Posterior Tibial Nerve Stimulation: A Study of the Effects of Stimulation Parameters on Urodynamic Changes in Anesthetized Rats

by

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Abstract

Although posterior tibial nerve stimulation (PTNS) has been shown in both clinical and animal studies to mediate inhibition of bladder function, our understanding of the role of posterior tibial nerve (PTN) afferents that elicit this reflex is significantly limited. To this end, we investigated the effects of frequency-dependent PTNS in urethane-anesthetized rats undergoing repeated urodynamic fills. Electrical pulses applied at both 5 Hz and 10 Hz resulted in significant decreases in the frequency of bladder contractions during and after 10-minutes of stimulation (acute inhibition and post-stimulation effect). Selective electrical activation of the lateral or medial plantar nerves confirmed that each type of bladder reflex can be elicited by individual subsets of PTN afferents. Our findings indicate that PTNS-evoked bladder reflexes are tuned to specific stimulation frequencies. The neuromodulation of bladder function achieved by selective PTN branch stimulation provides a novel insight into these neural pathways and also suggests potential clinical application.
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List of Abbreviations

OAB, overactive bladder
PTNS, posterior tibial nerve stimulation
PTN, posterior tibial nerve
BRC, bladder rhythmic contractions
NCE, nerve cuff electrode
EMG, electromyography
EUS, external urethral sphincter
fEMG, foot EMG
AUEC, area under the EMG curve
NRC, nerve recruitment curve
AICI, average inter-contraction interval
aBCF, average bladder contraction frequency
SNS, sacral nerve stimulation
Chapter 1
Introduction & Background

1.1 Motivating Problem: Overactive Bladder (OAB)

Overactive bladder (OAB) is a disorder where the sufferer feels a sudden need to urinate despite the bladder not being completely full. The disorder is characterized by having to urinate eight or more times in 24 hours or at least twice a night. OAB may or may not be accompanied by urinary leakage episodes. One can imagine the debilitating effect that this frequent urination and inability to hold back urine may have on a person’s social life, confidence and sleeping habits. Presently, it is estimated that one in five or over 6 million Canadians over the age of 35 suffer from OAB (Corcos & Schick, 2004), where the overall economic impact of treating these patients is around $380 million in Canada alone and $3.9 billion worldwide (Irwin et al., 2009). Although OAB is a disorder that may be left undiagnosed and/or untreated without further effect on patient mortality, the profound impact on the quality of life of the individual patient is often the main motivating factor for seeking medical attention.

1.2 Organization of the Thesis

This thesis is comprised of five chapters. Chapter One (Introduction & Background) provides a brief overview of the treatment options for OAB followed by a broad literature review dealing with the history of posterior tibial nerve stimulation (PTNS) — the therapy which is at the centerfold of this thesis. The chapter concludes with a summary of the primary objectives of the research, which look to address specific gaps in the literature outlined in Section 1.5.2.

Chapter Two (Theory & Methods) provides a more specific literature review on the research objectives—or the areas where the gaps in the scientific literature were shown to exist. While the literature review in Chapter One served to give the reader a broad overview of PTNS, the brief overview of PTNS in Chapter Two strives to frame a rationale for the study within the context of prior work in this area and current state of PTNS animal models specifically. The second part of the chapter (Materials and Methods) outlines the experimental approach and analytical methods used to conduct the experiments, quantify the data and determine statistical significance.
Chapter Three (Results) provides a summary of the findings from the experiments conducted, referencing the embedded Figures.

Chapter Four (Discussion) frames the significance of these results within the context of current scientific literature—both the literature discussed in Chapters One & Two, as well as other relevant papers that are introduced in Chapter Five. This chapter concludes by providing a summary of the clinical implications of the results of this study, thereby connecting the significance of the work back to the thesis’ motivating problem: improving the lives of patients with OAB.

The final chapter, Chapter Five (Conclusions & Future Work), explores the future direction of this research and suggests potential studies aimed at further developing and optimizing PTNS therapy.

1.3 Overview of Treatment Options for Overactive Bladder

Once medical attention is sought and OAB is diagnosed, the typical treatment options available for patients are numerous. Behavioural training, pharmaceutical medication, bladder augmentation surgery, Botox and electrical neuromodulation are the main avenues for providing symptom relief. The two primary modalities of electrical neuromodulation are sacral nerve stimulation (SNS) and posterior tibial nerve stimulation (PTNS) – the topic of the present work.

1.3.1 Behavioural Training

Behavioural training involves exercises that attempt to strengthen the pelvic floor muscles. It is of course the least invasive method. However, not only is it relatively ineffective (Fantl et al., 1991), this treatment option is also very tedious and time-consuming. For these reasons, this treatment option has not been adopted for widespread use.

1.3.2 Pharmaceutical Medication

Pharmaceutical medication is typically the frontline treatment for OAB. Anti-cholinergic medication such as Oxybutynin or Tolterodine blocks the nerve signals to the bladder that initiate
bladder contractions. Clinical studies have shown these drugs to be effective in 50-70% of patients (Van Kerrebroeck et al., 2001). However, medication is associated with side-effects such as nausea, dry-mouth or increased heart-rate and hence is not tolerated well by some patients. In fact, many patients decide to discontinue use of medication as a result of these side-effects; choosing to tolerate the symptoms of OAB.

1.3.3 Surgical Interventions

For patients with severe OAB, doctors may recommend bladder augmentation surgery. In a typical surgery, the surgeon uses part of the intestinal smooth muscle to increase the size of the bladder. Although bladder capacity has been increased there is now a smaller percentage of the bladder with working mechanoreceptors that can respond to bladder fullness (Biers, Venn, & Greenwell, 2012). Surgery is obviously very invasive and is typically seen as a last line treatment for only the most severe patients. Furthermore, with the development of newer, safer and less invasive therapies, it appears as though the surgical route is being offered less often (Reyblat & Ginsberg, 2010).

1.3.4 Botox

Another treatment option involves injections of botulinum toxin (Botox) into the bladder musculature to relax the tissue and provide OAB symptom relief. Performed as an outpatient procedure under general or local anesthesia, studies have shown the effectiveness of Botox as a treatment for OAB (Dmochowski et al., 2010; Karsenty et al., 2008). In addition to neuromodulation and pharmaceutical medication, Botox is considered to be a frontline treatment option for OAB symptoms. However, there are inherent risks of infection, possible complications resulting from the surgical procedure and allergic reactions caused by receiving Botox treatment. Another side-effect caused by intravesicular injection of Botox involves the patient being unable to completely empty their bladder (i.e. urinary retention). This requires transurethral catheterization until sufficient reflex bladder activity returns. Nonetheless, five days following successful Botox injections, OAB symptom relief is noticed with peak symptom relief achieved at the two week mark. Patients can expect the therapy effects to last 6-9 months before another procedure is required.
1.3.5 Electrical Neuromodulation

Electrical neuromodulation—the topic of the present work—has been studied extensively in treating OAB. In addition to sacral nerve and posterior tibial nerve stimulation, the other neural targets that could potentially be used for treating OAB include the dorsal penile nerve (Wheeler, Walter, & Zaszczurynski, 1992), the clitoral nerve (Dalmose et al., 2003), the sacral nerve root (Martens & Heesakkers, 2011) and direct bladder stimulation (Geirsson, Fall, & Sullivan, 1995). The reason that many of these treatment modalities have not been clinically implemented is because of their invasiveness, cost, side-effects and therapeutic effectiveness.

In contrast, sacral nerve stimulation (SNS) has been shown to be particularly effective in treating overactive bladder (Peeren, Hoebeke, & Everaert, 2005). This stimulation technique involves an electrical stimulator that drives electrical pulses through a multi-contact lead-type electrode positioned at the third sacral root (S3) of the spinal cord. The procedure involves a two-stage surgery that results in a permanently implanted neurostimulation system. However, in addition to various stimulation-evoked side effects, many patients are either unwilling to undergo this procedure while others cannot afford the high cost of this treatment (Kantartzis & Shepherd, 2013).
1.4 A Comparison between Percutaneous and Transcutaneous PTNS

PTNS therapy can be achieved in one of two ways. The primary method (PPTNS) involves the use of a percutaneous needle electrode paired to a surface (return) electrode. It was adopted from acupuncture, and clinically implemented as the Stoller Method in the late 1990s. The second method (TPTNS), which has not been used widely in a clinical setting, utilizes a pair of surface electrodes adhered to the skin surface. As such, there is no penetration of the skin surface. The following pages will outline the primary differences between the two methods.

1.4.1 Percutaneous Posterior Tibial Nerve Stimulation (PPTNS)

Stimulating the nerve percutaneously is the more common and invasive method. Effective nerve stimulation involves a needle electrode that punctures the skin, the tip of which is carefully positioned in close proximity to the posterior tibial nerve. PTNS follows the typical protocol of stimulation sessions conducted once a week for twelve weeks. Each session is 30 minutes in duration and electrical pulses are applied at a frequency of 20Hz. Stimulation is set at the highest bearable amplitude that can exceed the foot twitch threshold (up to 10 mA). The most common pulse duration is 200 microseconds. This approach was first introduced by McGuire et al in his seminal paper (McGuire, Zhang, Horwinski, & Lytton, 1983) and has since been shown to be effective in 60-80% of patients (MacDiarmid et al., 2010; K. M. Peters, Carrico, MacDiarmid, et al., 2013). Since 1983, PPTNS has been shown in controlled clinical trials to be at least as, if not more effective in treating OAB symptoms than the available pharmaceutical medications (K. M. Peters et al., 2009). The limited number of animal studies that have investigated PTNS therapy have typically involved direct activation of the PTN, which is most similar to PPTNS (Tai, Chen, Shen, Wang, Liu, et al., 2011; Tai, Chen, Shen, Wang, Roppolo, et al., 2011).

However, PPTNS requires a trained medical professional to deliver the therapy. Following the initial 12-week treatment period, the patient is required to maintain the therapeutic effects of PTNS by repeated visits to the hospital (~every 3 weeks). The need for repeated clinical visits to maintain PTNS therapy results can incur significant long-term costs and inconvenience for
patients. Consequently, this may limit the widespread adoption of PTNS as a frontline treatment for OAB patients.

1.4.2 Transcutaneous Posterior Tibial Nerve Stimulation (TPTNS)

Transcutaneous stimulation, on the other hand, requires only a pair of surface electrodes that are adhered to the skin above and/or below the ankle. The stimulation protocol is the same as PPTNS, in that it follows the Stoller Method. Although this approach has received very little attention in the literature, more and more clinical trials are beginning to show that TPTNS can be an effective treatment option for OAB (Ammi et al., 2014; Souto, Reis, Palma, Palma, & Denardi, 2014; Tellenbach, Schneider, Mordasini, Thalmann, & Kessler, 2013). With respect to pre-clinical work, a comprehensive literature search revealed only a single study involving TPTNS. Tai et al showed that transcutaneous stimulation of the PTN in cats provided bladder inhibition while forefoot stimulation showed no such effects (Tai, Shen, Chen, Wang, Liu, et al., 2011).

Since surface electrodes are being used to activate the PTN, there is obviously a lack of specificity in the stimulation compared to PPTNS. One can expect the skin to be a major barrier in the movement of current between the surface electrode and the nerve. This limitation does not affect PPTNS. Thus, one can expect to see less nerve activation during TPTNS when compared to PPTNS. With this in mind, TPTNS does show promising results as a non-invasive alternative to PPTNS. Currently however, no standard treatment protocol or stimulation device exists for TPTNS.

The primary advantage of TPTNS therapy is that treatment of OAB can potentially be achieved without the presence of a medical professional or repeated injections of needle electrodes. This offers the additional advantage of conveniently treating oneself at home or at work. This can minimize the number of hospital visits and the overall long-term costs associated with treating OAB. However, due to the high electrical impedance of skin tissue, surface electrodes require comparatively larger stimulation amplitudes for electrically activating the posterior tibial nerve.
As a result, cutaneous afferents and other non-targeted nerve fibers are co-activated, which in turn can impose a limit on stimulation parameters.
1.5 Posterior Tibial Nerve Stimulation Therapy

The following sections provide a detailed literature review of PTNS therapy and the related nerve stimulation technology.

Posterior tibial nerve stimulation (PTNS) was introduced in the late 1990s as an alternative to conventional treatment options. In general, there are two methods of electrically activating the posterior tibial nerve: (1) percutaneous posterior tibial nerve stimulation (PPTNS) or, (2) transcutaneous tibial nerve stimulation (TPTNS). A comparison of these two methods has already been provided in Section 1.4. As a brief summary, the former method utilizes a needle electrode while the latter achieves electrical activation via a surface electrode.

Since the posterior tibial nerve (PTN) is a relatively superficial nerve located in the ankle, treatment is minimally invasive and thus relatively inexpensive compared to sacral neuromodulation (Chen, Bercik, Werner, & Thung, 2012). Additionally, PTNS therapy has been shown to elicit little to no side-effects (Svihra, Kurca, Luptak, & Kliment, 2002). Over a three-year period, PTNS has been confirmed to be effective for reducing OAB symptoms in patients who are refractory to pharmaceutical medication (K. M. Peters, Carrico, Wooldridge, Miller, & MacDiarmid, 2013). It is important to note however, as will be discussed in the ensuing chapters, that there is very little consensus on the inner workings of PTNS therapy.

1.5.1 The Anatomy of the Posterior Tibial Nerve

The posterior tibial nerve originates from the tibial branch of the sciatic nerve. The sciatic nerve is the broadest nerve in the human body, having a 2 cm diameter at its origin. As the sciatic nerve descends behind the thigh muscles proximal to the knee, it splits into the tibial nerve and the common peroneal nerve. The sciatic nerve receives its spinal contributions from L4-5 as well as S1-S3. The tibial branch is composed of all of these contributions while the common peroneal nerve has all but the S3 contribution.

The tibial nerve is the larger of the two branches. It descends along the back of the thigh and knee pit to the distal border of the popliteus muscle where it is overlapped by the two heads of
the gastrocnemius (calf muscle). As it begins its descent down the leg it is buried deep in the soleus and gastrocnemius muscles but by the last portion of the leg it is covered only by skin. It then continues vertically down the back of the leg to a point between the medial malleolus (the bony prominence on the medial side of the ankle) and the heel. Throughout its descent from the thigh into the ankle and foot, the tibial nerve forms the (a) articular and (b) muscular branches as well as (c) the sural branch. At the Achilles tendon, it branches off into the (d) common calcaneal nerve. As it travels further under the laciniate ligament (flexor retinaculum) in the foot, it divides into the (e) medial plantar nerve and the (f) lateral plantar nerve. For the purposes of this thesis, in addition to the main trunk of the nerve (at the level of the medial malleolus), we are most concerned with these two distal plantar branches of the PTN.

The **medial plantar nerve** is the largest terminal division of the tibial nerve arising under the laciniate ligament. It supplies the skin of the sole of the foot and medial side of the hallux, while also innervating the second and third toes as well as the medial side of the fourth toe.

The **lateral plantar nerve** innervates the skin of the fifth toe and lateral half of the fourth toe as well as the various muscles in the foot.

**1.5.2 A History of Posterior Tibial Nerve Stimulation**

In order to appreciate the rationale of this present study, it is important to understand the origins of PTNS therapy, as well as the successes and failures of previous clinical work. Additionally, in order to give the reader a greater breadth of knowledge in the field, it is important to contextualize our current understanding of PTNS within the plethora of scientific literature on the topic. A brief history of PTNS is given in the following pages outlining the most significant scientific milestones since its inception by EJ McGuire in 1983 (McGuire et al., 1983).

**1.5.2.1 Acupunctural Origins of PTNS**

Acupuncture, which originated in ancient China, has since been used for over 2000 years as a means for treating the symptoms of the overactive bladder (OAB). The spleen meridian 6 (SP6) acupuncture point, located three cun (i.e. finger breadths) superior to the middle of malleolus, has been labeled as the main stimulation point for reducing symptoms. OAB symptoms include
urinary frequency, urgency and nocturia regardless of the presence of urinary incontinence. Nonetheless, despite its deep ancient roots, this treatment approach has remained outside the realm of modern scientific medicine until McGuire’s seminal work in 1983.

### 1.5.2.2 The Birth of Posterior Tibial Nerve Stimulation: 1983

McGuire, inspired by traditional Chinese acupuncture, applied transcutaneous stimulation to the posterior tibial and common peroneal nerves of the ankle (Sp 6 location in Chinese acupuncture) in his initial experimental studies on non-human primates. The Sp 6 location is on the medial side of the ankle proximal to the medial malleolus. Managing to successfully inhibit detrusor muscle contractility in these first experiments, he conducted a subsequent clinical trial that achieved similar results (McGuire et al., 1983). Out of his initial twenty-two patients, 12 (55%) became completely dry while 8 (36%) showed improvements from baseline. This groundbreaking work laid the foundation for the developments in PTNS therapy that were to follow over the next three decades.

### 1.5.2.3 Foundations for PTNS: 1987

Five years later, Stoller observed in pig-tailed monkeys that acupuncture treatment on the SP 6 location (Stoller, Copeland, Millard, & Murnaghan, 1987) resulted in significant urodynamic changes. He discovered that acupuncture treatment eliminated inappropriate bladder contractions while maintaining the normal voiding sequence in monkeys with a history of bladder instability. In addition, he also noted that: (1) additional acupuncture sessions increased the duration of the interval of relief in between inappropriate contractions and; (2) acupuncture applied to anaesthetized limbs provided no such relief, suggesting that an intact periphery nervous system is necessary for results.

### 1.5.2.4 First Clinical Studies of PTNS Therapy: 1993

A study by Chang et al which investigated the long-term effects of acupuncture in women with OAB, showed that 6 of 8 patients achieved a stable detrusor muscle after acupuncture. The one year and three year follow-up showed that the effect was temporary and that follow up
treatments were required to be maintain improvements in bladder function (P. L. Chang, Wu, & Huang, 1993).

PTNS was also investigated in female patients with interstitial cystitis. (Geirsson, Wang, Lindstrom, & Fall, 1993). Overall, stimulation of the PTN was ineffective for this particular bladder disorder, where none of the six TPTNS patients and only one of six PPTNS patients achieved either subjective or objective therapeutic end-points.

Walter et al published a paper at the end of 1993 that provided interesting insight into the mechanism of PTNS therapy. (Walter, Wheeler, Robinson, & Wurster, 1993). The group attempted to acutely inhibit the cat bladder in spinal cord injured cats (n=5) by subcutaneously stimulating the tibial nerve. They found stimulation only caused leg spasms, and actually increased bladder activity. These results suggested that the effects of PTNS likely involve supraspinal circuits.

1.5.2.5 Limited Efficacy of TPTNS: 1996

Further attempts at transcutaneous stimulation of the posterior tibial nerve became infrequent. One group, which saw urodynamic improvements in only 2 out of 36 patients undergoing a 3-week stimulation protocol, described their experience with the use of the tibial nerve as “discouraging” (Hasan, Robson, Pridie, & Neal, 1996).

1.5.2.6 A Underlying Mechanism for PTNS: 1998

In 1998, Chang et al described a possible mechanism for the early success observed with PTNS therapy (C. J. Chang et al., 1998). They looked at a group of cats whose bladder had been noxiously stimulated by acetic acid. The researchers found that the expression of c-Fos, an indirect marker of neuronal activity, remained high in the spinal cords of all the cats except those that had received electro-acupuncture (EA) at the SP 6 location one hour prior to the bladder irritation. Their main conclusion was that this inhibition of C-fibre activity by EA may provide the underlying mechanism for neuromodulation through the posterior tibial nerve.
1.5.2.7 The Birth of PTNS and Stoller Afferent Nerve Stimulation: 1999

Over a decade after his initial work with pig-tailed monkeys, Stoller presented an abstract from his clinical trial on ninety patients treated for symptoms of overactive bladder (Stoller, 1999). This was quickly followed by his patent on the technique of PPTNS which is now known as Stoller Afferent Nerve Stimulation (SANS) technique (Malaney, Morris, Stoller, & Gleason, 1999). The method involved inserting a needle 5 cm cephalad to the medial malleolus, posterior to the margin of the tibia. The needle was inserted up until the medial edge of the fibula. Patients were stimulated with an adjustable current (0-9mA, highest bearable), once weekly for 20-30 minute sessions for 10 weeks. Stoller achieved an initial 81% objective success rate (defined by 50% decrease in symptoms); going even further to show that maintenance therapy at increased treatment intervals can maintain the results. This trial single-handedly sparked a wave of new studies involving PPTNS via the Stoller Method that has since been used for PTNS therapy in OAB patients.

1.5.2.8 The First Verification of SANS: 2000

Klinger et al. was the first to verify the SANS method by testing a 3-week, 12 session PTNS paradigm in 15 patients diagnosed with urgency-frequency syndrome stemming from OAB (Klingler, Pycha, Schmidbauer, & Marberger, 2000). The highlights of the study were a complete response achieved by 7 out of 15 (47%) patients, and a partial response achieved by 3 out of 15 (20%) patients. A complete response was considered a patient whose daytime voids dropped below 8 and night time voids below 2. There was also an increase in total bladder capacity after 12 weeks. The study concluded with an advisory against using the method on patients with a history of interstitial cystitis (IC), which corroborated findings by Gerrison et al 1993.

A year later, Govier et al conducted the first multi-centre trial with 53 patients enrolled in 5 sites across the United States (Govier, Litwiller, Nitti, Kreder, & Rosenblatt, 2001). The trial used the SANS method once weekly over the course of 12 weeks. The study’s primary efficacy end point was a 25% reduction in daytime voids in patients with 10 voids per day or more. At the end of the study, the investigators found 71% of the patients as treatment successes. The authors also
observed 25% and 21% mean reductions in daytime voids and night time voids, respectively. In addition, a 35% reduction in urge incontinence episodes was also observed.

Later that year, Van Balken et al published the results from a second prospective multi-centre trial; this time in Europe (M. R. van BALKEN et al., 2001). Using a 9 V, 20Hz stimulator, the amplitude was adjusted beyond the threshold for toe flexion, up to a tolerable level of 9 mA. Again, treatment was conducted for 30 minutes over the course of 12 weeks. This stimulation protocol was to become the gold standard for PTNS. Clinical efficacy was based on whether or not the patient requested to continue treatment after 12 weeks. In this study, 60% of all patients were deemed to have achieved a positive response and continued with long-term treatment.

These three initial trials illustrate the difficulty in comparing treatment success in the early days of PTNS. While van Balken et al used a subjective measure as their primary efficacy end point; both Klinger et al and Govier’s group chose to focus on the objective results. Even between Klinger and Govier, different objective efficacy end points were selected.

1.5.2.9 Further European Success: 2002

A Slovak group mimicked Stoller’s stimulation protocol in 28 female patients, who were divided into (1) SANS treatment, (2) drugs, and (3) no treatment groups (Svihra et al., 2002). The researchers observed increases in quality of life measures (as judged by the Incontinence Quality of Life Questionnaire) by a margin of 50% or greater in 60% of the SANS patients. The drugs however, fared better, achieving an 80% success rate.

PPTNS was first tested as a treatment modality for children by Hoebeke et al in Belgium (Hoebeke, Renson, Petillon, Vande Walle, & De Paepe, 2002). Thirty-two children underwent PTNS treatment for 6 weeks, and continued treatment for another 6 weeks, if there were improvements in symptoms. The study is highlighted by the following results: (1) a 25% disappearance and 36% improvement in urgency; (2) 16% incontinence cure rate and 52% improvement rate; (3) mean bladder capacity increased from an average of 185.16 mL to an
average of 279.19mL. However, the study was uncontrolled, since it permitted patients to use anti-cholinergic drugs throughout the 12 weeks.

1.5.2.10 The Vandoninck Trials: Early 2003

Vandoninck et al treated urge incontinence in 35 Dutch and Italian patients across 5 sites (V. Vandoninck, M. R. Van Balken, E. Finazzi Agro, F. Petta, C. Caltagirone, et al., 2003). They achieved 70% objective success rate (decrease in leakage episodes by 50% or more) and a 63% response rate (request to continue treatment). Additionally, 46% of the patients were completely cured, seeing no leakage after 12 weeks of PTNS.

The Vandoninck group followed up their initial work with a second trial that involved 39 patients (V. Vandoninck, M. R. Van Balken, E. Finazzi Agro, F. Petta, F. Micali, et al., 2003). This was a multi-centre European trial consisting of 12 weeks of treatment. At the end of this treatment period, twenty-three (59%) patients chose to seek long-term treatment and were considered subjective successes; while 16 (41%) patients were deemed objective treatment successes having seen their 24hr total catheterized volume reduced by 50% or more.

Vandoninck et al published their third paper in succession later that year, which described results from another study involving ninety consecutive patients (Vera Vandoninck et al., 2003). Following the same protocol as in their previous studies, urinary leakage episodes per day were decreased by 50% in 56% of the patients while 64% of patients requested to continue treatment, indicating subjective success. They noted that PTNS was not effective in treating detrusor instability. In both of Vandoninck’s 2003 trials, quality of life improved significantly.

