Estimated Distributions of Thermal and Vibration Sensory Thresholds for Healthy Subjects

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Preface

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This thesis document includes the text of one article to be submitted for publication to the journal of the American Association of Electrodiagnostic Medicine, *Muscle & Nerve*. The authors, listed in order, are: Gillian Bartlett, John D. Stewart, Robyn Tamblyn, Michal Abrahamowicz. The article presents the methods and results of a primary analysis of data collected by Gillian Bartlett as part of an original study of thermal and vibration sensory thresholds. Dr. M. Abrahamowicz acted as primary thesis supervisor, Dr. J. Stewart as co-supervisor and Dr. R. Tamblyn as thesis committee member. The research presented in the paper was inspired by the work of Drs. Abrahamowicz and Stewart on establishing an index measure of autonomic cardiovascular reflexes. Drs. Abrahamowicz, Stewart and Tamblyn reviewed drafts of the article prior to final submission for publication. Gillian Bartlett, MSc candidate, was responsible for determining the research question; establishing appropriate measurement procedures and designing the study; recruiting subjects; collecting all data; carrying out the statistical analysis; interpreting, organizing, and presenting the results; and writing the manuscript.
Abstract

Objective: To estimate the distributions of thermal and vibration sensory thresholds in a healthy population taking into account important covariates.

Methods: Sensory thresholds were measured using quantitative sensory testing for 148 volunteers free of neuropathic disease. Independent effects of age, gender, height, and skin temperature were estimated using multiple linear regression. Parametric and nonparametric methods were used to estimate the distributions of interest.

Results: Significant age-related increases were observed for vibration thresholds (p<0.0001) and in the foot for thermal thresholds (p<0.0002). Height was significantly associated with vibration thresholds in the foot (p<0.003). Selected percentiles are estimated for thermal trends in the hand. Age-adjusted continuous distributions for the remaining thresholds are presented with corrections for height where appropriate.

Conclusions: Our results provide reference values for thermal and vibration sensory thresholds of a healthy population. Future patient test results may be compared with estimated age-specific percentiles to assess levels of abnormality.
Résumé

**But:** Évaluer les distributions des seuils de sensibilité thermique et vibratoire dans une population en bonne santé, en tenant compte des covariables importantes.

**Méthodes:** Les seuils de sensibilité ont été mesurés par des essais sensoriels quantitatifs sur 148 sujets volontaires ne présentant aucunes neuropathies. Les effets indépendants de l'âge, du sexe, de la taille et de la température cutanée ont été estimés par une analyse par régression linéaire à variables multiples. Des tests paramétriques et non paramétriques ont été utilisés pour estimer les distributions pertinentes.

**Résultats:** Des augmentations significatives reliées à l'âge ont été observées pour les valeurs des seuils de sensibilité vibratoire (p<0.0001) et, dans le pied, pour les seuils de sensibilité thermique (p<0.0002). Une relation significative a été observée entre la taille des sujets et les seuils de sensibilité vibratoire dans le pied (p<0.003). Certains percentiles ont été estimés pour les tendances de la sensibilité thermique dans la main. Les distributions continues corrigées en fonction de l'âge concernant les autres seuils sont décrites en tenant compte des corrections en fonction de la taille des sujets, lorsqu'il est approprié.

**Conclusions:** Les résultats de notre étude fournissent des valeurs de référence pour les seuils de sensibilité thermique et vibratoire dans une population en bonne santé. