast, the correlation coefficient between the innervation ratio technique and the digital subtraction method is substantially lower at 0.54. Details of differences between the methods of quantification are shown in Table 2.2.

TABLE 2.2 GROUP RESULTS OF CROSS-OVER QUANTIFICATION.

Cohorts	Recording location	Techniques		
		Digital subtraction (mV)	Collision block (mV)	Innervation ratio (%)
		Median (range)	Median (range)	Median (range)
MGA group	Hypothenar	0.93 (0.37-1.87)	0.61 (0.38-1.80)	290 (163-546)
	FDI	2.06 (1.35-2.77)	1.74 (1.07-2.41)	305 (207-402)
Nerve transfer group	Hypothenar	2.20 (1.87-3.51) *	1.94 (1.55-3.55) *	745 (233-1972) *
	FDI	2.41 (1.18-3.68)	1.42 (1.21-3.50)	196 (95-343)
Combined	Hypothenar	1.99 (0.37-3.51)	1.79 (0.38-3.55)	453 (163-1972)
	FDI	2.21 (1.18-3.68)	1.42 (1.07-3.50)	217 (95-402)

*Indicates significant difference between nerve transfer and MGA groups.

TABLE 2.3 CMAP AMPLITUDE AND DURATION EVOKED BY MEDIAN NERVE STIMULATION AT THE WRIST AND ELBOW.

		Stimulation location (median *n*)			
		Wrist CMAP	Wrist CMAP	Elbow CMAP	Elbow CMAP
		n-peak amplitude (mV)	*n*-phase duration (ms)	*n*-peak amplitude (mV)	*n*-phase duration (ms)
Cohorts	Recording location	Median (range)	Median (range)	Median (range)	Median (range)
MGA group	Hypothenar	0.16 (0.08-0.29)	6.7 (5.8-7.1)	0.45 (0.14-1.58)	6.2 (6.1-11.7)
	FDI	0.61 (0.07-0.81)	6.9 (3.7-8.4)	1.27 (0.29-2.38)	7.8 (6.7-12.2)
Nerve transfer group	Hypothenar	0.43 (0.09-0.69)	7.3 (6.4-8.5)	2.21 (1.43-4.80)	8.4 (6.4-12.6)
	FDI	1.16 (0.92-2.08)	6.4 (4.1-10.5)	2.70 (1.11-5.19)	11.7 (6.0-12.9)

Note: N-peak denotes negative peak while *n*-phase denotes negative phase.
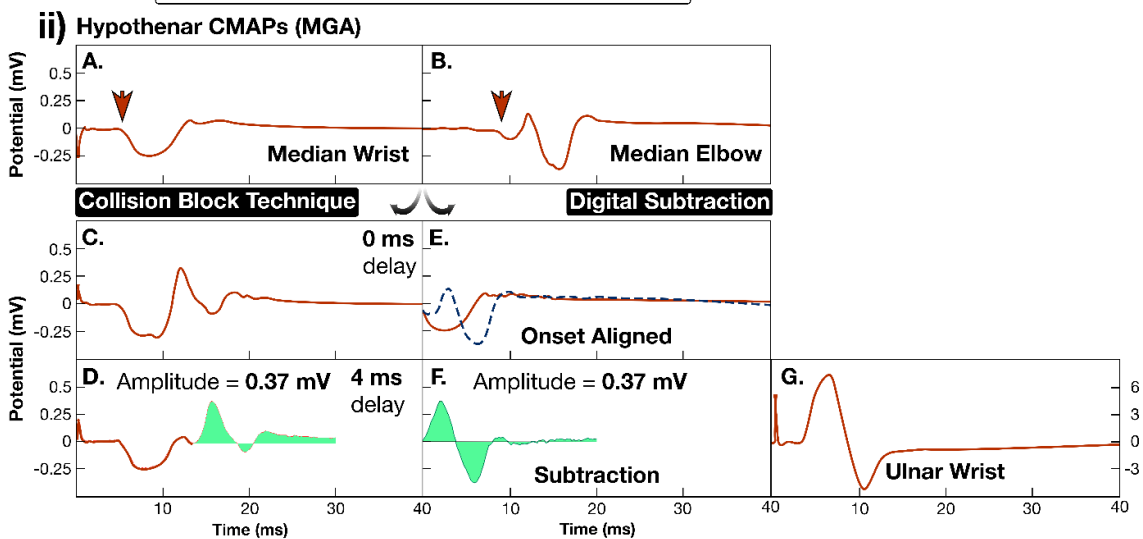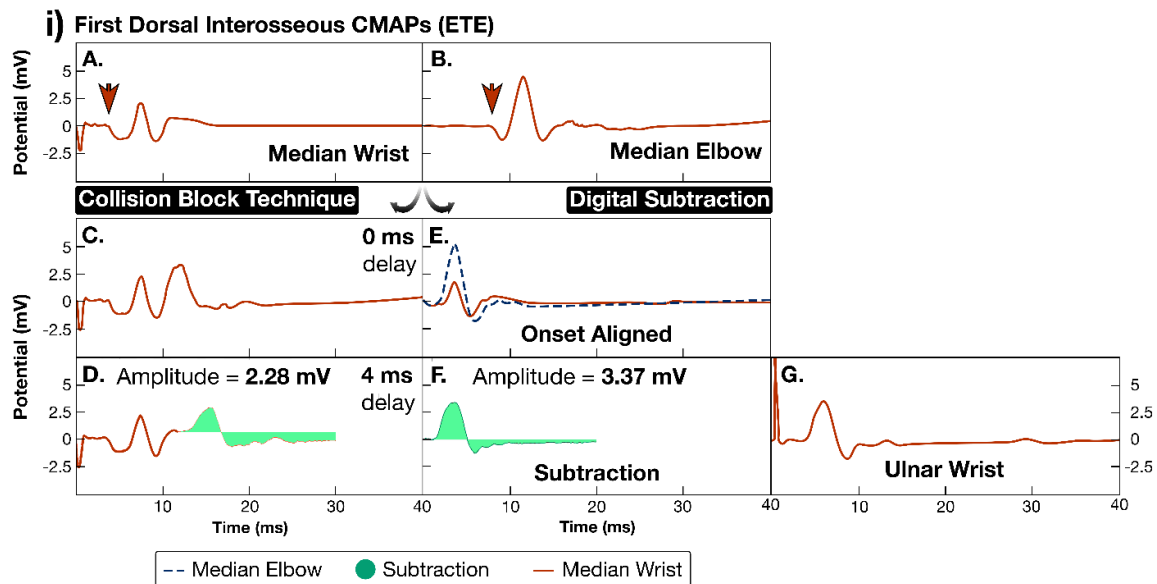
FIGURE 2.3 COMPARISON BETWEEN DIGITAL SUBTRACTION METHOD AND COLLISION TECHNIQUE.

(i) A typical example of recordings on the first dorsal interosseous muscle in a patient who had undergone anterior interosseous to ulnar nerve transfer to compare the waveforms obtained using digital subtraction and collision technique. As shown in panel D, the baseline of the collided waveform was lifted by the preceding wrist waveform resulting in a substantial reduction in the CMAP amplitude. In contrast, the digitally subtracted waveform (in F) much more closely resembles the CMAP evoked by stimulating the ulnar at the wrist (in G) (ii) Recordings from a subject with MGA to compare the results between the two techniques. In panel D, there is no distortion of the baseline at the hypothenar muscles because the size of the preceding CMAP evoked at the wrist is small. Reason for the much larger CMAP amplitude of the ulnar wrist (G) compared to the digitally subtracted waveform is because of the additional contribution from the native motor nerve fibers in the ulnar nerve.
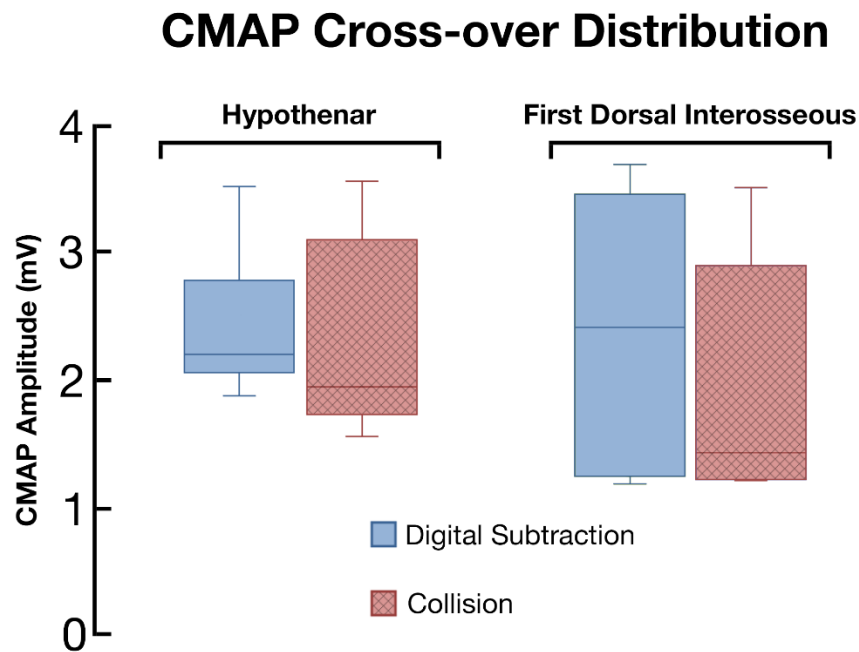
FIGURE 2.4 CROSS-OVER OF MOTOR FIBRES FROM THE AIN TO THE HYPOTHENAR AND FDI MUSCLE.

Note the close overlap of the results from the digital subtraction and collision technique. Notations used: The upper and lower limits of the box denote the 75th and 25th percentile while the upper and lower whiskers represent the 95th and 5th percentile. The horizontal line in the box is the median.

2.5 Discussion

In this paper, we described a technique using digital subtraction to quantify the amount of crossover innervation from the anterior interosseous nerve to the ulnar intrinsic hand muscles that can be applied to individuals with Martin-Gruber anastomosis and patients with high ulnar nerve injury undergoing end-to-end nerve transfer (Roy et al., 2016). The finding that the amount of crossover innervation from the anterior interosseous nerve to the first dorsal interosseous muscle was significantly greater than in the hypothenar muscles in subjects with Martin-Gruber anastomoses is in agreement with the literature (Amoiridis & Vlachonikolis, 2003; Kimura et al., 1976; Uchida & Sugioka, 1992).

Although prior techniques to quantify crossover innervation from Martin-Gruber anastomosis exist, they have important limitations (Kimura et al., 1976; Uchida & Sugioka, 1992). With the innervation ratio technique, the calculated values are highly influenced by the size of the CMAP evoked by stimulating the median nerve at the wrist which is used as the denominator in the calculation. This can be drastically different depending on the muscle groups as can be already seen from the results in this study. The innervation ratios are significantly higher in the hypothenar muscles because the hypothenar CMAPs evoked by stimulating the median nerve at the wrist are much smaller compared to the first dorsal interosseous muscle. This eliminates the possibility of directly comparing crossover innervation from the anterior interosseous nerve in different parts of

the hand. Secondly, since the CMAP recorded from the hypothenar muscles is often very small, it is particularly susceptible to noise or baseline displacement resulting in greater variability and measurement errors. This is the likely explanation for the lower correlation with results from the digital subtraction method.

In contrast, there is a high correlation between the digital subtraction and collision technique which provides support for criterion validity of the digital subtraction method (Kimura et al., 1976). However, the digital subtraction method offers a number of advantages over the collision technique. First, when using the collision technique, clear identification of the onset of the collided CMAP from the elbow is not always easy, as can be readily appreciated in the waveforms shown in Fig 2.3(i). This is primarily due to overlap of the waveforms evoked at the wrist and the elbow. This physiological constraint is attributable to the fact that the duration of a CMAP is close to 10 ms whereas the delay in elbow stimulation that would still allow for collision is only around 4 ms. The challenge is particularly severe with larger CMAPs such as those recorded from the first dorsal interosseous muscle. This could result in significant distortion of the baseline, particularly in cases in which the crossover innervations are small and in shorter individuals with less distance between the two sites of stimulation. In addition, there are practical constraints that have hampered widespread use of the method. First, the optimal amount of delay for collision varies in individuals depending on their arm length and the nerve conduction velocity. The need to individually fine tune the amount of delay consumes extra time. Second, the collision technique requires EMG machines equipped with stimulators with adjustable delays. For these reasons, the method has not seen widespread adoption since it was first described 40 years ago. In contrast, no extra data collection setup or equipment is required for the digital subtraction technique. Indeed, the required study protocol is the same as conventional motor nerve conduction studies. The additional steps needed

for data analysis can be performed on a personal computer or using web-based programs. For example, the script provided in the supplementary material can be run as a stand-alone program without the need for purchasing any additional software.

A physiological limitation to the digital subtraction technique is temporal dispersion and phase cancellation. With the longer distance from the recording electrode, the CMAP evoked at the elbow is slightly smaller and broader than that evoked at the wrist. However, in healthy subjects, the difference is relatively small. Furthermore, this physiological constraint is not unique to digital subtraction as it would also apply to the other techniques as well. A second limitation is a practical one in that the digital subtraction technique is currently not widely available on commercial EMG machines. However, since the algorithm for onset alignment and datapoint-by-datapoint subtraction is relatively simple, it can be easily adopted on commercial EMG machines without the need for additional hardware. Indeed, the technique described in this paper has already been incorporated by one manufacturer. It is possible that other manufacturers may follow suit if there is a sufficient clinical demand for this method.

2.6 References

Amoiridis G & Vlachonikolis IG (2003). Verification of the median-to-ulnar and ulnar-to-median nerve motor fiber anastomosis in the forearm: an electrophysiological study. Clin Neurophysiol 114, 94-98.

Haase SC & Chung KC (2002). Anterior interosseous nerve transfer to the motor branch of the ulnar nerve for high ulnar nerve injuries. Ann Plast Surg 49, 285-290.

Kimura J & Murphy MJ (1976). Electrophysiological study of anomalous innervation of intrinsic hand muscles. Arch Neurol 33, 842-844.

Martin SP, Schauer KT, Czyrny JJ & Ablove RH (2019). Electrophysiological Findings in Common Median-Ulnar Nerve Interconnections and Their Clinical Implications. J Hand Surg. Am 44, 884-894.

Novak CB & Mackinnon SE (2002). Distal anterior interosseous nerve transfer to the deep motor branch of the ulnar nerve for reconstruction of high ulnar nerve injuries. Journal of Reconstructive Microsurgery 18, 459-464.

Roy J, Henry BM, PA PE, Vikse J, Saganiak K, Walocha JA & Tomaszewski KA (2016). Median and ulnar nerve anastomoses in the upper limb: A meta-analysis. Muscle Nerve 54, 36-47.

Schenck TL, Stewart J, Lin S, Aichler M, Machens HG & Giunta RE (2015). Anatomical and histomorphometric observations on the transfer of the anterior interosseous nerve to the deep branch of the ulnar nerve. J Hand Surg Eur Vol 40, 591-596.

Uchida Y & Sugioka Y (1992). Electrodiagnosis of Martin-Gruber connection and its clinical importance in peripheral nerve surgery. J Hand Surg. Am 17, 54-59.

2.7 Acknowledgements

Chapter 3

Reverse End-to-Side Nerve Transfer for Severe Ulnar Nerve Injury: A Western Canadian Multicentre Prospective Nonrandomized Cohort Study

Matthew WT Curran MD MSc[1] | Jaret L Olson MD[1] | Michael J Morhart MSc MD[1] | Simon Wu BSc[1] | Raj Midha MD[2] | Michael Berger MD PhD[3] | K Ming Chan MD[1]

[1] University of Alberta, Faculty of Medicine, Edmonton, Alberta, Canada

[2] University of Calgary, Faculty of Medicine, Calgary, Alberta, Canada

[3] International Collaboration on Repair Discoveries (ICORD), University of British Columbia, Faculty of Medicine, Vancouver, BC, Canada

[4] University of British Columbia, Faculty of Medicine, Victoria, BC, Canada

3.1 Abstract

Background:

Reverse end-to-side (RETS) nerve transfer has become increasingly popular in patients with severe high ulnar nerve injury, but the reported outcomes have been inconsistent.

Objective:

To evaluate the "babysitting effect," we compared outcomes after anterior interosseous nerve RETS transfer with nerve decompression alone. To evaluate the source of regenerating axons, a group with end-to-end (ETE) transfer was used for comparisons.

Methods:

Electrophysiology measures were used to quantify the regeneration of anterior interosseous nerve (AIN) and ulnar nerve fibers while functional recovery was evaluated using key pinch and Semmes-Weinstein monofilaments. The subjects were followed post-surgically for 3 years.

Results:

Sixty-two subjects (RETS=25, ETE=16, and decompression=21) from 4 centres in Western Canada were enrolled. All subjects with severe ulnar nerve injury had nerve compression at the elbow except 10 in the ETE group had nerve laceration or traction injury. Post-surgically, no reinnervation from the AIN to the abductor digiti minimi muscles was seen in any of the RETS subjects. Although there was no significant improvement in compound muscle action potentials amplitudes and pressure detection thresholds in the decompression and RETS group, key pinch strength significantly improved in the RETS group ($P < .05$).

Conclusion:

The results from published clinical trials are conflicting in part because crossover regeneration from the donor nerve has never been measured. Unlike those with ETE nerve

transfers, we found that there was no crossover regeneration in the RETS group. The extent of reinnervation was also no different from decompression surgery alone. Based on these findings, the justifications for this surgical technique need to be carefully re-evaluated.

3.2 Introduction

High ulnar nerve injuries are common. Indeed, cubital tunnel syndrome is the second most common entrapment neuropathy, with a prevalence of 1.8% in the general population (An, Evanoff, Boyer, & Osei, 2017). In severe cases with marked axonal loss causing intrinsic hand muscle weakness, wasting, and sensory loss, results of decompression surgery are often poor. These are due to the challenge of the long distance of regeneration for the nerve fibers to reach target (Tong, Xu, Dong, Zhang, & Gu, 2017). Outcomes of nerve repair after laceration or traction injury are similarly poor (Rosberg et al., 2005).

Nerve transfers have gained increasing popularity in the past decade. In the original approach to reinnervate intrinsic hand musculature, a redundant branch of the anterior interosseous nerve (AIN) to the pronator quadratus muscle is transferred to the deep motor branch of the ulnar nerve through an end-to-end (ETE) coaptation (Haase & C Chung, 2002). Although it was shown to produce better outcomes compared with nerve reconstruction (Flores, 2015), transection of the ulnar nerve trunk presents a dilemma in incomplete injuries because ETE transfer would preclude the possibility of the remaining native motor nerve fibers from regenerating. To circumvent this, Isaacs et al (J. Isaacs et al., 2005) proposed a reverse end-to-side (RETS) transfer by coapting the end of the AIN to the side of the ulnar nerve through a perineurial window (Kale et al., 2011). A purported advantage of RETS is that in addition to providing a route for the donor nerve fibers to grow into the ulnar nerve, it might also augment regeneration of the remaining proximal ulnar nerve

fibers through the so-called "babysitting effect" (Barbour et al., 2012). There has been a growing body of literature on the RETS technique on patients with ulnar compression neuropathy (Dengler et al., 2020; Doherty, Miller, Larocerie-Salgado, Byers, & Ross, 2020; Head et al., 2020; McLeod et al., 2020; Xie et al., 2021), high ulnar nerve lacerations (Baltzer, Woo, Oh, & Moran, 2016; Koriem et al., 2020), and other etiologies (Davidge et al., 2015). However, the source of nerve fibers regenerating into the recipient nerve and restoration of function in RETS transfer has been inconsistent. Although some studies have shown benefits (Baltzer et al., 2016; Davidge et al., 2015), other studies did not find them to be efficacious (Pienaar et al., 2004; Pondaag & Gilbert, 2008).

3.2.1 Objective

To answer this critical question, we used a novel electrophysiological technique to evaluate the amount of crossover regeneration from the AIN into the ulnar nerve innervated muscles in patients with ETE transfer compared with those who underwent RETS (Wu, Curran, Hachisuka, Rajshekar, & Chan, 2022). Second, to evaluate the babysitting effect, we compared the physiological and functional outcomes in patients with severe high ulnar nerve injury who underwent RETS to those who had nerve decompression at the elbow alone.

3.3 Methods

3.3.1 Patient Selection

In this prospective multicentre study, patients seen at the University of Alberta, University of Calgary, and the University of British Columbia in Vancouver and Victoria were recruited. Ethics approval was obtained from all study sites, and all patients provided consent for enrollment in this study. The trial was registered with ClinicalTrails.gov (Trial No. NCT05242302). Inclusion criteria were (1) adults (older than 18 years) and (2) signs and symptoms of severe ulnar nerve axonal loss as

defined as McGowan III and confirmed by electrodiagnostic studies. Exclusion criteria were (1) previous cubital tunnel surgery, (2) presence of a Martin-Gruber anastomosis, (3) confounding neurological disorder, and (4) inability to provide voluntary consent.

3.3.2 Study Arms

This study consists of 3 parallel cohorts: (1) patients with cubital tunnel syndrome who received AIN RETS transfer, (2) patients with cubital tunnel syndrome who received decompression of the ulnar nerve at the elbow, and (3) patients who received AIN ETE for high ulnar nerve laceration or very severe ulnar compression neuropathy at the elbow.

3.3.3 Surgical Techniques

The AIN RETS transfer was performed as described by Barbour et al (Barbour et al., 2012). In brief, the deep ulnar motor branch was decompressed at the Guyon canal and traced proximally. The pronator quadratus muscle was exposed by retracting the flexor tendons to identify the AIN branch to the muscle. A perineurial window was made in the motor branch of the ulnar nerve, and the coaptation was performed. Patients in the AIN ETE group underwent the same dissection but with the AIN and motor branch of the ulnar nerve both cut and directly coapted in an end-to-end fashion. To provide a conduit for the ulnar sensory nerve fibers to regrow, the rest of the ulnar nerve trunk was left untouched. If the primary etiology was an ulnar nerve laceration, a primary nerve repair was also performed. All subjects in the AIN RETS and decompression groups and those with severe ulnar compression neuropathy in the AIN ETE group also underwent an in-situ decompression of the ulnar nerve at the elbow. If ulnar nerve subluxation was found after decompression, a subcutaneous transposition or blocking flap was performed.

3.3.4 Outcome Measures

The subjects were evaluated at baseline and the majority were followed annually for 3 years annually for 3 years after surgery.

3.3.5 Electrophysiological Analysis of AIN and Ulnar Nerve Contribution

Surface electrodes were placed over the abductor digiti minim (ADM) muscle in a tendon-belly monopolar configuration to record the maximum compound muscle action potentials (CMAPs) evoked by stimulating the ulnar and median nerves at the wrist and elbow (illustrated in Figure 1). Through a digital subtraction algorithm using custom written MATLAB program (MathWorks), the relative contribution of the AIN and ulnar nerve to the ADM muscle was quantified. In this technique, onset of the CMAP waveform evoked by stimulating the median nerve at the elbow that gives rise to the AIN was time shifted to align with the onset of the CMAP evoked by stimulating the median nerve at the wrist. The amount of crossover reinnervation from the AIN to the ADM muscle could be quantified by the resultant subtracted CMAP waveform. This is illustrated by an example shown in Figure 3.1.
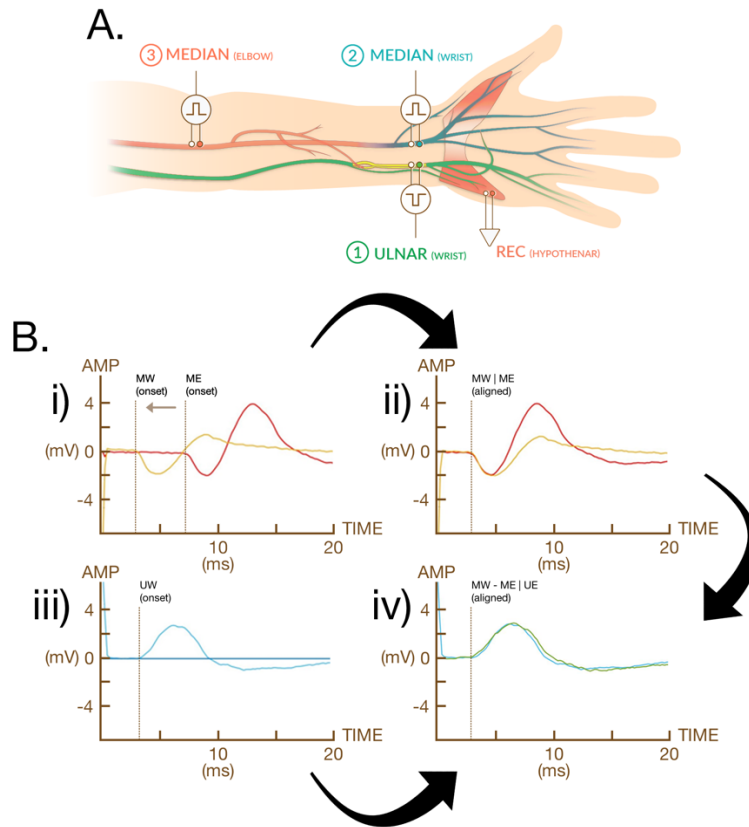
FIGURE 3.1 NOVEL DIGITAL SUBTRACTION CROSS-OVER QUANTIFICATION METHOD.

(a) Depiction of the sites of stimulation on the median and ulnar nerves while recording over the abductor digiti minimi muscle. (b) An illustration of the CMAPs evoked by stimulating the median nerve at the wrist and elbow in a subject with crossover innervation from the AIN. First, the CMAPs evoked by stimulating the median nerve at the wrist and elbow were onset aligned (i and ii). With the entire source of innervating motor fibers coming from the AIN, the digitally subtracted waveform (green line in iv) is similar to the CMAP evoked by stimulating the ulnar nerve at the wrist (light blue line in iii and iv). AIN, anterior interosseous nerve; AMP, amplitude; CMAPs, compound muscle action potentials; ME, median (elbow); MW, median (wrist); REC, recording electrodes.

3.3.6 Functional Outcomes

Key pinch strength designed to measure the first dorsal interosseous and adductor pollicis muscles was assessed using a pinch gauge dynamometer (B&L Engineering). Sensory return was assessed using Semmes-Weinstein Monofilament kit (Sammons Preston Roylan) to determine fine touch pressure detection threshold as previously described (Curran, Morhart, Olson, Hachisuka, & Chan, 2019).

3.3.7 Statistical Analysis

Demographic data were analyzed using the independent t test and reported as mean ± SD. Incomplete data because of missed follow-ups were evaluated according to the intention-to-treat principle. Because the electrophysiological and functional outcomes were found to have a non-normal distribution by the Shapiro-Wilk test ($P < .05$), nonparametric tests were used to determine significance. The Mann-Whitney U test was used to compare baseline outcomes between the groups. To determine improvement over the course of the follow-up within each group, the Kruskal-Wallis test was employed while the Wilcoxon signed rank test was used to determine significant differences between the RETS and decompression group. Statistical significance was defined as $P < .05$. All statistical analysis was calculated using STATA 14.

3.4 Results

3.4.1 Demographics

A total of 62 subjects were recruited: AIN RETS group = 25, AIN ETE group = 16, and decompression group = 21. The patient demographic data are presented in Table 3.1. They were predominately male (85%) with a mean age of 59 ± 15 years. Subjects in the AIN ETE group were significantly younger than those in the AIN RETS group ($P = .03$). Types of injury in the AIN ETE

group consisted of traction injury (n = 7; 44%) or laceration of the ulnar nerve at the elbow or higher (n = 3; 19%), while compression neuropathy at the elbow was the sole etiology in the both the AIN RETS and decompression groups.

TABLE 3.1 PATIENT DEMOGRAPHICS BETWEEN THE 3 STUDY GROUPS.

Age (mean ± SD)	58.9 ± 14.6	62.9 ± 13.9	45.2 ± 15.2[a]	60.4 ± 13.0
Sex (M:F)		21:4	14:2	18:3
Injury		Compression = 22 Laceration = 2 Traction = 1	Compression = 6 Traction = 7 Laceration = 3	Compression = 21
Affected limb (R:L)		13:12	6:10	9:12
Smoking status (Y:N)		5:20	4:12	7:14
Comorbidities		Cancer (n = 2) Hypoglycemia (n = 2) Inferior MI (n = 2) A-fib (n = 1) Asthma (n = 1) Diabetes (n = 1) LAFB (n = 1)	Diabetes (n = 2) IHD (n = 2) Cancer (n = 1) Inferior MI (n = 1) NSTA (n = 1)	Diabetes (n = 6) HTN (n = 3) Inferior MI (n = 3) IHD (n = 2) LAFB (n = 2) A-fib (n = 1) Bradycardia (n = 1) NSTA (n = 1)

A-fib, atrial fibrillation; AIN, anterior interosseous nerve; ETE, end-to-end; HTN, hypertension; IHD, ischemic heart disease; LAFB, left anterior fascicular block; MI, myocardial infarction; NSTA, nonspecific t abnormalities; RETS, reverse end-to-side.
[a]Denotes statistical significance ($P < .05$).
All data presented as mean ± SD unless otherwise indicated.

3.4.2 Baseline Characteristics

Before surgery, all subjects underwent needle EMG examination and were found to have evidence of severe motor unit loss with reduced recruitment. For those with cubital tunnel syndrome, there was a mixture of ongoing denervation in the form of positive sharp waves and fibrillation potentials, small polyphasic nascent motor units, and chronically remodeled large motor unit action potentials. Subjects in the ETE group had significantly lower CMAP amplitudes and key pinch strength compared with the RETS and decompression group ($P < .001$; Table 3.2). This is due to more severe injury in the ETE group relative to the other 2 groups. In addition, pressure sensation was more severely impaired in the ETE transfer subjects compared with the decompression group ($P < .01$) but was not significantly different from the RETS group (see Table 3.2). Owing to the differences in mechanisms of injury and baseline characteristics of the ETE

group, comparisons of postsurgical outcomes were made only between the decompression and the RETS group.

TABLE 3.2 BASELINE COMPARISON OF OUTCOMES BETWEEN EXPERIMENTAL GROUPS.

	AIN RETS	AIN ETE	Decompression
CMAP (mV)	2.6 ± 3.2	0.2 ± 0.6[a]	3.3 ± 3.1
Key pinch (kg)	2.18 ± 2.37	0.48 ± 0.90[a]	3.06 ± 2.04
SWMT (size)	4.38 ± 1.08	6.30 ± 0.85[β]	3.65 ± 0.58

AIN, anterior interosseous nerve; CMAP, compound muscle action potential; ETE, end-to-end; RETS, Reverse end-to-side; SWMT, Semmes-Weinstein Monofilament test.
[a]denotes statistical significance ($P < .05$) between the AIN ETE and the other 2 groups, while β denotes a significant difference between the AIN ETE and the decompression group.
Size of the monofilaments denotes $\log_{10}$ of their bending force measured in gram. All data presented as mean ± SD.

3.4.3 Source of Axonal Regeneration in AIN RETS

Based on the results from the digital datapoint-by-datapoint subtraction, none of the subjects in the RETS group showed any evidence of axonal growth from the AIN into the ADM muscle with a mean CMAP amplitude of 0.0 mV at all postsurgical follow-up time points. A typical example is shown in Figure 3.2A. In all cases, the ADM muscle was entirely innervated by motor axons from the ulnar nerve. By contrast, nerve regeneration from the AIN to the distal stump of the ulnar nerve was found in all subjects in the ETE group. A representative example of that is shown in Figure 3.2B.
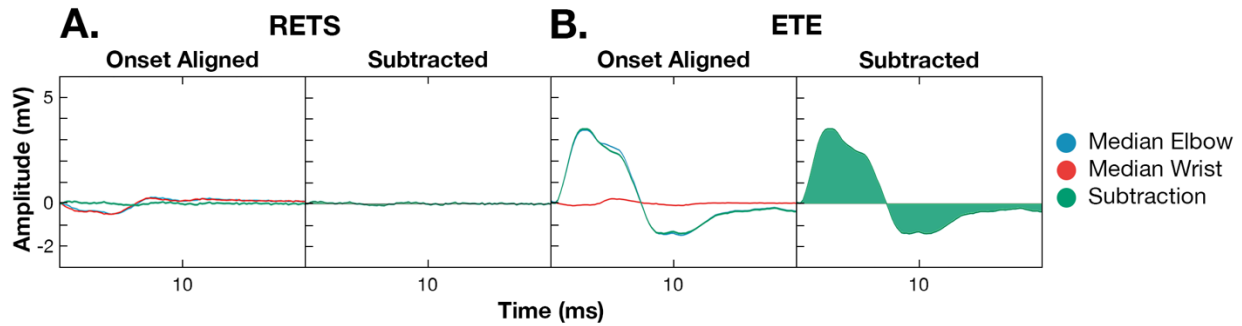
FIGURE 3.2 A REPRESENTATIVE RECORDING FROM A RETS SUBJECT AT YEAR 3 USING DIGITAL SUBTRACTION.