1.5.2.11 Acute Effects of Transcutaneous PTNS: Late 2003

Late 2003 was highlighted by the publishing of two journal articles that continued work from nearly a decade prior (Geirsson et al., 1993; Hasan et al., 1996). Both articles re-visited transcutaneous posterior tibial nerve stimulation (TPTNS) as a means of achieving acute inhibition of the urinary bladder.
The first paper was a trial conducted by Amarenco et al addressing whether or not TPTNS can achieve acute changes in bladder function in patients with OAB (Amarenco et al., 2003). The mean first voluntary detrusor contraction (1st IVDC) during standard cystometry increased from 163 ml before stimulation to 232 ml during 10 Hz stimulation. In similar fashion, the average maximum cystometric capacity (MCC) increased; from 221 ml to 277 ml. The study showed that 22 of the 44 patients (50%) were positive responders to the treatment, increases in either the 1st IVDC or MCC by 100 ml or 50% during stimulation, respectively.

The second paper was a case study of a 64-year old man with detrusor hyperreflexia (Andrews & Reynard, 2003). The subject showed increased bladder capacity (75-145 ml) in three bladder fills during TPTNS at 25 Hz. A follow-up treatment three weeks later showed similar results.

These two studies showed that, in contrast to previous work (Geirsson et al., 1993; Hasan et al., 1996), TPTNS has the potential to achieve clinically relevant effects such as temporary increases in bladder capacity.

1.5.2.12 The Spanish, Italian and Chinese Trials: 2004

A study conducted in Spain followed a 10 week, 30 minutes per session PPTNS protocol (Congregado Ruiz, Pena Outeirino, Campoy Martínez, Leon Duenas, & Leal López, 2004). Fifty-one patients were recruited, 26 of whom had frequency/urgency, 22 had urge incontinence and 3 with IC. The results showed statistically significant improvements in both daytime and night time voiding frequency and volumes, as well as reductions in leakage episodes and hypogastric pain. The trial called for further long-term randomized studies to confirm these findings.

An Italian study published in the same year showed similar results (De Gennaro et al., 2004). The trial was the second PPTNS trial on children. It followed the standard 12 week, 30 minute session protocol conducted in a total of 23 children varying from 4 to 17 years old. The main highlight of the study was the 80% success rate in the treatment of overactive bladder symptoms in 10 of the patients. Five of 9 patients were cured from incontinence. They also noted that in terms of pain, PPTNS is well-tolerated by children.
The last relevant study published in 2004 served as a follow-up to previous work on treating IC with PTNS (Zhao & Nordling, 2004). Unlike Geirsson et al, who in 1993 used TPTNS (Geirsson et al., 1993), Zhao et al followed the standard PPTNS protocol. Their conclusions, similar to Geirsson’s, were that over 10 weeks of treatment, PPTNS does not achieve statistically significant clinical effects on patients with refractory IC (although one patient was cured, 7% success rate).

1.5.2.13 PTNS Proven Effective, Now What: 2005

With a growing body of evidence to support the clinical efficacy of PPTNS, new studies aimed at optimizing the clinical use of PTNS therapy began to emerge. The following two studies exemplify this shift in attitude towards PTNS therapy.

The first is an interesting study by Karademir et al, which tried to determine the effects, if any, that anti-cholinergic drugs (oxybutynin hydrochloride) contributed to PPTNS treatment (Karademir et al., 2005). A total of 43 patients were divided randomly into two groups: group 1 received SANS treatment alone, while group 2 received SANS treatment in addition to 5 mg oxybutynin hydrochloride. Treatment sessions were 1hr long (20 Hz) and were conducted weekly for 8 weeks. Quality of life questionnaires, voiding diaries and urodynamic studies were used to evaluate the therapeutic response. Groups 1 and 2 achieved a 62% and 83% response rate, respectively, improvements in symptoms by >35% obtained by both groups. Although, the mean number of frequency and urgency symptoms decreased by 37% (Group 1) and 46% (Group 2), there was no statistically significant difference between the two groups. Ultimately, this Turkish study concluded that while both are effectively when used independently, anti-cholinergic drugs used in combination with PPTNS can increase the treatment response rate (although the response strength within responders will likely remain similar).

During this time, van der Pal et al attempted to determine whether PPTNS effects are permanent or if chronic treatment must be continued after the initial success (van der Pal, van Balken, Heesakkers, Debruyne, & Bemelmans, 2006). The trial enrolled 11 patients who were previously diagnosed with either OAB (seven or more voids per day), or urge incontinence (three or more
daily urge incontinence episodes). All the patients were currently on maintenance PTNS therapy. The trial looked to assess what effects a 6-week pause in treatment would have in symptoms. After suspending PTNS for a month and a half, nine (82%) of the eleven patients had symptoms deteriorate by 50% or more. However, with resumption in treatment, symptoms disappeared once again in all patients. The study concluded that maintenance PTNS is necessary for sustaining the improvements in bladder symptoms.

1.5.2.14 Continued Treatment Optimization: 2006

Another clinical trial published in the following year investigated the effects of increasing the stimulation protocol to three PTNS sessions per week. (Finazzi Agro et al., 2005). Thirty-five subjects were divided equally into two treatment groups. Eleven (63%) of the seventeen once-weekly treated patients exhibited a 50% or more reduction in bladder symptoms, while twelve (67%) of the eighteen thrice-weekly patients were deemed treatment successes. The author’s conclusion was that “the periodicity of PTNS sessions does not affect the end-results of PTNS treatment. Instead, the advantage of more frequent stimulation sessions is to achieve earlier clinical improvement”.

Nuhoglu et al provided a second study which looked at the maintenance properties of PPTNS (Nuhoğlu, Fidan, Ayyıldız, Ersoy, & Germiyanoğlu, 2006). This time, as opposed to a 6 week pause in treatment, patients who successfully responded to treatment were followed-up a full year after PTNS therapy was ceased. Of the original 35 patients treated for OAB (once weekly for 10 weeks), 19 (54%) were deemed successful (8 or less voids per 24hr, 1 or less episodes of urgency and the disappearance of incontinence). After a year, only eight (42%) of these 19 responders were able to maintain their initial results. The results of this study agreed with van der Pal (van der Pal, van Balken, et al., 2006) in concluding that PPTNS does work, but maintenance is required for long-term symptom relief.

In an attempt to improve patient selection for PTNS therapy, van Balken et al conducted a study to see if there are any patient factors that exist that can predetermine which of the patients will respond to PPTNS treatment (M. Van Balken, Vergunst, & Bemelmans, 2006). In one of the
largest clinical trials to date, 132 patients across 8 sites in Europe were treated with PPTNS. Of these patients, 83 were treated for OAB symptoms. The results showed that 37.3% of patients achieved a 50% or greater decrease in bladder symptoms, whereas 55.4% expressed a desire to continue treatment. Unfortunately, the study was not able to determine any prognostic factor. The only criterion of importance was a low SF Mental Component Summary score at baseline, which was a predictor for negative response to PPTNS.

1.5.2.15 Acute Effects of Percutaneous PTNS: 2007/2008

Despite the 2003 work of Amarenco et al confirming the acute effects of transcutaneous simulation in treating OAB symptoms (Amarenco et al., 2003), a second study by Fjorback et al four years later showed conflicting results (Fjorback et al., 2007). Eight patients with multiple sclerosis (MS) and neurogenic detrusor overactivity underwent two consecutive cystometries; one cystometry acting as a control and the second during PPTNS. Three of the patients were given an additional cystometry while undergoing TPTNS. Surprisingly, the study showed that detrusor contractions were not inhibited by electrical stimulation in any of the patients.

A follow-up study by Kabay et al almost a year later, successfully reproduced the earlier results of Amarenco (Kabay, Yucel, & Kabay, 2008). The study looked at the urodynamic results both before and during PPTNS (20Hz, Stoller Method) in 29 patients with multiple sclerosis and resultant neurogenic detrusor overactivity (Kabay, Apr 2008). The study showed that the mean 1st IVDC on standard cystometry was increased from 138 mL at baseline to 230 mL during PPTNS. Similarly mean maximum cystometric capacity (MCC) increased from 194 mL to 286mL. These results supported the effectiveness of PPTNS in the acute treatment of an overactive detrusor.

Several months later, Kabay et al followed up with a second study that was conducted in patients with Parkinson’s Disease (PD) (Kabay, Kabay, Yucel, & Ozden, 2009). The protocol involved PPTNS (20Hz) in 22 patients undergoing standard cystometry. The mean 1st IVDC increased from 145 mL to 245 mL during PPTNS, and the mean MCC increased from 205 mL to 301 mL.
during PPTNS. The authors concluded that PPTNS is acutely effective in suppressing an overactive detrusor.

In the midst of all this work on the acute effects of PPTNS, in June of 2008, Zhao et al published results from a clinical trial that looked at the effectiveness on PPTNS in 18 female Chinese patients with IC (Zhao, Bai, Zhou, Qi, & Du, 2008). Subjects underwent 30 min sessions twice a week (as opposed to the standard once a week protocol) for 5 weeks. The main highlight of this study was the rate of subjective success, which was determined by the patients (choice of no effect, some effect or significant effect). None of the patients indicated that the treatment had a significant effect. Ten patients described some effect, and the remaining eight stated that PPTNS yielded no effect. These results were in line with Zhao’s 2004 work on PPTNS (Zhao & Nordling, 2004), and Geirsson’s 1993 pioneering paper on TPTNS (Geirsson et al., 1993); both describing the ineffectiveness of the treatment for IC patients.

1.5.2.16 Placebo Controlled Trials of PTNS: Early 2009

At this point in the history of PTNS, the effectiveness of the treatment was clearly demonstrated by numerous experts in the field. However, the lack of any placebo-controlled studies undermined the clinical validity of PTNS therapy.

The first published placebo controlled trial was conducted in Brazil (Bellette et al., 2009). The treatment included eight sessions over 4 weeks in women presenting with OAB symptoms. The authors showed that all 21 TPTNS treated patients (Amarenco method) and all 16 sham TPTNS patients resulted in an increase in quality of life and decrease in frequency and urgency episodes. Although they were using an unproven sham method, the TPTNS group showed greater improvements than sham patients in both respects.

Later in the year, Peters et al published an effective PTNS sham method for use in controlled clinical trials (K. Peters, Carrico, & Burks, 2009). The study involved 30 patients (15 men, 15 women) divided in two groups. One group received sham PTNS on the right foot sham needle at PTNS site, with TENS pad on ipsilateral foot doing the actual sham stimulation) and actual PTNS on the left foot, while the PTNS configuration (sham vs. actual) was reversed in the
second group. By showing that only 33% (10/33) of patients were able to identify the correct foot that was actually stimulated by PTNS (compared to 50% by chance), the authors successfully validated this experimental paradigm.

1.5.2.17 PTNS and the Elderly: Early 2009

Early 2009 also saw the publication of a retrospective study looking at the effectiveness of PTNS in elderly patients (Zinkgraf, Quinn, Ketterhagen, Kreuziger, & Stevenson, 2009). To our knowledge, this was the first trial of its kind. It looked at the voiding diaries of 26 women with OAB and urge incontinence (wet group), and 7 women with OAB but without urge incontinence (dry group). Upon completing 12 weeks of weekly 30 minute sessions, 8 (62%) of the 13 wet group patients who finished the trial (13 dropped out for various reasons) saw marked objective improvements in symptoms. Four of the seven dry patients completed the trial and all saw at least a 50% decrease in daytime frequency and nocturia. The study showed the clinical effectiveness of PTNS in a patient population with a mean age of 75.

1.5.2.18 More on the Placebo Effect, Signs of Neural Plasticity: April 2009

After showing that the periodicity of PTNS treatment did not affect the end-result of treating OAB (Finazzi Agro et al., 2005), Finazzi-Agro followed-up with an important paper in April 2009 that discussed neural plasticity as a possible mechanism of action for PTNS (Finazzi-Agro et al., 2009). The group treated 24 patients with OAB symptoms (12, 30 minute sessions over 4 weeks); 16 underwent actual PTNS and 8 were assigned to the sham PTNS group (same sensation as PTNS but no/very little nerve activation). The actual PTNS group achieved a 62.5% response rate, as judged by a 50% decrease in OAB symptoms, while the sham group did not have any responders. The authors further looked at long latency somatosensory evoked potentials (LL-SEP) in all of the patients both before and after the 4 week regimen. In their most basic sense, these signals reflect information processing in the brain after stimulation of the peripheral nervous system. The P80, P100 and P200 waves are of most relevance when discussing the posterior tibial nerve. The results showed that while the mean amplitude of the P200 wave did not change over the 4 weeks in either group, the P80 and P100 mean amplitudes increased.
significantly in the actual PTNS treatment group, while remaining much the same in the sham group. Within the actual PTNS group, there was a larger increase in the mean amplitude of the P80 and P100 waves in the clinical responders when compared to the non-responders. This led them to conclude that PTNS effectiveness may be attributed to an increase in synaptic efficiency caused by modifications PTNS induces in the somatosensory pathway.

1.5.2.19 Long-term PPTNS for MS Patients: Summer 2009

Kabay et al began investigating the long-term effects of PPTNS in a group of 19 MS patients exhibiting OAB symptoms (Kabay et al., 2008). This was in contrast to this research group’s two previous trials (S. C. Kabay et al., 2009; Kabay et al., 2008) that studied the acute effects of PTNS. Nineteen MS patients with neurogenic detrusor overactivity (NDO) were enrolled, and underwent the standard 12 week PTNS protocol. The study saw mean 1st IVDC increase from 124mL at baseline to 218mL after 12 weeks. Mean MCC improved from 200 mL to 267 mL. Statistically significant improvements were also noted in detrusor pressure at the maximal flow rate. The authors described these results as promising but called for a multi-centre prospective trial to verify their results.

1.5.2.20 The First Phase of OrBIT Trial: September 2009

Results from the first randomized, multi-center trial involving PTNS therapy was published by Peters et al (K. M. Peters et al., 2009). Although the trial was not placebo controlled, it compared the clinical efficacy of PPTNS to a major anti-cholinergic medication, Tolterodine. The study involved 50 male and 50 female patients; all with urinary frequency symptoms. Of the 100 patients, 41 of 50 and 43 of 50 patients saw the trial to completion in the PTNS and Tolterodine groups, respectively. Success was determined objectively based on voiding diaries and subjectively via a global response assessment (GRA) of OAB symptoms filled out after the study by both the patient and the investigator. Subjectively, 79.5% of PTNS patients were seen as being cured or showing significant symptom improvements by both the patient and the investigator. 54.8% of Tolterodine patients believed they improved while 60.5% of patients were seen as improved or cured from the perspective of the investigator. The difference between the two groups was not statistically significant. Objectively, 73.2% of PTNS patients and 74.4%
Tolterodine patients saw a significant reduction in voids/day. More PTNS patients saw improvements in nocturia symptoms (70% vs. 61%) and urge incontinence episodes (80% vs. 73%), while Tolterodine did better to reduce the number of moderate to severe daily urgency episodes (70.7% improvement in PTNS vs. 75.6% improvement in Tolterodine). The absence of any statistical differences in clinical measures between the PTNS and Tolterodine arms allowed Peters et al to conclude that PTNS therapy can achieve the same clinical effects as drugs in treating OAB symptoms.

1.5.2.21 More Work on PTNS and Children: Late 2009

Another study investigated the effectiveness of PTNS in pediatric patients. This involved a group of 36 children with a variety of bladder disorders, including: overactive and underactive bladder, and dysfunctional voiding (Capitanucci et al., 2009). Stoller type stimulation was conducted once weekly for 12 weeks. Follow-ups were conducted at the 1 and 2 year mark. At the end of 3 months, twelve of the 14 (86%) overactive bladder cases showed an improvement; and 5 (36%) of these 12 patients were considered cured. At the one year mark, five (42%) of the original twelve patients who responded to PTNS maintained their results while six went into relapse (one relapsed at 6 months and was responding well to chronic stimulation). At the two year follow-up, five (42%) patients remained improved/cured, one relapsed, five were responding to well to chronic stimulation while the final patient was only showing a partial response to chronic stimulation. The study showed that PPTNS is effective in treating overactive bladder in children and that chronic stimulation is needed to maintain results.

1.5.2.22 The Second Phase of the OrBIT Trial: January 2010

This phase of OrBIT consisted of yearlong tracking of the progress of 33 of the 35 PTNS responders from the trial’s first phase. Thirty-two patients were reevaluated at 6 months and 25 returned for revaluation at the one year mark (MacDiarmid et al., 2010). However, unlike previous studies, this trial followed patients as they continued to receive chronic PTNS treatment, where the interval between stimulation sessions was increased to 3 weeks. The GRA continued to show improvements at the 6 and 12 month mark in 94% and 96% of the responders. After a year, daily voids, urge incontinence and nocturia episodes had decreased by an average
of 2.8, 1.6 and 0.8 episodes, respectively. 97% of the patients were able to maintain therapeutic results attained at the end of the original 12 weeks of PTNS therapy. The outcome of this trial strongly supported the long-term effectiveness of PTNS for patients with chronic OAB symptoms.

1.5.2.23 The SUmiT Trial and Level 1 Scientific Evidence: April 2010

Using their proven sham method from early 2009, Peters et al went on to provide the first scientific evidence of PTNS effectiveness. The study was a large multicenter, double-blind, randomized and placebo controlled trial coined the SUmiT trial (K. M. Peters et al., 2010). It looked at the GRA scores, quality of life and OAB symptom questionnaires, and voiding diaries of 220 adults undergoing 12 weeks of either PTNS or sham PTNS (1:1 ratio). Sixty of the 110 PTNS (54.5%) patients showed marked improvement in the GRA compared to 23 of the 110 (20.9%) sham patients. The discrepancy was statistically significant. Voiding diary parameters (frequency, nocturia, urgency and incontinence episodes) all showed statistically significant improvements from baseline to 12 weeks in PTNS patients when compared to sham. Quality of life measures also showed statistically significant increases in the PTNS group when compared to the sham group. This was the first Level 1 clinical evidence that supported the effectiveness of PTNS.

1.5.2.24 Late 2010: Another Successful Placebo Controlled Trial, PTNS in Cats, and the 6 Week PTNS Protocol

Finazzi-Agro et produced a second study, that mimicked the SUmiT trial (K. M. Peters et al., 2010). Using the same sham PTNS method devised by Peters et al (K. Peters et al., 2009), the study randomized 35 females with incontinence stemming from detrusor overactivity into a PTNS treatment (18 patients) or sham treatment (17 patients) arms (Finazzi-Agro et al., 2010). Again, 12 sessions were conducted; this time however, the sessions were spaced out over the course of just 4 weeks (3 times weekly), as opposed to the standard 12 week protocol. The investigators were looking for improvements in quality of life score as well as a 50% or greater reduction in urge incontinence episodes. Seventeen PTNS patients and fifteen sham patients completed the study. Of these, 12 (71%) PTNS patients and none (0%) of the sham subjects were
found to respond to PTNS therapy. Improvements seen in quality of life scores were deemed to be statistically significant in the PTNS group. Quality of life score improvements were seen in the placebo group but they were not statistically significant. The study showed more compelling evidence that PTNS clinical success is unlikely to be attributable to the placebo effect.

Yoong et al questioned the necessity of the standard 12 week protocol in a journal article in December of 2010 (Yoong, Ridout, Damodaram, & Dadswell, 2010). Forty-three female patients underwent a shortened 6 week treatment protocol and returned for a 6 month follow-up. The six treatments were conducted once weekly at 20 Hz using the Stoller Method and the Urgent PC device. It was completed by all forty-three women. The authors were looking for a 50% or greater reduction in frequency episodes and 25% reduction in Incontinence Impact Questionnaire as well as the disappearance of bothersome OAB symptoms. An overall 70% response rate was achieved. Median daytime frequency was cut down to 6.9 from 11.8 episodes at baseline. Likewise, median nocturnal frequency was reduced to 2.4 from 3.5 and median incontinence episodes to 2.4 from 3.5. All these results were statistically significant and no adverse side effects were reported. The authors also showed that OAB symptoms returned without further PTNS within an average of 3 weeks. Understandably, the authors called for a randomized controlled trial comparing the 6 and 12 week protocols. The study implied that a shorter protocol would ultimately have time and cost saving benefits in that it would much sooner identify responders (who can commit to earlier to chronic PTNS stimulation) and non-responders (who know to seek other solutions).

During this period, animal experiments were being conducted to better understand the underlying mechanism of PTNS therapy. One such study was published by Tai et al, where the authors conducted experiments in spinally intact, anesthetized cats (Tai, Shen, Chen, Wang, Roppolo, et al., 2011). Using nerve cuff electrodes implanted on the posterior tibial nerve, the authors investigated changes in bladder capacity that were elicited by both short duration (3-5 minute) and long duration (30 min) PTNS. Two different frequencies were used: 5 Hz and 30 Hz. The main finding of the study was that PTNS could increase bladder capacity by 182% and 161% from baseline after short and long term nerve stimulation, respectively. In addition, the long term
stimulation effect was shown to last for up to 2 hours post-stimulation. Further 5 Hz stimulation during a second cystometry was able to further increase the bladder capacity by about 30%, while 30 Hz stimulation had no such effect. The study concluded that “neuromodulation induces a long-lasting post-stimulation inhibitory effect that is useful in treating overactive bladder symptoms.

1.5.2.25 Foot Stimulation in Cats and a Long Overdue Success with Transcutaneous PTNS: Early 2011

Tai et al further investigated the feasibility of reflexively inhibiting the urinary bladder in cats by electrically activating PTN afferents located in the foot (Tai, Shen, Chen, Wang, Liu, et al., 2011). This was a novel study design that tested the feasibility of applying TPTNS to multiple sites on the foot. Initially, stimulation was applied to the front paw (i.e. human hand) and hind paw (i.e. human heel) but the former was discontinued after a lack of evoked responses. Five and 20 Hz stimulation of the fore foot during cystometry was able to increase the cat bladder capacity by 153% and 137%, respectively. However, unlike their previous study that maintained the results for 2 hours post-stimulation (Tai, Shen, Chen, Wang, Roppolo, et al., 2011), transcutaneous foot stimulation did not elicit any carryover effect.

De Seze revived the largely unpopular TPTNS approach (almost 8 years since the Amarenco et al study) by investigating this nerve stimulation method in 70 MS patients (de Seze et al., 2011). The study followed a 12-week, 20 minute, once a week treatment protocol. Urgency and frequency voiding diary measurements were the primary outcomes while incontinence episodes, symptom score and quality of life were secondary outcomes. These measurements were taken at baseline, day 30 and day 90. 83% of the patients saw clinical improvements in their primary outcomes, while a 30% or greater increase in cystometric capacity was seen in 51% of the patients. Sixty-two (62%) percent of the patients achieved improvements in incontinence at day 30 (with a reduction of 2.7 leakages per week) while 45% achieved complete continence. Furthermore, statistically significant improvements were seen in the rest of the primary and secondary outcomes on day 30 as well. All results remained statistically significant at day 90.
This study was the first in nearly a decade since the initial Amarenco et al study that supported the use of TPTNS as an effective means of treating OAB symptoms.

1.5.2.26 More PTNS in MS Patients and the Optimal Time to Restart Treatment: Late 2011

Coincidentally, a couple of months following the de Seze paper, another paper involving MS patients was published (Gobbi et al., 2011). This study involved 21 patients who received standard PPTNS (30 min sessions, 20 Hz) over 12 weeks. Mean day time frequency was reduced from 9 at baseline to 6 after treatment while mean nocturia episodes was reduced from 3 to 1. Overall, there was an 89% subjective success rate. Additionally, statistically significant improvements in quality of life scores were noted as well.

In November of 2011, Marchal et al answered the question: “how long after the end of treatment, should treatment be restarted?” The study involved 53 subjects followed-up for a maximum of 24 months (Marchal et al., 2011) after initial PPTNS therapy. Cure was defined as 50% reduction in daytime and nighttime voids, 50% decrease in the incontinence questionnaire (ICIQ-SF) and a 50% improvement in 2 or more of the urodynamic parameters. In contrast, improvement was characterized by using 25% improvements in bladder symptoms. The study followed a completely unconventional PTNS protocol: the first 8 weeks were once weekly treatments, followed by 8 weeks of twice weekly treatments concluded by 8 weeks of once monthly treatments for a total of 6 months. At 6 months, 49 of 53 (92%) of subjects were deemed to have been cured or improved. At 12 months, the improvement/cure rate remained consistent in 39 of 43 (92%) patients. Only 16 patients were followed up at 2 years, and only 10 of these (63%) were considered cured or improved. With worsening of symptoms and quality of life scores occurring at 24 months, the researchers concluded that this was the optimal point at which to restart treatment.

1.5.2.27 The STEP Study: 2012/2013

Using the 50 patients that responded to PTNS in the previous SUmiT trial (K. M. Peters et al., 2010), Peters et al investigated the long-term efficacy of PTNS therapy in these patients (K. M.
Peters, Carrico, MacDiarmid, et al., 2013). Thirty-five of these subjects remained at the two year follow-up. After the 12 weekly PTNS sessions, the patients underwent a 14-week tapering off protocol (to once monthly sessions) after which a chronic treatment plan was individualized for each patient. Voiding diaries were conducted every 6 months and symptom improvement questionnaires every 3 months. All voiding diary measurements saw statistically significant improvement at each of the time intervals from baseline to 24 months. Similarly, health-related quality of life scores and OAB-q symptom severity scores showed similar statistically significant improvements at each time point. The authors concluded that after a successful initial 12-week treatment protocol, an average of 1.3 treatments per month or a treatment session every 23 days was sufficient for maintaining control of OAB symptoms. Peters et al. recently published the 3 year follow-up data of these patients, which showed similar results maintained with PTNS (K. M. Peters, Carrico, Wooldridge, et al., 2013).