(a) In this case, the digitally subtracted waveform between the median wrist and elbow compound muscle action potentials was flat, indicating that there was no crossover innervation from the anterior interosseous nerve which was the case in all RETS subjects in this study. (b) By contrast, in a representative end-to-end subject at year 3, nerve regeneration from the anterior interosseous nerve to the distal stump of the ulnar nerve was found. ETS, end-to-side; RETS, reverse end-to-side.

3.4.4 Comparison of Postsurgical Outcomes Between RETS and Decompression

The results of the physiological and functional outcomes are presented in Table 3.3. There was no significant change in CMAP in the RETS and decompression groups even after 3 years. It went from 2.6 ± 3.2 to 3.2 ± 2.5 mV in the RETS group while in the decompression group from 3.3 ± 3.1 to 4.0 ± 3.3 mV. The strength recovery of the first dorsal interosseous and adductor pollicis muscles as reflected by the key pinch test showed significant improvement in the RETS groups but not in the decompression group. By year 3, there was a 2.6-fold increase (from 1.56 ± 0.85 to 4.13 ± 2.52 kg; P < .01) in the RETS group, whereas the change from 3.11 ± 2.09 at baseline to 4.05 ± 2.20 kg at year 3 in the decompression group was not significant. Potential reasons for this discrepancy will be further explored in the next section. For sensory recovery, there was no significant change in

the Semmes-Weinstein monofilament scores in the RETS (from 4.60 ± 1.08 to 3.83 ± 1.21) and

decompression group (from 3.65 ± 0.58 to 3.44 ± 0.29).

TABLE 3.3 COMPARISON OF POSTOPERATIVE OUTCOMES BETWEEN THE RETS AND DECOMPRESSION GROUPS.

Outcome	Group	Baseline	Year 1	Year 2	Year 3
CMAP (mV)	AIN RETS (n = 25)	2.6 ± 3.2	2.4 ± 3.1	2.6 ± 3.1	3.2 ± 2.5
	Decompression (n = 21)	3.3 ± 3.1	3.3 ± 3.1	3.6 ± 3.3	4.0 ± 3.3
Key pinch (kg)	AIN RETS	2.18 ± 2.37	3.64 ± 2.66 *	4.06 ± 3.09 *	4.13 ± 2.52[a]
	Decompression	3.06 ± 2.04	3.36 ± 2.22	3.84 ± 2.39	3.95 ± 2.20
SWMT (size)	AIN RETS	4.38 ± 1.08	4.24 ± 0.92	4.26 ± 0.96	4.01 ± 0.89
	Decompression	3.65 ± 0.58	3.54 ± 0.42	3.52 ± 0.38	3.44 ± 0.29

AIN, anterior interosseous nerve; CMAP, compound muscle action potential; ETE, end-to-end; RETS, Reverse end-to-side; SWMT, Semmes-Weinstein Monofilament Test.
[a]denotes that the outcomes were statistically different from baseline within the RETS or decompression groups.
Outcomes reported in mean ± SD. Year 3 outcomes were not statistically different between RETS and decompression

3.4.5 Physiological and Functional Outcomes in the ETE Group

Motor reinnervation as revealed by a gradual increase in the CMAP amplitudes in the ADM

muscle after surgery was presented (see Table 3.4). This reached significance after the first year, and

even greater gains were observed in subsequent years. By year 3, the mean CMAP amplitude

increased from 0.2 ± 0.6 mV at baseline to 3.3 ± 0.6 mV (P < .01). The strength recovery of key

pinch test also showed significant improvement. There was a 6.3–fold increase (from 0.48 ± 0.90 at

baseline to 3.00 ± 0.45 kg at year 3; P < .05). Although there was a gradual improvement of pressure

sensation, it took substantially longer than muscle reinnervation. The improvement became

significant by year 3 (from 6.30 ± 0.35 to 4.48 ± 0.25 monofilament size; P < .05).

TABLE 3.4 PHYSIOLOGICAL AND FUNCTIONAL OUTCOMES IN THE AIN ETE TRANSFER GROUP.

Group	Outcome	Baseline	Year 1	Year 2	Year 3
AIN ETE (n = 16)	CMAP (mV)	0.24 ± 0.60	1.20 ± 2.13[a]	2.04 ± 2.01[a]	3.34 ± 2.29[a]
	Key pinch (kg)	0.48 ± 0.90	2.49 ± 1.98[a]	3.19 ± 2.12[a]	3.00 ± 1.67[a]
	SWMT (size)	6.30 ± 0.85	5.49 ± 1.30	4.59 ± 0.93	4.48 ± 0.86[a]

[a]denotes outcomes that were statistically different from baseline.
Outcomes reported in mean ± SD. The abbreviations used are the same as in the previous tables.

3.5 Discussion

In this prospective nonrandomized cohort study, we could not find any evidence of crossover axonal regeneration from the AIN to reinnervate the ADM muscle in any of the AIN RETS subjects. The motor reinnervation seen in the RETS group is due to regeneration of the native ulnar axons, which occurred at a pace similar to that in the decompression group. However, subjects in the RETS group did show a significant increase in key pinch strength compared with baseline.

Of the 11 clinical trials published on AIN to ulnar nerve RETS transfer to date, 7 of them are retrospective chart reviews without a control group (Chen et al., 2021; Davidge et al., 2015; Dengler et al., 2020; Doherty et al., 2020; George, Burahee, Sanders, & Power, 2022; Head et al., 2020; McLeod et al., 2020).[9-11,13,16,21,22] This makes it difficult to firmly attribute whether the improvement seen is due to the nerve transfer itself or whether the improvement was only a result of the spontaneous recovery of the partially injured ulnar nerve. Although there was a control group in the studies by Flores (Flores, 2015) and Koriem et al (Koriem et al., 2020), they comprise patients with complete high ulnar nerve transection. The comparison may not be appropriate because the outcomes of nerve graft reconstruction in high ulnar nerve injury are known to be poor and are therefore not commonly performed. To date, probably, the strongest clinical data for the potential efficacy of RETS to improve hand function are from a large prospective randomized controlled trial by Xie et al (Xie et al., 2021). However, the mean CMAP (primary outcome at the final time point) reported in the RETS group at 17.17 mV is well above that seen in healthy subjects (12.9 mV) (Kim, 2011). Biologically, it is difficult to reconcile this electrophysiological discrepancy. Further verification of this will be needed in future studies.

This study differs from previous RETS studies in 1 major respect. Although crossover regeneration from the AIN into the ulnar nerve through a perineurial window is a major premise of the technique, this has not been evaluated in previous studies. We specifically quantified this but could not find any evidence of innervation in the ADM muscle by the AIN even up to 3 years after the procedure.

To evaluate the babysitting effect that infers a preservation effect on the target tissue, we compared results from the RETS group with patients who underwent only decompression of the ulnar nerve at the elbow and found no difference in the extent of reinnervation between the groups. However, patients in the RETS group did show a significant improvement in key pinch strength. Based on our results, this unexpected finding cannot be attributed to greater nerve regeneration. Rather, it may have a number of other plausible explanations. One possibility is that in addition to nerve transfer, the ulnar nerve was also released at Guyon canal which is another well-known potential compression site (Brubacher & Leversedge, 2017). A second potential explanation is the presence of frequency-dependent conduction block. This has been shown to occur in focal compression neuropathies resulting in failure of action potential propagation and weakness during tetanic contraction (Watson & Doherty, 2010). This may also explain the lack of improvement in pinch strength seen in the chronic compression injury group who only had decompression at the elbow.

Limitations

A major limitation of this study is that the allocation of treatment was not randomized that could result in observer bias. However, because the indications for each of the treatments are different, it is difficult to ethically justify assigning surgery such as RETS to patients with high ulnar transection on the basis of clinical equipoise. A second constraint is the lack of blinding. This has

not been performed in any of the clinical trials published because the surgical scars make the type of procedures performed obvious. We tried to mitigate this by using objective physiological outcomes including neurophysiology that are less prone to subjective interpretation and bias. Third, because technically it is more difficult to accurately quantify crossover reinnervation from the AIN to the first dorsal interosseous muscle because of contemplation of the much larger CMAP from the neighboring median innervated thenar muscles, we elected to focus on the ADM muscle which is much less affected by volume conduction. A further rationale for this decision is also based on prior observations that with the shorter distance from the nerve transfer site, reinnervation was found to be more robust in the ADM muscle (Head et al., 2020). Finally, although motor unit number estimation would be a more accurate measure of nerve regeneration, it is a specialized technique that cannot be feasibly performed across all study sites. Furthermore, in a previous study, the pattern of postoperative changes in CMAP and motor unit action potentials in patients with cubital tunnel syndrome was found to be very similar, with reinnervation occurring very slowly over years (Power, Morhart, Olson, & Chan, 2019).

3.6 Conclusion

Although RETS nerve transfer surgery has become increasingly popular, the results from published clinical trials are conflicting, in part because crossover regeneration from the donor nerve has never been measured. In this multicentre prospective study, we found that unlike patients with ETE nerve transfers, there was no crossover regeneration in any of the subjects in the RETS group. The extent of reinnervation was also no different from decompression surgery alone. Based on these findings, the justifications for this surgical technique need to be carefully re-evaluated.

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3.8 Disclosures

The authors have no personal, financial, or institutional interest in any of the drugs, materials, or devices described in this article.

3.9 References

An TW, Evanoff BA, Boyer MI, Osei DA. The prevalence of cubital tunnel syndrome: a cross-sectional study in a U.S. metropolitan cohort. J Bone Joint Surg Am. 2017;99(5):408-416.

Tong J, Xu B, Dong Z, Zhang C, Gu Y. Surgical outcome for severe cubital tunnel syndrome in patients aged >70 years: a mean follow-up of 4.5 years. Acta Neurochir. 2017;159(5):917-923.

Rosberg HE, Carlsson KS, Hojgard S, Lindgren B, Lundborg G, Dahlin LB. Injury to the human median and ulnar nerves in the forearm—analysis of costs for treatment and rehabilitation of 69 patients in southern Sweden. J Hand Surg Br. 2005;30(1):35-39.

Haase SC, Chung KC. Anterior interosseous nerve transfer to the motor branch of the ulnar nerve for high ulnar nerve injuries. Ann Plast Surg. 2002;49(3):285-290.

Flores LP. Comparativestudyofnervegraftingversusdistalnervetransferfortreatment of proximal injuries of the ulnar nerve. J Reconstr Microsurg. 2015;31(9):647-653.

Isaacs J, Allen D, Chen LE, Nunley J 2nd. Reverse end-to-side neurotization. J Reconstr Microsurg. 2005;21(1):43-48.

Kale SS, Glaus SW, Yee A, et al. Reverse end-to-side nerve transfer: from animal model to clinical use. J Hand Surg Am. 2011;36(10):1631-1639.e1632.

Barbour J, Yee A, Kahn LC, Mackinnon SE. Supercharged end-to-side anterior interosseous to ulnar motor nerve transfer for intrinsic musculature reinnervation. J Hand Surg Am. 2012;37(10):2150-2159.

Dengler J, Dolen U, Patterson JMM, et al. Supercharge end-to-side anterior interosseous-to-ulnar motor nerve transfer restores intrinsic function in cubital tunnel syndrome. Plast Reconstr Surg. 2020;146(4):808-818.

McLeod GJ, Peters BR, Quaife T, Clark TA, Giuffre JL. Anterior interosseous-to-ulnar motor nerve transfers: a single center's experience in restoring intrinsic hand function. Hand (N Y). 2022;17(4):609-614.