1.5.2.28 Recent Work Involving Transcutaneous PTNS: 2014

In 2014, additional studies showed further clinical evidence in support of the effectiveness of tPTNS. In one study involving 75 women, the authors showed that improvements in clinical symptoms of OAB were best achieved when tPTNS is used in combination with oxybutynin (Souto et al., 2014).

A second study showed that tPTNS alone was effective in improving OAB symptoms in 50% of patients who were refractory to anti-cholinergic medication (Ammi et al., 2014). In both of these studies, 10Hz was the selected stimulation frequency, while the chosen amplitude as the highest bearable one without causing any subjective pain. Presumably, both these factors were selected based on the prior work by Amarenco et al. (Amarenco et al., 2003) and de Seze et al. (de Seze et al., 2011).

The final study, looked at TPTNS as a means for treating refractory OAB. Using both subjective and objective measures in 42 patients, the group observed a 39% (objective, i.e. voiding diary) and 50% (subjective, patient satisfaction) response in the patients after the 12-week treatment
protocol. The group concluded that TPTNS is a safe and effective way to counter OAB—so much so that it warrants a randomized, placebo-controlled trial.

1.6 Research Objectives

As outlined in the previous sections, PTNS therapy has made significant advancements as an effective treatment option for OAB since the pioneering work by McGuire et al in 1983. However, a review of the brief history of PTNS, highlights numerous gaps in the scientific literature. The most notable of these are: (1) the lack of a physiological rationale for setting the stimulation parameters; and (2) the unknown role of the PTN branches in this reflex pathway. Over the last two years, I have focused on testing our main hypothesis: that PTNS-evoked bladder reflexes are mediated by the individual PTN branches. To this end, I have focused on the following two primary objectives. The two primary objectives of my research, shaped by the gaps in PTNS literature, are outlined below:

1. Exploration of varying PTNS parameters to improve treatment efficacy

2. Targeting the distal branches of the PTN to compare the effect to PTN trunk stimulation

Each of these research objectives is outlined in more detail in the following subsections. The rationale for addressing each of the objectives, as well as the relevant literature and methods used are analyzed in further detail in the following chapter.

1.6.1 Varying PTNS Parameters

To date, all clinical work on PTNS has used a frequency of 20Hz and an arbitrary amplitude—the highest tolerable amplitude or the one which causes the hallux (big toe) to fan out (defined as 1T). Since these parameters were initially selected and shown to work by McGuire in 1983, there has been very little motivation to experiment with different parameters, particularly given that the current settings can achieve clinical effectiveness.
However, it is important to note that the current PTNS response rate varies from 60-80%, and the reduction of bladder symptoms are also quite notable (~50%). It is possible that fine tuning the stimulation parameters (e.g., frequency and amplitude of stimulation) can help achieve even higher clinical response rates and overall outcomes. Thus, one of the main goals of this thesis is to further investigate these parameters, and explore ways to optimize the PTNS treatment modality. As will be discussed in the Theory & Methods section, the work in this thesis looks at frequencies of 2, 5, 10, 20 and 50Hz as well as amplitudes of varying from the 1-10T.

1.6.2 Targeting the Distal Plantar Branches of the PTN

All studies to date, clinical and animal, have applied PTNS at the level of the medial malleolus, just above the ankle. While previous work has looked at the whole nerve, there are multiple pathways that are derived from this nerve trunk and the purpose of this thesis was to determine whether electrical stimulation had any effect on the bladder. Electrical stimulation was applied independently to the medial and lateral plantar branches of the PTN while the effects on bladder activity were being recorded. This is the first study that has investigated the effects of selective PTN branch stimulation on bladder function. The completion of this research objective will not only provide insight into the neural mechanisms of PTNS reflexes but it will also help in developing alternative methods of delivering PTNS therapy.
Chapter 2
Theory & Methods

2.1 Study Rationale

The following section will outline the current preclinical literature of PTNS and its effects on the lower urinary tract function (i.e. the bladder). This outline will serve to provide the overall rationale for the experimental procedure used for this work; why specific stimulation parameters were tested, experimental approaches used and stimulation protocols carried out.

2.1.1 Electrical Neuromodulation of the Lower Urinary Tract

Electrical neuromodulation of the lower urinary tract has been investigated over the last several decades, where direct electrical activation of various sensory nerve afferents has identified multiple reflex pathways that can influence bladder function. Studies show that reflexive inhibition of the bladder can be achieved through sacral spinal nerve stimulation (SNS) (Comiter, Mazar, Phull, & Salkini, 2010; Jezernik, Grill, & Sinkjaer, 2001; Su, Nickles, & Nelson, 2012a; Walter, Siderous, Robinson, Wheeler, & Wurster, 1992) or by electrical activation of the pudendal nerve (Tai, Chen, Shen, Wang, Liu, et al., 2011; Tai, Chen, Shen, Wang, Roppolo, et al., 2011; Tai, Shen, Chen, Wang, Liu, et al., 2011; Tai, Shen, Chen, Wang, Roppolo, et al., 2011; Woock, Yoo, & Grill, 2008). Both clinical and animal data suggest that these inhibitory reflexes involve a direct spinal cord-mediated pathway that exhibits maximum response at stimulation frequencies between 5 Hz and 20 Hz (Snellings, Yoo, & Grill, 2012; van der Pal, Heesakkers, & Bemelmans, 2006). Conversely, studies have also identified specific sensory nerves that can electrically activate excitatory bladder reflexes (e.g., evoke sustained bladder contractions). These include the urethral sensory and dorsal genital branches of the pudendal nerve in cats, where maximum excitatory and inhibitory effects are observed during nerve stimulation at 2-5 Hz and 25-40 Hz, respectively (J. Wang et al., 2009; Woock et al., 2008; Yoo, Woock, & Grill, 2008).
2.1.2 The PTNS Mechanism

As an alternative approach, posterior tibial nerve stimulation (PTNS) has been investigated as a means of modulating bladder function. Clinical studies have shown that PTNS-induced neural plastic changes can reduce OAB symptoms (McGuire et al., 1983) over long periods (i.e., months) with repeated nerve stimulation (Finazzi-Agro et al., 2010; Finazzi Agro et al., 2005; McGuire et al., 1983; K. M. Peters, Carrico, MacDiarmid, et al., 2013; K. M. Peters et al., 2010; K. M. Peters, Carrico, Wooldridge, et al., 2013; V. Vandoninck, M. R. Van Balken, E. Finazzi Agro, F. Petta, C. Caltagirone, et al., 2003; V. Vandoninck, M. R. van Balken, E. Finazzi Agro, F. Petta, F. Micali, et al., 2003). The nature of this bladder-inhibitory reflex pathway has been studied in various animal models, where acute changes in bladder function have been described in response to finite-duration PTNS. In anesthetized cats, reflex inhibition of bladder function (e.g., increased bladder capacity) was achieved by both direct PTN and transcutaneous hind foot stimulation at frequencies of either 5Hz or 20Hz (G. Chen et al., 2012; Tai, Chen, Shen, Wang, Roppolo, et al., 2011; Tai, Shen, Chen, Wang, Roppolo, et al., 2011). More recently, Su et al reported robust bladder inhibition in anesthetized rats (Su et al., 2012a), where the authors showed that PTNS-evoked responses were tuned to a very narrow range of stimulation parameters: frequency of 10Hz and amplitude of 3T (T=foot twitch threshold). Given the multitude of bladder reflexes that can be evoked by electrical nerve stimulation (e.g., pudendal nerve reflexes, sacral nerve stimulation and intravesical stimulation), these recent findings further validate the rat as an appropriate model for investigating bladder neuromodulation reflexes evoked by PTNS (H. Y. Chang, Cheng, Chen, Peng, & de Groat, 2006; Jiang, 1998; Y. Wang & Hassouna, 2000; Zvara, Sahi, & Hassouna, 1998).

2.1.3 Limitations with Current PTNS Parameters

However, there are significant limitations in our understanding of which specific stimulation parameter(s) are relevant to modulating bladder function by PTNS. Such variables include the diameter of the recruited axons, total number of activated fibers, specific anatomical subset(s) of fiber bundles (i.e., nerve fascicles), and the frequency at which axons are pulsed. The current
clinical data indicates that therapeutic effects can be achieved by PTNS applied at 20 Hz (Finazzi-Agro et al., 2010; K. M. Peters, Carrico, MacDiarmid, et al., 2013; K. M. Peters et al., 2010; K. M. Peters, Carrico, Wooldridge, et al., 2013; K. M. Peters et al., 2009) and even 10 Hz (de Seze et al., 2011). That this bladder-inhibitory reflex also appears to work in cases of neurogenic bladder (e.g., multiple sclerosis and spinal cord injury) suggests PTNS may possibly involve a spinal-mediated reflex pathway (Andrews & Reynard, 2003; de Seze et al., 2011; Gobbi et al., 2011; S. Kabay et al., 2009). As demonstrated in previous work involving pudendal nerve stimulation (Boggs, Wenzel, Gustafson, & Grill, 2006a; Woock et al., 2008; Yoo et al., 2008), characterization of stimulation parameters that can modulate bladder function is a key step in understanding the mechanisms of PTNS.

2.1.4 The Goals of this Work

In this study, we investigated the input-output relationship between PTNS and changes in bladder function. Using an urodynamic (continuous-fill) bladder model in anesthetized rats, we varied the stimulation parameters over a broad range of amplitudes and frequencies. The results of this study showed that PTNS can elicit a complex set of bladder responses that strongly depend on the stimulation frequency. PTNS-evoked responses were identified as either an inhibitory or excitatory bladder reflex, where each response was further classified as acute or prolonged. In addition, we found that selective electrical activation of individual PTN branches (medial and lateral plantar branches) can also modulate bladder function.
2.2 Materials and Methods

All surgeries and procedures were approved by the University of Toronto Animal Care Committee in accordance with the regulations of the Ontario Animal Research Act (Toronto, ON, Canada). Experiments were conducted on female Sprague-Dawley rats (n=11) weighing 250-300g. Anesthesia was initially induced with 5% isoflurane (induction chamber) and maintained with a gas mask (2-3% isoflurane, O\textsubscript{2} flowrate: 0.1 L/min) for the duration of the surgical procedure (2-3 hrs). Once set-up was complete, the anesthesia was transitioned from isoflurane to urethane (2 IP injections, 5 minutes apart, 1.2 g/kg). Heart rate (300-400 beats/min), blood O\textsubscript{2} level (97-100%) and blood temperature (37-39° Celsius) were monitored throughout the experiment. If needed, supplemental doses of urethane (0.25-0.5 dose, 1.2g/kg) were administered. The animal was maintained on urethane for 8-10hrs. At the end of the experiment the animal was euthanized via overdose of isoflurane. (5% isoflurane, delivered over 10-15 minutes).

Figure 2. Experimental set-up in urethane anesthetized rats. Repeated bladder contracts were generated by continuous infusion of saline through a suprapubic catheter.
2.2.1 Electrical Stimulation of the PTN and PTN Branches

A surgical incision was made on the medial ankle rostral to the foot. This exposed the posterior tibial nerve and allowed for the implant of a custom-made bipolar (inter-electrode distance=5mm) platinum nerve cuff electrode (NCE). The NCE was connected to an isolated pulse stimulator (A-M Systems, Carlsborg, WA, USA) which delivered constant-current pulses (0.2ms, square pulses) at varying amplitudes and frequencies. Selective electrical stimulation of the medial and lateral plantar branches was achieved by fine dissection of the PTN trunk at the initial incision site. The NCE was then placed on either branch and secured via 6-0 sutures. The stimulation amplitude was slowly increased until the first foot twitch was observed. Stimulation evoked twitches of the hallux and the lateral digits indicated the selective activation of the medial and lateral plantar branches, respectively.

![Figure 2](image)

**Figure 2.** Detailed illustration of the surgically implanted nerve cuff and foot EMG electrodes. A bipolar nerve cuff electrode was implanted on the PTN trunk and connected to a constant-current source. Foot EMG was recorded by inserting insulated wires (1) between the hallux and 2nd digit, and (2) at the mid-sole of the foot.

2.2.2 Bladder Pressure Recording and Saline Infusion

Following an abdominal incision to expose the bladder (Figure 1), a small cut was made at the dome of the bladder, through which a cannula (PE50) was inserted and secured with 6-0 sutures.
The abdomen was closed using 4-0 sutures. Externally, the cannula was connected in series to a pressure transducer (Utah Med, Midvale, UT, USA) and an infusion pump (Harvard Apparatus, Holliston, MA, USA). Bladder pressure signals were conditioned using a bridge amplifier (iWorx, Dover, NH, USA).