Doherty CD, Miller TA, Larocerie-Salgado J, Byers BA, Ross DC. Reverse end-to-side anterior interosseous nerve-to-ulnar motor transfer for severe ulnar neuropathy. Plast Reconstr Surg. 2020;146(3):306e-313e.

Xie Q, Shao X, Song X, et al. Ulnar nerve decompression and transposition with versus without supercharged end-to-side motor nerve transfer for advanced cubital tunnel syndrome: a randomized comparison study. J Neurosurg. 2021;136(3): 845-855.

Head LK, Zhang ZZ, Hicks K, Wolff G, Boyd KU. Evaluation of intrinsic hand musculature reinnervation following supercharge end-to-side anterior interosseous to ulnar motor nerve transfer. Plast Reconstr Surg. 2020;146(1): 128-132.

Koriem E, El-Mahy MM, Atiyya AN, Diab RA. Comparison between super-charged ulnar nerve repair by anterior interosseous nerve transfer and isolated ulnar nerve repair in proximal ulnar nerve injuries. J Hand Surg Am. 2020;45(2): 104-110.

Baltzer H, Woo A, Oh C, Moran SL. Comparison of ulnar intrinsic function following supercharge end-to-side anterior interosseous-to-ulnar motor nerve transfer: a matched cohort study of proximal ulnar nerve injury patients. Plast Reconstr Surg. 2016;138(6):1264-1272.

Davidge KM, Yee A, Moore AM, Mackinnon SE. The Supercharge end-to-side anterior interosseous-to-ulnar motor nerve transfer for restoring intrinsic function: clinical experience. Plast Reconstr Surg. 2015;136(3):344e-352e.

Pondaag W, Gilbert A. Results of end-to-side nerve coaptation in severe obstetric brachial plexus lesions. Neurosurgery. 2008;62(3):656-663.

Pienaar C, Swan MC, De Jager W, Solomons M. Clinical experience with end-to-side nerve transfer. J Hand Surg Br. 2004;29(5):438-443.

Wu S, Curran MWT, Hachisuka A, Rajshekar M, Chan KM. A new method to quantify innervation of the ulnar intrinsic hand muscles by the anterior interosseous nerve in Martin–Gruber anastomosis and nerve transfer surgery. Muscle Nerve. 2022;66(3):297-303.

Curran MWT, Morhart MJ, Olson JL, Hachisuka A, Chan KM. Acetyl-l-carnitine to enhance nerve regeneration in carpal tunnel syndrome: a double-blind, randomized controlled trial. Plast Reconstr Surg. 2019;143(1): 111e-120e.

Chen SH, Mao SH, Lan CY, et al. End-to-side anterior interosseous nerve transfer: a valuable alternative for traumatic high ulnar nerve palsy. Ann Plast Surg. 2021; 86(2S suppl 1):S102-S107.

George SC, Burahee AS, Sanders AD, Power DM. Outcomes of anterior interosseous nerve transfer to restore intrinsic muscle function after high ulnar nerve injury. J Plast Reconstr Aesthet Surg. 2022;75(2):703-710.

Kim DH. Ulnar nerve conduction study of the first dorsal interosseous muscle in Korean subjects. Ann Rehabil Med. 2011;35(5):658-663.

Brubacher JW, Leversedge FJ. Ulnar neuropathy in cyclists. Hand Clin. 2017; 33(1):199-205.

Watson BV, Doherty TJ. Frequency-dependent conduction block in ulnar neuropathy localized to the elbow. Clin Neurophysiol. 2010;121(12): 2111-2116.

Power HA, Morhart MJ, Olson JL, Chan KM. Postsurgical electrical stimulation enhances recovery following surgery for severe cubital tunnel syndrome: a double-blind randomized controlled trial. Neurosurgery. 2020;86(6): 769-777.

3.10 Acknowledgments

3.11 Comment

Reverse end-to-side (RETS) is one of several end-to-side (ETS) nerve reconstruction strategies that involve the creation of a perineurial window in the side of a recipient nerve to which a transected donor nerve is coaptated. The theoretical benefits of ETS over traditional end-to-end (ETE) transfers include reduced morbidity of the donor nerve, potentially allowing both donor and native axons to grow through the site of the transfer, as well as a possible "babysitting effect" by which the presence of healthy donor axons enhances regeneration of the injured nerve.

Outcomes after proximal ulnar injury are notoriously poor, resulting in potentially debilitating hand weakness. With the goal of augmenting hand recovery, some have advocated for ETS transfer of the pronator quadratus branch of the anterior interosseous nerve (AIN) to the distal

ulnar nerve. However, no studies have demonstrated primacy of ETS over ETE or even ulnar decompression.

In this timely and relevant study, the authors quantify the relative contribution of the native ulnar and donor AIN nerves to ulnar-innervated hand muscles in patients who underwent AIN to distal ulnar ETS or ETE. Interestingly, while the ETE group did have AIN contribution to ulnar-innervated muscles, the ETS group had only ulnar contribution, suggesting no donor axon regeneration in the ETS group. Moreover, the authors found minimal difference in clinical outcomes between those who underwent ETS and those who underwent ulnar decompression alone, suggesting no clinically meaningful "babysitting effect." These data necessitate a careful re-evaluation of the utility of ETS techniques for nerve reconstruction.

— *Whitney E. Muhlestein Lynda J.-S. Yang Ann Arbor, Michigan, USA*

Chapter 4

Conclusion, Limitations, and Future Directions

4.1 Conclusion

It has been seventeen years since the RETS nerve transfer was conceived in the rodent model. The original intention of RETS to expand on the pool of regenerating axons, hinges on the presence of axonal crossover through an epineurial or perineurial window. While this has been demonstrated in rodent models using retrograde labels, RETS has experienced an astonishing pace of translation and is now used in many clinical settings across North America, Europe, and Asia, despite the lack of evidence for crossover regeneration in humans. Although its initial therapeutic applications were to augment a depleting pool of regenerating axons, the current enthusiasm in the literature and clinics may be overtaking the current evidence. This is apparent in that clinical scenarios in which RETS was never intended, are being recommended.

To methodically assess whether crossover regeneration occurs in RETS nerve transfers in humans, we constructed an objective electrophysiological technique that could quantify the degree of axonal crossover called digital subtraction. Using ETE nerve transfer and MGA patients, we compared the digital subtraction technique to previous techniques for measuring crossover. We found that the digital subtraction technique was more accurate and easier to implement using standard electrophysiological equipment. Armed with an objective tool to interrogate axonal crossover, we implemented this technique as our primary outcome measure in a multicentre prospective cohort study on patients with severe ulnar neuropathy. The effects of axonal crossover were assessed by comparing patients who underwent RETS and ETE AIN to ulnar nerve transfers. In addition to growing the pool of regenerating axons, the RETS transfer also potentiates the baby-sitting. An end-organ preserving phenomenon which occurs through an as yet unknown mechanism. To measure the effects of baby-sitting, the RETS group was compared to patients that had only underwent decompression surgery. Not only was crossover regeneration from the donor nerve not

present in the RETS group, electrophysiological and functional outcomes were similar between RETS and decompression alone. The latter indicates that there was no baby-sitting effect in the RETS patients.

4.2 Limitations

A physiological limitation to the digital subtraction technique is temporal dispersion and phase cancellation. With the longer distance from the recording electrode, the CMAP evoked at the elbow is slightly smaller and broader than that evoked at the wrist. However, in healthy subjects, the difference is relatively small. Furthermore, this physiological constraint is not unique to digital subtraction as it would also apply to the other techniques as well. A second limitation is a practical one in that the digital subtraction technique is currently not widely available on commercial EMG machines. However, since the algorithm for onset alignment and datapoint-by-datapoint subtraction is relatively simple, it can be easily adopted on commercial EMG machines without the need for additional hardware.

A major limitation of the multicentre prospective clinical trial was that the allocation of treatment was not randomized that could result in observer bias. However, because the indications for each of the treatments was different, it was difficult to ethically justify assigning surgery such as RETS to patients with high ulnar transection based on clinical equipoise. A second constraint was the lack of blinding. This has not been performed in any of the clinical trials published because the surgical scars make the type of procedures performed obvious. We attempted to mitigate this by using objective physiological outcomes including neurophysiology that are less prone to subjective interpretation and bias. Third, because technically it is more difficult to accurately quantify crossover reinnervation from the AIN to the first dorsal interosseous muscle because of contemplation of the

much larger CMAP from the neighboring median innervated thenar muscles, we elected to focus on the ADM muscle which is much less affected by volume conduction. A further rationale for this decision is also based on prior observations that with the shorter distance from the nerve transfer site, reinnervation was found to be more robust in the ADM muscle. Finally, although motor unit number estimation would be a more accurate measure of nerve regeneration, it is a specialized technique that cannot be feasibly performed across all study sites. Furthermore, in a previous study, the pattern of postoperative changes in CMAP and motor unit action potentials in patients with cubital tunnel syndrome was found to be very similar, with reinnervation occurring very slowly over years.

4.3 Future Directions

Using retrograde labels, axonal crossover in rodent models is well supported in the literature. While retrograde labels are not feasible in humans, methods like motor unit number estimates and digital subtraction, which measure axonal reinnervation and crossover regeneration respectively, exist as suitable surrogates. These techniques represent important outcome measures in blinded and randomized clinical studies. While the application of these techniques in clinical trials can help us determine the efficacy of RETS nerve transfer, or lack thereof, they fail to deduce the underlying physiology by which crossover regeneration across epineurial and perineurial tissues and the baby-sitting effect occur. Further basic science investigation in molecular underpinnings of the axon's ability to traverse nervous connective tissues and its muscle preserving effects will give us a fundamental mechanistic understanding of the RETS nerve transfer. A comprehensive understanding of RETS will bolster the literature on nerve transfers, ensure proper clinical translation of the surgery, and improve outcomes for patients with severe nerve injuries.

Bibliography

Amoiridis, G., & Vlachonikolis, I. G. (2003). Verification of the median-to-ulnar and ulnar-to-median nerve motor fiber anastomosis in the forearm: An electrophysiological study. *Clinical Neurophysiology*, *114*, 94–98.

Amr, S. M., & Moharram, A. N. (2005). Repair of brachial plexus lesions by end-to-side side-to-side grafting neurorrhaphy: Experience based on 11 cases. *Microsurgery*, *25*, 126–146.

Amr, S. M., Moharram, A. N., & Abdel-Meguid, K. M. S. (2006). Augmentation of partially regenerated nerves by end-to-side side-to-side grafting neurotization: Experience based on eight late obstetric brachial plexus cases. *Journal of Brachial Plexus and Peripheral Nerve Injury*, *1*, 6–6. PubMed (17147803).

An, T. W., Evanoff, B. A., Boyer, M. I., & Osei, D. A. (2017). The Prevalence of Cubital Tunnel Syndrome: A Cross-Sectional Study in a U.S. Metropolitan Cohort. *The Journal of Bone and Joint Surgery. American Volume*, *99*, 408–416.

Anastakis, D. J., Malessy, M. J. A., Chen, R., Davis, K. D., & Mikulis, D. (2008). Cortical plasticity following nerve transfer in the upper extremity. *Hand Clinics*, *24*, 425–444, vi–vii.

Angius, D., Wang, H., Spinner, R. J., Gutierrez-Cotto, Y., Yaszemski, M. J., & Windebank, A. J. (2012). A systematic review of animal models used to study nerve regeneration in tissue-engineered scaffolds. *Biomaterials*, *33*, 8034–8039.

Artiaco, S., Tos, P., Conforti, L. G., Geuna, S., & Battiston, B. (2010). Termino-lateral nerve suture in lesions of the digital nerves: Clinical experience and literature review. *Journal of Hand Surgery (European Volume)*, *35*, 109–114.

Babcock, W. W. (1927). A standard technique for operations on peripheral nerves with especial reference to the closure of large gaps. *Surgery, Gynecology & Obstetrics*, *45*, 78.

Baltzer, H., Woo, A., Oh, C., & Moran, S. L. (2016). Comparison of Ulnar Intrinsic Function following Supercharge End-to-Side Anterior Interosseous–to–Ulnar Motor Nerve Transfer: A Matched

Cohort Study of Proximal Ulnar Nerve Injury Patients. *Plastic and Reconstructive Surgery*, *138*, 1264–1272.

Bao, B., Wei, H., Zhu, H., & Zheng, X. (2022). Transfer of Soleus Muscular Branch of Tibial Nerve to Deep Fibular Nerve to Repair Foot Drop After Common Peroneal Nerve Injury: A Retrospective Study. *Frontiers in Neurology*, *13*, 745746.

Barbour, J., Yee, A., Kahn, L. C., & Mackinnon, S. E. (2012). Supercharged End-to-Side Anterior Interosseous to Ulnar Motor Nerve Transfer for Intrinsic Musculature Reinnervation. *The Journal of Hand Surgery*, *37*, 2150–2159.

Beris, A. E., & Lykissas, M. G. (2009). Chapter 13 Experimental Results in End-To-Side Neurorrhaphy. In *International Review of Neurobiology* (Vol. 87, pp. 269–279). Academic Press.

Bray, D., Thomas, C., & Shaw, G. (1978). Growth cone formation in cultures of sensory neurons. *Proceedings of the National Academy of Sciences*, *75*, 5226–5229.

Brubacher, J. W., & Leversedge, F. J. (2017). Ulnar Neuropathy in Cyclists. *Hand Clinics*, *33*, 199–205.

Brushart, T. M., Hoffman, P. N., Royall, R. M., Murinson, B. B., Witzel, C., & Gordon, T. (2002). Electrical Stimulation Promotes Motoneuron Regeneration without Increasing Its Speed or Conditioning the Neuron. *The Journal of Neuroscience*, *22*, 6631.

Buterbaugh, K. L., & Shah, A. S. (2016). The natural history and management of brachial plexus birth palsy. *Current Reviews in Musculoskeletal Medicine*, *9*, 418–426.

Cajal, S. R. y. (1991). *Cajal's Degeneration and Regeneration of the Nervous System* (J. DeFelipe & E. G. Jones, Eds.; R. M. May, Trans.). New York: Oxford University Press.

Cederna, P., Kalliainen, L., Urbanchek, M., M. Rovak, J., & Kuzon, W. (2001). "Donor" Muscle Structure and Function after End-to-Side Neurorrhaphy. *Plastic and Reconstructive Surgery*, *107*, 789–796.

Chan, K. M., Olson, J. L., Morhart, M., Lin, T., & Guilfoyle, R. (2014). Outcomes of Nerve Transfer versus Nerve Graft in Ulnar Nerve Laceration. *Canadian Journal of Neurological Sciences / Journal Canadien Des Sciences Neurologiques*, *39*, 242–244.

Chen, S.-H., Mao, S.-H., Lan, C.-Y., Huang, R.-W., Lee, C.-H., Hsu, C.-C., … Chuang, D. C.-C. (2021). End-to-Side Anterior Interosseous Nerve Transfer: A Valuable Alternative for Traumatic High Ulnar Nerve Palsy. *Annals of Plastic Surgery*, *86*, S102–S107.

Cheng, L., Khan, M., & Mudge, A. W. (1995). Calcitonin gene-related peptide promotes Schwann cell proliferation. *The Journal of Cell Biology*, *129*, 789–796.

Cleveland, D. W., & Hoffman, P. N. (1991). Neuronal and glial cytoskeletons. *Current Opinion in Neurobiology*, *1*, 346–353.

Coroneos, C. J., Voineskos, S. H., Christakis, M. K., Thoma, A., Bain, J. R., & Brouwers, M. C. (2017). Obstetrical brachial plexus injury (OBPI): Canada's national clinical practice guideline. *BMJ Open*, *7*, e014141.

Curran, M. W. T., Morhart, M. J., Olson, J. L., Hachisuka, A., & Chan, K. M. (2019). Acetyl-L-Carnitine to Enhance Nerve Regeneration in Carpal Tunnel Syndrome: A Double-Blind, Randomized, Controlled Trial. *Plastic and Reconstructive Surgery*, *143*, 111e–120e.

Davidge, K. M., Yee, A., Moore, A. M., & Mackinnon, S. E. (2015). The Supercharge End-to-Side Anterior Interosseous–to–Ulnar Motor Nerve Transfer for Restoring Intrinsic Function: Clinical Experience. *Plastic and Reconstructive Surgery*, *136*, 344e–352e.

Davis, L., Dou, P., DeWit, M., & Kater, S. B. (1992). Protein synthesis within neuronal growth cones. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *12*, 4867–4877.

Dengler, J., Dolen, U., Patterson, J. M. M., Davidge, K. M., Kahn, L. C., Yee, A., & Mackinnon, S. E. (2020). Supercharge End-to-Side Anterior Interosseous–to–Ulnar Motor Nerve Transfer Restores Intrinsic Function in Cubital Tunnel Syndrome. *Plastic and Reconstructive Surgery*, *146*.

Doherty, C. D., Miller, T. A., Larocerie-Salgado, J., Byers, B. A., & Ross, D. C. (2020). Reverse End-to-Side Anterior Interosseous Nerve-to-Ulnar Motor Transfer for Severe Ulnar Neuropathy. *Plastic and Reconstructive Surgery*, *146*, 306e–313e.

Dorsi, M. J., Hsu, W., & Belzberg, A. J. (2010). Epidemiology of brachial plexus injury in the pediatric multitrauma population in the United States: Clinical article. *Journal of Neurosurgery: Pediatrics PED*, *5*, 573–577.

Dunlop, R. L. E., Wormald, J. C. R., & Jain, A. (2019). Outcome of surgical repair of adult digital nerve injury: A systematic review. *BMJ Open*, *9*, e025443.

Farber, S. J., Glaus, S. W., Moore, A. M., Hunter, D. A., Mackinnon, S. E., & Johnson, P. J. (2013). Supercharge Nerve Transfer to Enhance Motor Recovery: A Laboratory Study. *The Journal of Hand Surgery*, *38*, 466–477.

Ferraresi, S., Garozzo, D., Ravenni, R., Dainese, R., De Grandis, D., & Buffatti, P. (2002). Hemihypoglossal Nerve Transfer in Brachial Plexus Repair: Technique and Results. *Neurosurgery*, *50*, 332–335.

Flores, L. P. (2015). Comparative Study of Nerve Grafting versus Distal Nerve Transfer for Treatment of Proximal Injuries of the Ulnar Nerve. *J Reconstr Microsurg*, *31*, 647–653.

Franciosi, L. F., Modestti, C., & Mueller, S. F. (1998). Neurotization of the biceps muscle by end-to-side neurorraphy between ulnar and musculocutaneous nerves: A series of five cases. *Annales de Chirurgie de La Main et Du Membre Supérieur*, *17*, 362–367.

Fu, S. Y., & Gordon, T. (1995). *Contributing factors to poor functional recovery after delayed nerve repair: Prolonged denervation. 15*, 3886–3895.

Fu, Susan Y., & Gordon, T. (1997). The cellular and molecular basis of peripheral nerve regeneration. *Molecular Neurobiology*, *14*, 67–116.

Garg, R., Merrell, G. A., Hillstrom, H. J., & Wolfe, S. W. (2011). Comparison of nerve transfers and nerve grafting for traumatic upper plexus palsy: A systematic review and analysis. *The Journal of Bone and Joint Surgery. American Volume*, *93*, 819–829.

Gaudet, A. D., Popovich, P. G., & Ramer, M. S. (2011). Wallerian degeneration: Gaining perspective on inflammatory events after peripheral nerve injury. *Journal of Neuroinflammation*, *8*, 110.

George, S. C., Burahee, A. S., Sanders, A. D., & Power, D. M. (2022). Outcomes of anterior interosseous nerve transfer to restore intrinsic muscle function after high ulnar nerve injury. *Journal of Plastic, Reconstructive & Aesthetic Surgery : JPRAS*, *75*, 703–710.

Gesslbauer, B., Hruby, L. A., Roche, A. D., Farina, D., Blumer, R., & Aszmann, O. C. (2017). Axonal components of nerves innervating the human arm. *Annals of Neurology*, *82*, 396–408.

Gordon, T., & Stein, R. B. (1982). Time course and extent of recovery in reinnervated motor units of cat triceps surae muscles. *The Journal of Physiology*, *323*, 307–323.

Gordon, T., & Tyreman, N. (2010). Sprouting capacity of lumbar motoneurons in normal and hemisected spinal cords of the rat. *The Journal of Physiology*, *588*, 2745–2768.

Gordon, Tessa, Sulaiman, O., & Boyd, J. G. (2003). Experimental strategies to promote functional recovery after peripheral nerve injuries. *Journal of the Peripheral Nervous System : JPNS*, *8*, 236–250.

Grafstein, B. (1975). The nerve cell body response to axotomy. *Experimental Neurology*, *48*, 32–51.

Grossman, J. A. I., DiTaranto, P., Yaylali, I., Alfonso, I., Ramos, L. E., & Price, A. E. (2004). Shoulder Function Following Late Neurolysis and Bypass Grafting for Upper Brachial Plexus Birth Injuries. *The Journal of Hand Surgery: British & European Volume*, *29*, 356–358.

Gutmann, E., Gutmann, L., Medawar, P. B., & Young, J. Z. (1942). The Rate of Regeneration of Nerve. *Journal of Experimental Biology*, *19*, 14–44.

Haase, S., & C Chung, K. (2002). Anterior Interosseous Nerve Transfer to the Motor Branch of the Ulnar Nerve for High Ulnar Nerve Injuries. *Annals of Plastic Surgery*, *49*, 285–290.

Hayashi, A., Pannucci, C., Moradzadeh, A., Kawamura, D., Magill, C., Hunter, D. A., … Myckatyn, T. M. (2008). Axotomy or compression is required for axonal sprouting following end-to-side neurorrhaphy. *Experimental Neurology*, *211*, 539–550.

Head, L. K., Zhang, Z. Z., Hicks, K., Wolff, G., & Boyd, K. U. (2020). Evaluation of Intrinsic Hand Musculature Reinnervation following Supercharge End-to-Side Anterior Interosseous–to–Ulnar Motor Nerve Transfer. *Plastic and Reconstructive Surgery*, *146*.