2.2.3 Foot EMG and EUS EMG Recording

A pair of 35-gauge, PFA-coated, multi-stranded, stainless steel wire electrodes (A-M Systems, Carlsborg, WA, USA) were used to record foot and external urethral sphincter (EUS) EMG activity. Recording of the foot EMG (fEMG) was achieved by inserting one of the electrodes between the hallux and long digit and the midsole of the foot (Figure 2). For EUS EMG recordings, a pair of electrodes were injected bilateral to the urethral meatus (Figure 1).

2.2.4 Data Conditioning and Acquisition

EMG electrode signals were filtered and amplified (EMG: Bandwidth= 100Hz-3000Hz, Gain=1000) through a low-noise voltage preamplifier (Stanford Research Systems, Sunnyvale, CA, USA). All data was acquired using a PowerLab data acquisition system (AD Instruments, Colorado Springs, CO) and integrated onto the computer via LabChart software (AD Instruments, Colorado Springs, CO) at a sampling rate of 20k samples per second.

2.2.5 Stimulation Amplitude Settings

In each rat, the minimum stimulus amplitude that evoked any skeletal muscle contractions of the foot during stimulation of the PTN trunk, the lateral branch and the medial branch was defined as T\textsuperscript{m}, T\textsuperscript{m-l} and T\textsuperscript{m-m}, respectively. The tibial nerve was stimulated at 1, 2, 4, 6, 8 and 10 times T (biphasic, square, pulse width=200µs, 2Hz, 10 sec) resulting in 20 signals recorded by the fEMG electrodes. At each amplitude (Figure 3A), all 20 signals were averaged and the area under the EMG curve (AUEC) was calculated and normalized by the AUEC at 10T. The AUEC (% activation) was plotted against amplitude to obtain an EMG recruitment curve (Figure 3B). From
the recruitment curve we can infer about the percentage of nerve activation. This was repeated for stimulation of the medial and lateral branches of the tibial nerve at multiples of $T_{m-m}$ and $T_{m-l}$ respectively (Figure 3B). Based on these recruitment characteristics the default stimulation amplitude for all nerves (PTN, lateral plantar and medial plantar) was set to 6T.

2.2.6 Acquisition and Interpretation of BRC Data

Periodic bladder rhythmic contractions (BRC) were achieved by continuous saline infusion (0.1-0.3 mL/min) via the suprapubic catheter. The urinary bladder was repeatedly filled and emptied by either sustained contractions or by passive leakage through the urethral meatus. Increases in bladder pressure (>15 cmH$_2$O) with concomitant high-frequency bursts in EUS EMG activity were indicative of a single contraction. The experimental protocol involved an initial control period of BRC (no stimulation) lasting 10 min, followed by a 10 min stimulation period during which the tibial nerve was stimulated at 6$xT_m$ and a given frequency. This process of alternating 10 min control and stimulation periods was repeated for 2, 5, 10, 20 and 50Hz, applied in a random fashion. In some cases, passive leakage of the urinary bladder was observed. These cases were characterized by a period of sustained and constant high-pressure (20-30 cmH$_2$O) in the bladder, during which no bladder contractions or characteristic high-frequency EUS EMG bursts were observed. Although 50Hz stimulation was able to restore BRC, these results were not factored into our data. In these cases of passive leakage, upon resumption of bladder contractions, the bladder was usually allowed a 30 minute period to recover before PTNS was resumed.

The average inter-contraction interval (AICI) was determined for both the 10-min “stimulation” and “post-stimulation” periods. Using the AICI, we determined the average bladder contraction frequency (aBCF) over each 10min period. The aBCF for each period was normalized using the control period preceding the “stimulation” and expressed as the % change in aBCF with respect to control (Figure 4). Throughout all experiments, we defined a “10% decrease in aBCF” as the minimum level of change in bladder function. Any changes in aBCF above 10% were deemed inhibitory. The time needed to complete the stimulation protocol varied from 8-10hrs. Data was processed using LabChart (AD Instruments, Colorado Springs, CO) and was further analyzed.
using MatLab (MathWorks, Torrance, CA, USA) and MS Excel (Microsoft, Redmond, Washington, 2011). The entire process was repeated for the medial and lateral plantar branches using stimulation amplitudes of $6xT_{m,m}$ and $6xT_{m,l}$, respectively.

### 2.2.7 Statistical Significance

In our statistical approach, we considered 2Hz stimulation (PTN trunk) as our control parameter. In our previous work using an identical experimental setup, we observed that 2Hz stimulation had no effect on the bladder either during or immediately following the stimulation period (Kovacevic & Yoo, 2014). This has also been shown to hold true in previous animal studies by other groups (Su et al., 2012a). Using 2Hz as our control stimulation parameter, we applied a Mann-Whitney U test (non-parametric test) at each stimulation frequency using a standard confidence interval of 95%. All data are summarized as mean +/- SE, unless otherwise indicated.
Chapter 3
Results

Experiments were conducted in eleven rats, where the fEMG was recorded in response to PTNS in every study. In eight (8/11) of these experiments, we generated additional fEMG recruitment curves for the medial and lateral PTN branches. Reflex BRCs and PTNS-evoked changes in bladder function were achieved in every experiment. However due to the premature expiration of 4 rats, complete sets of data (PTNS + PTN branch stimulation) we were obtained in only 7 experiments.

Figure 3. Sample fEMG recording data. A. Recordings from a single experiment during electrical stimulation of the PTN trunk, where 20 pulses were applied at 2 Hz. Each line represents the fEMG activity averaged over 20 pulses and repeated at different amplitudes (1Tm, 4Tm, 10Tm, where Tm=10 μA). Very little difference in fEMG is observed between PTNS at 4Tm and 10Tm. B. The recruitment curves of fEMG is compared among electrical stimulation of the PTN (n=11), medial plantar (n=7), and lateral plantar (n=7) nerves. All activity is normalized to the % activation at 10T for stimulation of each neural target. The threshold values (mean ± SD) for evoking an fEMG response during PTN trunk, lateral and medial plantar branch are 18.7 ± 15.6 μA, 18.8 ± 11.1 μA and 89.1 ± 141.6μA, respectively. PTN trunk is maximally activated at 4Tm, whereas full fEMG activation by medial and lateral branches occur at 6Tm-m, and 6Tm-l, respectively.
3.1 Electrical Recruitment of the Posterior Tibial Nerve

The threshold amplitude for eliciting a foot twitch during PTNS (Tm) was 18.7±15.6μA (n=11, range: 5-60μA). Figure 3B shows that the fEMG reaches maximum muscle fiber activation at 6Tm, with 64.7±32.5% (n=11, range: 4.5-103.3%) achieved at 2Tm, and 89.3±16.0% (n=11, range: 45.5-104.9%) attained at 4Tm. Both the waveform (Figure 3A) and the high level of fEMG (Vpp or RI) suggests that smaller Aδ fibres were fully recruited at or beyond 6Tm.

Selective electrical stimulation of the medial and lateral branches of the PTN showed fEMG thresholds of 18.8±11.1μA (Tm-m, n=7, range: 6-40μA) and 89.1±141.6μA (Tm-l, n=7, range: 5-400μA), respectively. The data indicates that full activation of the medial branch occurred at 8Tm-m corresponding to 99.4±4.0% (n=8, range: 92.6-105.1%), and also at 8Tm-l, 95.8±3% (n=8, range: 91.2-99.3%) for the lateral branch. It is noted that the initial activation threshold of the medial nerve appears to occur at a lower amplitude than that of the lateral branch. Similar to PTN trunk stimulation, electrical stimulation of either nerve branch approximates maximum recruitment by 6T stimulation.

Figure 4. The effects of PTNS frequency on changes in urodynamic bladder function. Changes in the average bladder contraction frequency (aBCF, mean ± SE) were calculated
both during stimulation (stim, 10-minute pulse train) and following the termination of PTNS (post, 10-minute duration). The aBCF significantly decreased during PTNS (labeled stim) at 5Hz (n=10) and 10Hz (n=11), as well as during the 5Hz (n=10) post-stimulation period (Mann-Whitney Test, p < 0.05).

3.2 PTNS-Evoked Changes in Bladder Function

Based on the results from the fEMG recruitment, we opted to use 6T as the stimulation amplitude. By using this level of stimulation amplitude, we are confident in the activity of the majority of the nerve fibres of the posterior tibial nerve trunk. This also eliminated one variable (stimulation amplitude) and permitted a study of different stimulation frequencies and target nerves.

Our data shows that PTNS-evoked inhibition of the urinary bladder was statistically significant when electrical pulses were applied at 5Hz and 10Hz. Referring to Figure , PTNS at 10Hz exhibited the strongest inhibitory effect, decreasing aBCF by 41.8 ± 11.6% (n=11, range: 5.1% to 100%, p=0.002) during the stimulation period. An example of acute inhibition of the bladder during 10Hz PTNS is shown in Figure 5A. However, PTNS at 10 Hz failed to elicit significant bladder inhibition during the post-stimulation period (decrease in aBCF by 9.1±13.5%, n=11, range: -48.5% to 100%, p=0.248). Acute bladder inhibition (as measured by at least a 10% decrease in the aBCF) during 10Hz stimulation was observed in 8 of 11 (72%) experiments (Figure 6A); whereas prolonged bladder inhibition was found to occur in only 6 of 11 (55%) experiments (Figure 6B).
Figure 5. Electrical neuromodulation of bladder function as a result of PTNS (trunk) applied at various stimulation frequencies. A. PTNS applied at 10Hz inhibited the bladder completely during the stimulation period. B. 5Hz inhibition during the post-stimulation period. C. After 45 mins of bladder inactivity, 50Hz excitatory effect. D. 50Hz PTNS exciting the bladder during regular bladder contractions.

PTNS at 5Hz resulted in statistically significant bladder inhibition both during and following the 10-minutes of electrical stimulation. Acute bladder inhibition (Figure 4) was characterized by a 23.9±11.0% (n=10, range: -30.9% to 100%, p=0.024) decrease in aBCF during the stimulation period; whereas prolonged bladder inhibition (Figure 5B) resulted in a 35.1±13.9% (n=10, range: -30.9% to 100%, p=0.01) decrease in aBCF (Figure 4). Both types of responses were observed in 50% (5 of 10 rats, Figure 6A) and 80% (8 of 10 rats, Figure 6B) of experiments, respectively. Although the prolonged bladder-inhibitory effect typically disappeared within the 10-minute post-stimulation period (9 out of 11 rats), longer periods (30-60min) of suppressed bladder activity was observed in 2 rats (duration = 27.5 ± SE min, Figure 8).
Figure 6. Frequency-dependent distribution of experiments that exhibited PTNS-evoked inhibitory or excitatory bladder responses. A. Distribution of the effects of PTNS at different frequencies during the 10min stimulation period (n=11). PTNS applied at 10Hz caused at least a 10% inhibitory effect in 8 of 11 (72%) of experiments. Stimulation at 5Hz (5 of 10) and 20Hz (6 of 11) was effective in about half the experiments, whereas 2Hz and 50Hz stimulation was relatively ineffective. B. Distribution of the effects of PTNS at different frequencies during the 10min post-stimulation period (n=11). PTNS caused at least a 10% decrease in aBCF in 8 of 10 (80%) animals at 5Hz, 8 of 11 (72%) of experiments at 20Hz, and 6 of 11 experiments at 10 Hz.

In one experiment, PTNS applied at 10Hz (6T_m, duration= 5min) resulted in an exceptionally long (approximately 1 hr) period of post-stimulus bladder inhibition (Figure 8B). During this period of bladder inactivity, external pressure applied to the abdominal wall (at the 15min mark) failed to induce bladder contractions. BRCs resumed only following a 10-minute train of pulses applied to the PTN at 50Hz and 6T_m. This ‘extended period’ of bladder inhibition was not repeatable in subsequent experiments. As indicated in Figure 4, bladder-inhibition following PTNS trials applied at 10 Hz were typically non-existent or wore off well before the end of the 10 min post-stimulation period.

3.3 Excitatory Bladder Reflex Evoked by PTNS
In contrast to the suppression of bladder activity elicited by PTNS applied between 5 and 10 Hz, PTNS at 50Hz resulted in an excitatory shift in bladder function (increase in aBCF, Figure 5D). This resulted in a 34.8±19.2% (n=9, range: -33.1% to 142.6%, p=0.06) increase in aBCF during nerve stimulation (Figure 4). Although the average response was large, the overall effect was found to be marginally not statistically significant. This effect often carried over into the post-stimulation period, where the aBCF increased by 37.4±21.5% (±standard error, n=9, range: 27.7% to 123.5%, p=0.07). Again, the results were marginally not statistically significant. As shown in Figure 7, 4 of 9 (44%) experiments exhibited at least a 10% increase in aBCF during the stimulation period, while 5 of 9 (56%) exhibited at least a 10% increase in aBCF during the post-stimulation period.

![Figure 7. Frequency-dependent distribution of experiments that exhibited 50Hz PTNS-evoked excitatory bladder responses (PTN trunk stimulation).](image)

The excitatory effect of PTNS at 50 Hz was clearly demonstrated after 50Hz PTNS was applied during prolonged periods of bladder inhibition (1hr period following 10Hz PTNS, Figure 8B). During this period, there was constant leakage of saline through the urethral meatus but no EMG bursting activity (i.e. no bladder contractions). Instead, only random EUS EMG spikes (corresponding to each drop of saline) were recorded. As shown in Figure 8B, BRCs resumed within 5 minutes of 50Hz PTNS. In another experiment (Figure 5B and C), 5Hz PTNS resulted
in a similar leaky bladder that persisted following a PTNS trial applied at 5 Hz. In this example, BRCs were restored within 8 minutes of PTNS applied at 50Hz.

Figure 8. Prolonged inhibitory effects of PTNS (PTN trunk). A. 5Hz stimulation results in a 30min post-stimulation inhibitory effect. B. 10Hz stimulation inhibits the bladder for a period of up to 60min post-stimulation. Manual pressure was not able to induce bladder contractions but upon application of 50Hz PTNS, bladder contractions resumed.

3.4 Effects of Selective Medial Plantar Nerve Stimulation

Electrical stimulation of the medial plantar branch at 10 Hz resulted in a statistically significant inhibition of bladder function both during and following the 10-minute period of stimulation (Figure 9). The average decrease in aBCF was 38.9±6.3% (n=7, range: 17.3% to 66.7%, p<0.001) and 27.7±12.1% (n=7, range: -25.0% to 68.1%, p=0.03), respectively. Acute inhibition of bladder function was observed in all 7 of 7 (100%) experiments (i.e., minimum 10% decrease
in aBCF, Figure 10A). Similarly, prolonged bladder inhibition was observed in 5 of 7 (71%) of experiments (Figure 10B).

Figure 9. The effects of frequency-dependent, medial PTN branch (medial plantar nerve) stimulation on bladder function was quantified by percent change in aBCF, both during (Stim, 10 minutes) and after (Post, 10 minutes) the continuous train of electrical pulses. There were statistically significant decreases in aBCF during the 5 Hz (n=7) and 10 Hz (n=8) post-stimulation period, as well as the 10Hz (n=7) stimulation period (Mann-Whitney test, p<0.05). A post-stimulus increase in aBCF was observed at 20 Hz PTNS, but we were not able to determine whether this was an excitatory response, or a rebound effect elicited by the bladder inhibition observed during the 20Hz stimulation period.

Selective medial plantar branch stimulation also elicited significant bladder inhibition at 5 Hz, but only during the post-stimulation period. As shown in Figure 9, the aBCF decreased by an average of 29.7±15.2% (n=7, range: -12.7% to 48.6%, p=0.05). During this prolonged inhibitory response, at least a 10% decrease in aBCF was seen in 5 of 7 (71%) of experiments, while another 50% decrease in aBCF was observed in 2 of 7 (29%) experiments (Figure 10B). There were no statistically significant changes in aBCF during 2Hz, 20Hz or 50Hz stimulation.
Figure 10. Frequency-dependent distribution of experiments that exhibited medial PTN branch-evoked inhibitory bladder responses. A. Distribution of the effects of medial PTN branch stimulation at different frequencies during the 10min stimulation period (n=7). Electrical stimulation applied at 10Hz caused at least a 10% inhibitory effect in all (100%) experiments. Stimulation at 2Hz (1 of 6), 5Hz (1 of 7), 20Hz (2 of 5) and 50Hz (1 of 6) was relatively ineffective in inhibition the bladder. B. Distribution of the effects of medial PTN branch stimulation at different frequencies during the 10min post-stimulation period (n=7). Stimulation resulted in at least a 10% decrease in aBCF in 5 of 7 (71%) experiments at 5 Hz and in 5 of 7 (71%) of experiments during 10Hz stimulation. Simulation at 2Hz and 50 Hz had a very modest effect (both 3 of 6 experiments), whereas 20Hz stimulation did not have any effect during the post-stimulation period.

3.5 Effects of Selective Lateral Plantar Nerve Stimulation
Lateral plantar nerve stimulation at 10Hz induced statistically significant bladder inhibition during both the stimulation and post-stimulation periods (Figure 11). During stimulation the aBCF decreased by an average of 14.4±4.9 % (n=6, range: -1.8% to 29.2%, p=0.02), with a minimum 10% reduction in aBCF observed in 4 of 6 (67%) experiments (Figure 12A). Figure 12B shows that in all six experiments, the aBCF decreased during the post-stimulation period by at least 10%, with an overall decrease in aBCF of 33.3±6.8% (±standard error, n=6, range: 14.8% to 58.6%, p=0.002, Figure 11).
Interestingly, lateral PTN branch stimulation at 20Hz also induced a statistically significant decrease in aBCF during both the stimulation and post-stimulation periods (Figure 11). Acute bladder inhibition resulted in a 21.9±7.5% (n=6, range: 0.0% to 53.0%, p=0.02) decrease in aBCF, and prolonged bladder inhibition exhibited a 17.7±4.9 % (n=6, range: -2.6% to 34.1%, p=0.008) drop in aBCF. Figure 12 shows that 20Hz stimulation of the lateral PTN branch resulted in a minimum 10% decrease in aBCF in 4 of 6 (67%) of experiments during the stimulation period and in 5 of 6 (83%) experiments during the post-stimulation period. Lateral PTN branch stimulation at 2Hz and 5Hz did not have any effect on the urinary bladder.

Selective electrical activation of the lateral plantar nerve at 50 Hz showed evidence of an excitatory bladder reflex, both during and following the 10-minute duration of stimulation. This resulted in increases in aBCF of 50.3±35.7% and 131.0±102.1%, respectively (Figure 11). Although this effect was not consistent (only 3/7 and 4/6 rats showed at least 10% aBCF excitation) and therefore, not statistically significant (p = 0.104), these responses confirmed that the bladder excitatory response elicited by PTNS at 50 Hz (Figure 4) was mediated specifically by afferents contained within the later plantar nerve.
Figure 12. Frequency-dependent distribution of experiments that exhibited lateral PTN branch-evoked inhibitory bladder responses. A. Distribution of the effects of lateral PTN branch stimulation at different frequencies during the 10 min stimulation period (n=7) show that nerve stimulation at 10Hz and 20Hz caused at least a 10% inhibitory effect in 4 of 6 (66%) experiments while stimulation at 2Hz (1 of 7), 5Hz (1 of 6), and 50Hz (1 of 7) was comparatively ineffective in eliciting a bladder response. Distribution of the effects of lateral PTN branch stimulation at different frequencies during the 10 min post-stimulation period (n=7) show that stimulation caused at least a 10% decrease in aBCF in all 6 of 6 (100%) experiments at 10Hz, and in 5 of 6 (84%) of experiments at 20Hz. Lateral PTNS had limited inhibitory effects at 2Hz (3 of 7 experiments) and 5Hz (2 of 6 experiments), and 50Hz (1 of 6 of experiments).
Chapter 4
Discussion

In this study, we significantly expand upon our knowledge of the PTN-to-bladder inhibitory reflex and, in addition, describe for the first time an excitatory reflex mediated by PTN or LPN afferents. We also show, for the first time, that bladder inhibition can be elicited by selective PTN branch stimulation. Our findings indicate that electrical stimulation of the posterior tibial nerve trunk primarily mediates bladder-inhibitory reflexes that can be characterized as either acute or prolonged responses, and that these reflex pathways are tuned to specific stimulation frequency values: 10 Hz (acute) and 5 Hz (acute and prolonged). Selective electrical activation of individual branches of the PTN (medial and lateral plantar) also shows that both types of inhibitory responses can be elicited by activation of either neural pathway. Interestingly, we also describe an excitatory bladder reflex that is evoked by PTNS applied at stimulation frequencies (50 Hz) above those that elicit bladder inhibition. Selective PTN branch stimulation confirmed that this excitatory reflex is mediated by afferents located within the lateral plantar nerve.

4.1 The Inhibitory Effects of PTN Afferents

Previous studies in cats have shown that a broad range of frequencies (5Hz, 20Hz, 30Hz) have an inhibitory effect on the urinary bladder (Tai, Shen, Chen, Wang, Liu, et al., 2011; Tai, Shen, Chen, Wang, Roppolo, et al., 2011). Although, the present study did not consider 30Hz stimulation, we have observed that stimulation within a similar frequency range (5-20Hz) is required for inhibition of the urinary bladder in rats. Reflex bladder inhibition by PTNS within this range has been shown previously to evoke an acute bladder inhibitory reflex in rats (Su et al., 2012a). Interestingly, this study observed bladder inhibition only during PTNS at 10Hz, while failing to observe statistically significant responses at other tested frequencies. In comparison, we have shown statistically significant bladder inhibition at 5Hz (entire PTN, medial branch), 10Hz (entire PTN, medial & lateral branch) and 20Hz (medial branch only). Other studies, such as that by Matsuta et al have shown a similar 5Hz post-stimulation effect induced by PTNS in rats. Although, this is in agreement with our results, it is interesting to note
that Matsuta et al did not observe the 5Hz acute inhibitory effect. We hypothesize that this is likely due to the fact that they were applying PTNS at 2-4T_m compared to the present study, which used 6T_m (Matsuta, Roppolo, de Groat, & Tai, 2014).

4.2 Considerations of PTNS Amplitude

It is interesting to note that Xu et al observed significant PTNS-evoked bladder inhibition only at stimulation amplitudes of 3T_m, while our study required pulses applied at 6T_m to evoke bladder inhibition. One of the main causes of this discrepancy is the difference in how T_m was experimentally determined. We defined T_m by directly measuring the evoked fEMG signal; whereas the Su et al study defined motor threshold by visually observing foot twitches. As a consequence, our T_m (0.018mA) value was much lower than that of the Xu et al study (0.16mA), which translated to current pulse amplitudes of approximately 0.1 mA (6T_m) and 0.5 mA (3T_m), respectively. This difference could also be attributed to the method by which the PTN was electrically activated. Our custom-fabricated bipolar nerve cuff electrodes (platinum contacts embedded in a silicone elastomer) likely provided enhanced nerve fiber activation at lower amplitudes, compared to bare-wire monopolar electrodes. In a cat model, Tai et al showed that the bladder-inhibitory effects of PTNS could be further augmented by increasing the stimulation amplitude, from 2T_m to 4T_m (Tai, Chen, Shen, Wang, Roppolo, et al., 2011; Tai, Shen, Chen, Wang, Roppolo, et al., 2011). These authors also used a nerve cuff electrode to directly activate the PTN, but instead used visually observed foot twitches to define T_m. Since one would expect that directly measuring the fEMG provides a more sensitive means of determining motor threshold (T_m) than by visual observation, we estimated that this cat study achieved a similar level of PTN fiber activation to that of our present study.

Additionally, Matsuta et al did not observe acute bladder inhibition while stimulating the PTN at amplitudes of 2-4T_m during cystometrograms (CMGs) in rats (Tm determined by visually observed foot twitches) despite observing prolonged inhibition after 30 minutes of stimulation (Matsuta et al., 2014). This is in line with the results from our preliminary work (Kovacevic & Yoo, 2013) where PTNS applied at 2–4T did not result in bladder inhibition and follows with our reasoning for selecting 6T_m as the stimulation amplitude. Consequently, the absence of acute
bladder inhibition in response to 5 Hz PTNS suggests that Matsuta et al did not recruit the full complement of PTN fibers.

![Graph showing ENG activity](image)

**Figure 13.** ENG Activity elicited by $200T_m$ stimulation. Stimulation applied to the PTN via a bipolar nerve cuff electrode. ENG recording was performed proximally at a distance of 3cm proximal to the stimulating electrode. A tripolar nerve cuff recording electrode was placed on the sciatic nerve to perform the recordings. Results shown above are from a single trial at $200T_m$. Stimulus is applied at $t=0$ms. Observed C-fibre activity can be seen at $t=2.5$ms. The action potential is shown to occur at $t=1$ms.