Hildebrand, C., Mustafa, G. Y., & Waxman, S. G. (1986). Remodelling of internodes in regenerated rat sciatic nerve: Electron microscopic observations. *Journal of Neurocytology*, *15*, 681–692.

Höke, A., Gordon, T., Zochodne, D. W., & Sulaiman, O. A. R. (2002). A decline in glial cell-line-derived neurotrophic factor expression is associated with impaired regeneration after long-term Schwann cell denervation. *Experimental Neurology*, *173*, 77–85.

Huckhagel, T., Nüchtern, J., Regelsberger, J., & Lefering, R. (2018). Nerve injury in severe trauma with upper extremity involvement: Evaluation of 49,382 patients from the TraumaRegister DGU® between 2002 and 2015. *Scandinavian Journal of Trauma, Resuscitation and Emergency Medicine*, *26*, 76.

Isaacs, J. (2021). Reverse End-to-Side (Supercharging) Nerve Transfer: Conceptualization, Validation, and Translation. *HAND*, 1558944720988076.

Isaacs, J., Allen, D., Chen, L. E., & Nunley, J. (2005). Reverse End-to-Side Neurotization. *Journal of Reconstructive Microsurgery.*, *21*, 43–48.

Isaacs, J. E., Cheatham, S., Gagnon, E. B., Razavi, A., & McDowell, C. L. (2008). Reverse End-to-Side Neurotization in a Regenerating Nerve. *Journal of Reconstructive Microsurgery.*, *24*, 489–496.

Jaeger, M., Braga Silva, J., Gehlen, D., Pereira Filho, G., Zettler, C., Antonieta Lopes de Souza, M., … Sebben, A. (2011). End-to-End Versus End-to-Side Motor and Sensory Neurorrhaphy in the Repair of the Acute Muscle Denervation. *Annals of Plastic Surgery*, *67*, 391–396.

Kale, S. S., Glaus, S. W., Yee, A., Nicoson, M. C., Hunter, D. A., Mackinnon, S. E., & Johnson, P. J. (2011). Reverse End-to-Side Nerve Transfer: From Animal Model to Clinical Use. *The Journal of Hand Surgery*, *36*, 1631-1639.e2.

Karsy, M., Watkins, R., Jensen, M. R., Guan, J., Brock, A. A., & Mahan, M. A. (2019). Trends and Cost Analysis of Upper Extremity Nerve Injury Using the National (Nationwide) Inpatient Sample. *World Neurosurgery*, *123*, e488–e500.

Kattla, S., & Lowery, M. M. (2010). Fatigue related changes in electromyographic coherence between synergistic hand muscles. *Experimental Brain Research*, *202*, 89–99.

Kim, D. H. (2011). Ulnar nerve conduction study of the first dorsal interosseous muscle in korean subjects. *Annals of Rehabilitation Medicine*, *35*, 658–663.

Kimura, J., Murphy, M. J., & Varda, D. J. (1976). Electrophysiological Study of Anomalous Innervation of Intrinsic Hand Muscles. *Archives of Neurology*, *33*, 842–844.

Koriem, E., El-Mahy, M. M., Atiyya, A. N., & Diab, R. A. (2020). Comparison Between Supercharged Ulnar Nerve Repair by Anterior Interosseous Nerve Transfer and Isolated Ulnar Nerve Repair in Proximal Ulnar Nerve Injuries. *The Journal of Hand Surgery*, *45*, 104–110.

Kuliasha, C., Kuliasha, C., Spearman, B., Atkinson, E. W., Furniturewalla, A., Rustogi, P., … Judy, J. (2018). *ROBUST AND SCALABLE TISSUE-ENGINEERINED ELECTRONIC NERVE INTERFACES (TEENI)* (p. 49).

Ladak, A., Morhart, M., O'Grady, K., Wong, J. N., Chan, K. M., Watt, M. J., & Olson, J. L. (2013). Distal Nerve Transfers Are Effective in Treating Patients with Upper Trunk Obstetrical Brachial Plexus Injuries: An Early Experience. *Plastic and Reconstructive Surgery*, *132*.

Latinovic, R., Gulliford, M. C., & Hughes, R. A. C. (2006). Incidence of common compressive neuropathies in primary care. *Journal of Neurology, Neurosurgery & Psychiatry*, *77*, 263.

Leechavengvongs, S., Ngamlamiat, K., Malungpaishrope, K., Uerpairotkit, C., Witoonchart, K., & Kulkittiya, S. (2011). End-to-Side Radial Sensory to Median Nerve Transfer to Restore Sensation and Relieve Pain in C5 and C6 Nerve Root Avulsion. *The Journal of Hand Surgery*, *36*, 209–215.

Liao, W.-C., Chen, J.-R., Wang, Y.-J., & Tseng, G.-F. (2009). The efficacy of end-to-end and end-to-side nerve repair (neurorrhaphy) in the rat brachial plexus. *Journal of Anatomy*, *215*, 506–521.

Lin, J. C., Schwentker-Colizza, A., Curtis, C. G., & Clarke, H. M. (2009). Final Results of Grafting versus Neurolysis in Obstetrical Brachial Plexus Palsy. *Plastic and Reconstructive Surgery*, *123*.

Lohman, R., Bullock, F., McNaughton, T., & Siemionow, M. (1997). End-to-end vs. End-to-side neurorrhaphy. *Journal of Reconstructive Microsurgery.*, *13*, 135–136.

Luff, A. R., Hatcher, D. D., & Torkko, K. (1988). Enlarged motor units resulting from partial denervation of cat hindlimb muscles. *Journal of Neurophysiology*, *59*, 1377–1394.

Lundborg, G. (2000). A 25-year perspective of peripheral nerve surgery: Evolving neuroscientific concepts and clinical significance. *The Journal of Hand Surgery*, *25*, 391–414.

Lundborg, G., Zhao, Q., Kanje, M., Danielsen, N., & Kerns, J. M. (1994). Can Sensory and Motor Collateral Sprouting be Induced from Intact Peripheral Nerve by End-to-Side Anastomosis? *Journal of Hand Surgery*, *19*, 277–282.

Mackinnon, S. E. (1988). Surgery of the peripheral nerve. *Carpal Tunnel Syndrome*, 146–169.

Mackinnon, S. E. (2013). In Reply. *The Journal of Hand Surgery*, *38*, 618–619.

Martin, S. P., Schauer, K. T., Czyrny, J. J., & Ablove, R. H. (2019). Electrophysiological Findings in Common Median-Ulnar Nerve Interconnections and Their Clinical Implications. *The Journal of Hand Surgery*, *44*, 884–894.

Matsis, R., Chou, J., & Clode, N. (2021). Outcome of carpal tunnel decompression with pre-surgical diagnosis determined on general practitioner assessment and nerve conduction study. *Journal of Clinical Orthopaedics and Trauma*, *13*, 15–18.

Matsuda, K., Kakibuchi, M., Sotsuka, Y., Kubo, T., Shibata, M., & Hosokawa, K. (2015). End-to-side "loop" graft for total facial nerve reconstruction: Over 10 years experience. *Journal of Plastic, Reconstructive & Aesthetic Surgery : JPRAS*, *68*, 1054–1063.

McLeod, G. J., Peters, B. R., Quaife, T., Clark, T. A., & Giuffre, J. L. (2020). Anterior Interosseous–to–Ulnar Motor Nerve Transfers: A Single Center's Experience in Restoring Intrinsic Hand Function. *HAND*, 1558944720928482.

Mennen, U. (1998). End-to-side nerve suture in the human patient. *Hand Surgery*, *03*, 7–15.

Mennen, U. (2003). End-to-side Nerve Suture in Clinical Practice. *Hand Surgery*, *08*, 33–42.

Mennen, U., van der Westhuizen, M., & Eggers, I. (2003). Re-innervation of M. Biceps by End-to-side Nerve Suture. *Hand Surgery*, *08*, 25–31.

Mohanty, C. B., Bhat, D., & Devi, B. I. (2015). Role of Central Plasticity in the Outcome of Peripheral Nerve Regeneration. *Neurosurgery*, *77*, 418–423.

Moldovan, M., Sørensen, J., & Krarup, C. (2006). Comparison of the fastest regenerating motor and sensory myelinated axons in the same peripheral nerve. *Brain*, *129*, 2471–2483.

Morris, J. H., Hudson, A. R., & Weddell, G. (1972). A study of degeneration and regeneration in the divided rat sciatic nerve based on electron microscopy. II. The development of the "regenerating unit". *Zeitschrift Fur Zellforschung Und Mikroskopische Anatomie (Vienna, Austria : 1948)*, *124*, 103–130.

Nadi, M., Ramachandran, S., Islam, A., Forden, J., Guo, G. F., & Midha, R. (2018). Testing the effectiveness and the contribution of experimental supercharge (reversed) end-to-side nerve transfer. *Journal of Neurosurgery*, *130*, 702–711.

Nath, R. K., Lyons, A. B., & Paizi, M. (2008). Successful management of foot drop by nerve transfers to the deep peroneal nerve. *Journal of Reconstructive Microsurgery*, *24*, 419–427.

Noble, J., Munro, C. A., Prasad, V. S. S. V., & Midha, R. (1998). *Analysis of Upper and Lower Extremity Peripheral Nerve Injuries in a Population of Patients with Multiple Injuries*. *45*, 116–122.

Novak, C. B., & Mackinnon, S. E. (2002). Distal Anterior Interosseous Nerve Transfer to the Deep Motor Branch of the Ulnar Nerve for Reconstruction of High Ulnar Nerve Injuries. *Journal of Reconstructive Microsurgery.*, *18*, 459–464.

Oberlin, C., Béal, D., Leechavengvongs, S., Salon, A., Dauge, M. C., & Sarcy, J. J. (1994). Nerve transfer to biceps muscle using a part of ulnar nerve for C5-C6 avulsion of the brachial plexus: Anatomical study and report of four cases. *The Journal of Hand Surgery*, *19*, 232–237.

Okochi, M., Ueda, K., Okochi, H., Asai, E., Sakaba, T., & Kajikawa, A. (2016). Facial reanimation using hypoglossal-facial neurorrhaphy with end-to-side coaptation between the jump interpositional nerve graft and hypoglossal nerve: Outcome and duration of preoperative paralysis. *Microsurgery*, *36*, 460–466.

Olawale A. R. Sulaiman, & Tessa Gordon. (2018). *A rat study of the use of end-to-side peripheral nerve repair as a "babysitting" technique to reduce the deleterious effect of chronic denervation*. 1.

Pannucci, C., Myckatyn, T. M., Mackinnon, S. E., & Hayashi, A. (2007). End-to-side nerve repair: Review of the literature. *Restorative Neurology and Neuroscience*, *25*, 45–63.

Pienaar, C., Swan, M. C., De Jager, W., & Solomons, M. (2004). Clinical Experience with End-to-Side Nerve Transfer. *Journal of Hand Surgery*, *29*, 438–443.

Pondaag, W., & Gilbert, A. (2008). RESULTS OF END-TO-SIDE NERVE COAPTATION IN SEVERE OBSTETRIC BRACHIAL PLEXUS LESIONS. *Neurosurgery*, *62*, 656–663.