#### 4.3 The Relationship between Fiber Diameter and Bladder Reflexes

Although we failed to evoke any changes in bladder or EUS function during PTNS applied with pulse amplitudes below $6T_m$, (Kovacevic & Yoo, 2013), we found that almost 98.9% of
myelinated fiber activity was elicited at this level of stimulation (Figure 3B). As we increased the amplitude up to $200T_m$, we began to observe neural activity with a latency ($t=2.5\text{ms}$) corresponding to unmyelinated C-fibres (Figure 13). Earlier work by Sato et al examined the recruitment of different nerve fiber types within the left hind limb of cats using almost identical stimulation parameters (bipolar, monophasic square pulse, 0.2ms pulse width) and a stimulation-recording electrode configuration (ENG recording-electrode located 2 centimeters proximal to stimulating electrode) (Sato, Sato, & Schmidt, 1980). The authors concluded that small-diameter Aδ fibres (group III) were activated in the 4-10$T_{\text{ENG}}$ range while C-fibres (group IV) were activated in the 100-200$T_{\text{ENG}}$ range. Accordingly, all bladder reflexes elicited by PTNS in our study ($6T_m$), very likely involved the electrical recruitment of small Aδ fibres, with very limited activation of unmyelinated C-fibers.

Interestingly, however, reflex excitation of the urinary bladder has been reported in spinally-intact anesthetized cats by applying ice to the hind paws for a duration of 2-5 minutes (McPherson, 1966). The resulting decrease in the interval between bladder contractions (increased bladder contraction frequency) was attributed to unmyelinated C-fibres (tonic nociceptors) that were activated by this low-temperature input. Assuming a similar bladder-excitatory reflex – that is mediated by high activation threshold C-fibers – also exists in rats, this particular reflex pathway may partially explain the diminished effectiveness of PTNS in inhibiting the bladder at stimulation amplitudes above $3T_m$ (Su et al., 2012a). Given that we limited the stimulation amplitude up to $6T_m$, it is difficult to comment on the further effects of higher PTNS amplitudes on reflex bladder inhibition in this study.

### 4.4 PTNS-Meditated Excitation of the Bladder

The results of this study also present evidence of a PTNS-evoked bladder excitatory reflex that is mediated by high-frequency (50 Hz) stimulation of myelinated afferent fibers. Based on the electrical recruitment properties of unmyelinated C-fibers in mammals (Sato et al., 1980; Yoo et al., 2013), it is highly unlikely that this reflex was mediated by the C-fiber mechanism reported by McPherson et al (McPherson, 1966). The frequency (50 Hz) at which this excitatory PTNS reflex is evoked is clearly higher than the typical frequency values (10-30Hz) used to evoke
bladder inhibition by PTNS (Su, Nickles, & Nelson, 2012b; Tai, Chen, Shen, Wang, Roppolo, et al., 2011). In fact, it is also higher than the broad range of frequencies (2-35 Hz) required to elicit stimulation-evoked bladder contractions, whether mediated by the pudendal nerve trunk (Boggs, Wenzel, Gustafson, & Grill, 2006b), the dorsal genital nerve (Woock et al., 2008), or the urethral sensory branch (Yoo et al., 2008).

The novel observation of this high-frequency (50 Hz) excitatory bladder reflex in rats may be due, in part, to the experimental setup. Unlike previous PTNS studies that tested the effects of PTNS under isovolumetric conditions (G. Chen et al., 2012; Mally et al., 2012; Schwen et al., 2013a, 2013b; Su et al., 2012a; Tai, Chen, Shen, Wang, Liu, et al., 2011; Tai, Chen, Shen, Wang, Roppolo, et al., 2011; Tai, Shen, Chen, Wang, Liu, et al., 2011; Tai, Shen, Chen, Wang, Roppolo, et al., 2011), our study involved a continuous bladder infusion (i.e. urodynamic) method. Continuous and/or periodic neural feedback of lower urinary tract function was provided by pelvic and pudendal afferents during each repeated bladder filling and voiding. We speculate that pudendal afferent feedback played a significant role in unmasking this otherwise quiescent 50 Hz excitatory reflex. This hypothesis is supported by previous work of Peng et al (Peng, Chen, Cheng, & Grill, 2008), where they showed significant reduction of bladder voiding efficiency in anesthetized rats following unilateral and bilateral transection of the sensory branches of the pudendal nerve. (Peng et al., 2008). Anatomically, a potential interaction between PTN and pudendal nerve afferents is suggested by Pacheco et al (Pacheco et al., 1997; Pacheco, Martinez-Gomez, Whipple, Beyer, & Komisaruk, 1989), where they showed that the lumbosacral trunk nerve (LSN) and the L₆-S₁ nerve are common to both the pudendal (primarily L₆-S₁) and sciatic (primarily LSN) nerves. More specifically, in addition to the LSN being made up of contributions from L₄-L₆ and therefore sharing the L₆ contribution with the L₆-S₁ nerve more distally, the LSN also contributes a branch to the pudendal nerve and its corresponding sensory branch before continuing on to become the sciatic nerve and eventually the tibial nerve. This provides evidence for an anatomical mechanism through which the pudendal afferent nerves can converge and interact with the stimulus arising from the PTN or its distal branches. Further work is needed to investigate the interaction between pudendal afferents and PTNS-evoked
bladder reflexes and its potential application for addressing clinical symptoms of detrusor underactivity (Taylor & Kuchel, 2006).

4.5 Specific PTN Afferents Independently Modulate Bladder Function

Compared to the frequency-dependent characteristics of PTNS (nerve trunk) in the rat (Figure 4), selective electrical activation of each individual PTN branch exhibited markedly different patterns of bladder modulation. Medial plantar nerve stimulation resulted in both robust prolonged (5 Hz and 10 Hz) and acute (10 Hz) bladder inhibition, while high-frequency (50 Hz) stimulation failed to show any evidence of a bladder-excitative reflex. Similarly, lateral plantar nerve stimulation exhibited both prolonged and acute bladder inhibition, but at higher stimulation frequencies (10 Hz and 20 Hz). This data also showed that the high frequency (50 Hz) bladder-excitative reflex is mediated by the lateral plantar nerve. It is interesting to note that while (whole nerve) PTNS failed to elicit post-stimulus bladder inhibition at 10Hz, we clearly observed this prolonged reflex in response to 10 Hz stimulation of either the medial or lateral plantar nerve. In analogous fashion, lateral plantar nerve stimulation at 20Hz resulted in robust bladder inhibition, but the same input failed to elicit any effect when delivered via the medial plantar nerve or PTN trunk. These comparisons underscore a complex pattern of interaction among various sensory inputs that (as mentioned above) may also include the pudendal sensory nerve afferents. More importantly, these findings could have significant clinical implications for optimizing PTNS therapy, which is currently delivered primarily by 20 Hz stimulation. Further work is needed to clarify the neural mechanisms that underlie these multiple reflex pathways.

4.6 Clinical Implications

The results of this study – in addition to work by other researchers – suggest that the current PTNS protocol used to treat OAB symptoms may be enhanced by modifying the stimulation parameters. This is similar to that achieved by InterStim Therapy, where sacral nerve stimulation is adjusted over a broad frequency range (10-20Hz) and amplitudes to optimize the suppression of urinary incontinence (Peeren et al., 2005). Given both the current evidence of a post-stimulus bladder-inhibitory reflex (prolonged bladder inhibition lasting up to 0.5 hr) and prior reports of
similar prolonged inhibition (up to 2 hr) in cats (G. Chen et al., 2012; Tai, Shen, Chen, Wang, Roppolo, et al., 2011) and rats (Matsuta et al., 2014), it appears that 5 Hz or 10 Hz may be potentially effective stimulation parameters for PTNS therapy.

However, all pre-clinical data – including this study – indicate that robust neuromodulation of the urinary bladder requires the stimulation amplitude to exceed the foot motor threshold, which is the current ‘target amplitude’ used in clinical PTNS therapy. The physiological evidence clearly suggests that improved control of lower urinary tract function requires the electrical activation of small diameter myelinated axons, which in rats is achieved by PTNS at $6T_m$. Further research is needed to determine whether this therapeutic limitation can be circumvented by lower stimulation frequencies, which may provide a more tolerable sensation to patients undergoing PTNS therapy (McGrath, Gracely, Dubner, & Heft, 1983). In addition, further clinical translational studies are needed to elucidate the potential therapeutic effects of selective electrical activation of individual branches of the posterior tibial nerve (e.g., medial plantar and lateral plantar nerves).
Chapter 5
Conclusions & Future Work

In order to improve patient outcome in the treatment of overactive bladder through PTNS therapy, it is important to understand the underlying mechanism. Once the mechanism is fully understood, steps can be taken to optimize the therapy and improve patient outcomes.

With this in mind, the work completed in this thesis is of dual importance. Firstly, the study improves our current understanding of PTNS by providing evidence for which diameter nerve fibres might be responsible for therapeutic effectiveness and by showing for the first time that the distal branches of the PTN might play separate roles in the overall PTNS mechanism. Secondly, we provide a rationale for specific alteration of stimulation parameters as a means to potentially optimize the therapy, and ultimately improve response rates. As an example, the study suggests both 5Hz and 10Hz as potential candidates for alternate PTNS stimulation parameters.

Finally, the 50Hz bladder excitatory mechanism which was uncovered during the experimental work has applications beyond overactive bladder. Possible applications of this new phenomenon are likely found within patient cohorts dealing with urinary retention or underactive bladder.

5.1 Developing the Animal Model

Although the current animal model has been sufficient to show the overall frequency tuning characteristics of the PTNS bladder inhibition phenomenon, there are some downfalls to the stimulation protocol. For example, it cannot be confirmed that the inhibition caused by stimulation at one frequency wears off completely during the 10 min post-stimulation period before stimulation at the next frequency is conducted. The selection of the current stimulation protocol is rooted in exploring all frequencies in a reasonable amount of time while using minimal number of animals. As a next step, it would be suggested to explore individual frequencies in single animals.
Additionally, the delivery of urethane anesthesia to the animals often had an effect on the frequency of bladder contractions. Future work should focus on finding solutions for delivery of a consistent dose of urethane anesthesia, either through continuous IP infusion or other means. This would help increase the reproducibility of results.

5.2 Exploring Clinical Implications

The findings of this study will only make a real impact on the relevant patient cohorts when they make their way to a clinical setting. The logical progression from the work done in this thesis is to explore the 5Hz and 10Hz acute and post-stimulate effects in the form of a clinical trial. A suggested initial trial would be to study three groups of patients undergoing a typical PTNS protocol under either 5Hz, 10Hz or the classic 20Hz stimulation and comparing the patient outcomes through urodynamic tests as well as pre and post-treatment quality of life surveys.

It is also possible that different patients respond to PTNS applied at different stimulation frequencies. As such, it might be useful for the non-responders from the aforementioned clinical trials to undergo a second PTNS trial at an alternate frequency. Similarly, patients who have undergone previous clinical trials with 20Hz PTNS but did not see a response, should be asked to undergo PTNS at 10Hz or 5Hz. As the PTNS effect has already been verified clinically, the next wave of clinical trials should focus optimization of treatment and customization for individual patients.

The results of this study suggest that the most consistent PTNS bladder inhibition was achieved during stimulation of the medial plantar branch. Similarly, there was a very consistent post-stimulation effect after stimulation was applied to the lateral plantar branch. Therefore, it would be useful to explore alternate PTNS treatment approaches. One inexpensive clinical trial would involve transcutaneous stimulation of the medial plantar branch at the hallux (big toe). Likewise, the lateral branch can be accessed transcutaneously through the lateral digits of the foot. Clinicians might also be inclined to apply percutaneous PTNS to the medial and lateral plantar branches of the foot.
5.3 The Excitatory Phenomenon

The 50Hz excitatory phenomenon observed in this study has never been before shown in an animal model. Although the results were not very consistent, it is important to note that this finding was unexpected. Naturally, the experimental paradigm used (i.e. continuous filling and emptying of the bladder throughout the experiment) was not optimized to investigate an excitatory response on the bladder. Future investigation of this excitatory reflex should involve a different experimental approach. An isovolumetric study, where the bladder is maintained at a constant volume, would likely be a better proving ground for this effect, ultimately yielding more consistent results.

In any case, as far the clinical application of 50Hz PTNS goes, its first use can come in the form of urinary retention or underactive bladder patients. A clinical trial can be set-up where these patients undergo the classic PTNS therapy protocol (one stimulation session, once a week over twelve weeks) but at a frequency of 50Hz. Success with this trial would provide a novel method for treating patients with these clinical indications.

5.4 Other Relevant Work

The work described in this thesis deals with experiments attempting to optimize PTNS therapy by the varying stimulation parameters (frequency and amplitude) and target nerves (medial and lateral plantar branch). Over the course of the last two years, I have also conducted work dealing with the optimization of the PTNS by other means—most notably, the use of an implantable device which enhances transcutaneous PTNS. These studies were first validated using finite element modelling (i.e. computer simulations) and then proven in an animal model (rat model). Although experimental procedures were done in conjunction with the work described in this thesis, we felt that this secondary study was both beyond the scope of the thesis, has therefore been omitted from the main body of the thesis.
References


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