Power, H. A., Morhart, M. J., Olson, J. L., & Chan, K. M. (2019). Postsurgical Electrical Stimulation Enhances Recovery Following Surgery for Severe Cubital Tunnel Syndrome: A Double-Blind Randomized Controlled Trial. *Neurosurgery*, *86*, 769–777.

Power, H. A., Morhart, M. J., Olson, J. L., & Chan, K. M. (2020). Postsurgical Electrical Stimulation Enhances Recovery Following Surgery for Severe Cubital Tunnel Syndrome: A Double-Blind Randomized Controlled Trial. *Neurosurgery*, *86*.

Puonti, H. K., Jääskeläinen, S. K., Hallikainen, H. K., & Partanen, T. A. (2011). A new approach to microneurovascular TRAM-flap breast reconstruction – a pilot study. *Journal of Plastic, Reconstructive & Aesthetic Surgery*, *64*, 346–352.

Puonti, H. K., Jääskeläinen, S. K., Hallikainen, H. K., & Partanen, T. A. (2017). Improved sensory recovery with a novel dual neurorrhaphy technique for breast reconstruction with free muscle sparing TRAM flap technique. *Microsurgery*, *37*, 21–28.

Rapp, E., Lallemand, S., Ehrler, S., Buch, N., & Foucher, G. (1999). Restoration of sensation over the contact surfaces of the thumb-index pinch grip using the terminal branches of the superficial branch of the radial nerve. *Annales de Chirurgie de La Main et Du Membre Supérieur*, *18*, 179–183.

Rosberg, H. E., Carlsson, K. S., Höjgård, S., Lindgren, B., Lundborg, G., & Dahlin, L. B. (2005). Injury to the human median and ulnar nerves in the forearm—Analysis of costs for treatment and rehabilitation of 69 patients in southern Sweden. *Journal of Hand Surgery (Edinburgh, Scotland)*, *30*, 35–39.

Rovak, J. M., Cederna, P. S., & Kuzon Jr, W. M. (2001). Terminolateral Neurorrhaphy: A Review of the Literature. *Journal of Reconstructive Microsurgery.*, *17*, 615–624.

Roy, J., Henry, B. M., PĘkala, P. A., Vikse, J., Saganiak, K., Walocha, J. A., & Tomaszewski, K. A. (2016). Median and ulnar nerve anastomoses in the upper limb: A meta-analysis. *Muscle & Nerve*, *54*, 36–47.

Sabatier, M. J., & English, A. W. (2015). Pathways Mediating Activity-Induced Enhancement of Recovery From Peripheral Nerve Injury. *Exercise and Sport Sciences Reviews*, *43*, 163–171.

Said, S. I., & Mutt, V. (1970). Potent peripheral and splanchnic vasodilator peptide from normal gut. *Nature*, *225*, 863–864.

Sallam, A. A., El-Deeb, M. S., & Imam, M. A. (2017). Nerve Transfer Versus Nerve Graft for Reconstruction of High Ulnar Nerve Injuries. *The Journal of Hand Surgery*, *42*, 265–273.

Schenck, T. L., Stewart, J., Lin, S., Aichler, M., Machens, H.-G., & Giunta, R. E. (2015). Anatomical and histomorphometric observations on the transfer of the anterior interosseous nerve to the deep branch of the ulnar nerve. *The Journal of Hand Surgery, European Volume*, *40*, 591–596.

Schmidt, C. E., & Leach, J. B. (2003). Neural Tissue Engineering: Strategies for Repair and Regeneration. *Annual Review of Biomedical Engineering*, *5*, 293–347.

Schreiber, J. J., Byun, D. J., Khair, M. M., Rosenblatt, L., Lee, S. K., & Wolfe, S. W. (2015). Optimal axon counts for brachial plexus nerve transfers to restore elbow flexion. *Plastic and Reconstructive Surgery*, *135*, 135e–141e.

Sforza, C., Frigerio, A., Mapelli, A., Tarabbia, F., Annoni, I., Colombo, V., … Biglioli, F. (2015). Double-powered free gracilis muscle transfer for smile reanimation: A longitudinal optoelectronic study. *Journal of Plastic, Reconstructive & Aesthetic Surgery*, *68*, 930–939.

Snyder-Warwick, A. K., Yee, A., & Mackinnon, S. E. (2017). New Insights and Applications in the Treatment of Nerve Injuries. In H. Till, M. Thomson, J. E. Foker, G. W. Holcomb III, & K. M. Khan (Eds.), *Esophageal and Gastric Disorders in Infancy and Childhood* (pp. 549–570). Berlin, Heidelberg: Springer Berlin Heidelberg.

Socolovsky, M., Martins, R. S., Di Masi, G., & Siqueira, M. (2012). Upper brachial plexus injuries: Grafts vs ulnar fascicle transfer to restore biceps muscle function. *Neurosurgery*, *71*, ons227-232.

Su, D. Y., Wan, H., Li, D. Z., Qiao, H., Schumacher, M., & Liu, S. (2020). Analysis of Preoperative Factors Influencing Hypoglossal-facial 'Side'-to-side Neurorrhaphy for Facial Paralysis after Excision of Acoustic Neuroma. *Biomedical and Environmental Sciences : BES*, *33*, 30–36.

Sunderland, S. (1951). A CLASSIFICATION OF PERIPHERAL NERVE INJURIES PRODUCING

    LOSS OF FUNCTION. *Brain*, *74*, 491–516.

Thorsén, F., Rosberg, H.-E., Steen Carlsson, K., & Dahlin, L. B. (2012). Digital nerve injuries:

    Epidemiology, results, costs, and impact on daily life. *Journal of Plastic Surgery and Hand Surgery*, *46*,

    184–190.

Tong, J., Xu, B., Dong, Z., Zhang, C., & Gu, Y. (2017a). Surgical outcome for severe cubital tunnel

    syndrome in patients aged >70 years: A mean follow-up of 4.5 years. *Acta Neurochirurgica*, *159*,

    917–923.

Tong, J., Xu, B., Dong, Z., Zhang, C., & Gu, Y. (2017b). Surgical outcome for severe cubital tunnel

    syndrome in patients aged >70 years: A mean follow-up of 4.5 years. *Acta Neurochirurgica*, *159*,

    917–923.

Uchida, Y., & Sugioka, Y. (1992). Electrodiagnosis of Martin-Gruber connection and its clinical

    importance in peripheral nerve surgery. *Journal of Hand Surgery*, *17*, 54–59.

Ueda, K., Akiyoshi, K., Suzuki, Y., Ohkouchi, M., Hirose, T., Asai, E., & Tateshita, T. (2007).

    Combination of Hypoglossal-Facial Nerve Jump Graft by End-to-Side Neurorrhaphy and Cross-

    Face Nerve Graft for the Treatment of Facial Paralysis. *J Reconstr Microsurg*, *23*, 181–187.

Vargas, M. E., & Barres, B. A. (2007). Why Is Wallerian Degeneration in the CNS So Slow? *Annual*

    Review of Neuroscience, *30*, 153–179.

Viterbo, F. (1993). A new method for treatment of facial palsy: The crossface nerve transplantation with

    end-to-side neurorrhaphy. *Rev Soc Bras Cir Plast Estet Reconstr*, *8*, 29–38.

Viterbo, Fausto, Amr, A. H., Stipp, E. J., & Reis, F. J. (2009). End-to-Side Neurorrhaphy: Past,

    Present,and Future. *Plastic and Reconstructive Surgery*, *124*, e351–e358.

Viterbo, Fausto, Brock, R. S., Maciel, F., Ayestaray, B., Garbino, J. A., & Rodrigues, C. P. (2017). End-to-side versus end-to-end neurorrhaphy at the peroneal nerve in rats. *Acta Cirurgica Brasileira*, *32*, 697–705.

Viterbo, Fausto, Romão, A., Brock, R. S., & Joethy, J. (2014). Facial Reanimation Utilizing Combined Orthodromic Temporalis Muscle Flap and End-to-Side Cross-Face Nerve Grafts. *Aesthetic Plastic Surgery*, *38*, 788–795.

Viterbo, Fausto, Trindade, J. C., Hoshino, K., & Mazzoni Neto, A. (1992). Latero-terminal neurorrhaphy without removal of the epineural sheath. Experimental study in rats. *Revista Paulista de Medicina*, *110*, 267–275.

Voche, P., & Ouattara, D. (2005). End-to-side neurorrhaphy for defects of palmar sensory digital nerves. *British Journal of Plastic Surgery*, *58*, 239–244.

Wagstaff, L. J., Gomez-Sanchez, J. A., Fazal, S. V., Otto, G. W., Kilpatrick, A. M., Michael, K., … Jessen, K. R. (2021). Failures of nerve regeneration caused by aging or chronic denervation are rescued by restoring Schwann cell c-Jun. *ELife*, *10*, e62232.

Waller, A. V., & Owen, R. (1850). XX. Experiments on the section of the glossopharyngeal and hypoglossal nerves of the frog, and observations of the alterations produced thereby in the structure of their primitive fibres. *Philosophical Transactions of the Royal Society of London*, *140*, 423–429.

Wang, S., Li, P., Xue, Y., Yiu, H., Li, Y.-C., & Wang, H. (2013). Contralateral C7 nerve transfer with direct coaptation to restore lower trunk function after traumatic brachial plexus avulsion. *The Journal of Bone and Joint Surgery. American Volume*, *95*, 821–827, S1-2.

Watson, B. V., & Doherty, T. J. (2010). Frequency-dependent conduction block in ulnar neuropathy localized to the elbow. *Clinical Neurophysiology : Official Journal of the International Federation of Clinical Neurophysiology*, *121*, 2111–2116.

Wu, S., Curran, M. W. T., Hachisuka, A., Rajshekar, M., & Chan, K. M. (2022). A new method to quantify innervation of the ulnar intrinsic hand muscles by the anterior interosseous nerve in Martin-Gruber anastomosis and nerve transfer surgery. *Muscle & Nerve*. https://doi.org/10.1002/mus.27654

Xie, Q., Shao, X., Song, X., Wang, F., Zhang, X., Wang, L., … Lyu, L. (2021). Ulnar nerve decompression and transposition with versus without supercharged end-to-side motor nerve transfer for advanced cubital tunnel syndrome: A randomized comparison study. *Journal of Neurosurgery*, 1–11.

Yang, L. J.-S., Chang, K. W.-C., & Chung, K. C. (2012). A systematic review of nerve transfer and nerve repair for the treatment of adult upper brachial plexus injury. *Neurosurgery*, *71*, 417–429; discussion 429.

You, S., Petrov, T., Chung, P. H., & Gordon, T. (1997). The expression of the low affinity nerve growth factor receptor in long-term denervated Schwann cells. *Glia*, *20*, 87–100.

Zarina S. Ali, Gregory G. Heuer, Ryan W. F. Faught, Shriya H. Kaneriya, Umar A. Sheikh, Idrees S. Syed, … Eric L. Zager. (2015). *Upper brachial plexus injury in adults: Comparative effectiveness of different repair techniques %J Journal of Neurosurgery JNS*. *122*, 195.

Zochodne, D. W. (2008). *Neurobiology of Peripheral Nerve Regeneration*. Cambridge: Cambridge University Press. Cambridge Core.

Zochodne, D. W. (2012). The challenges and beauty of peripheral nerve regrowth. *Journal of the Peripheral Nervous System : JPNS*, *17*, 1–18.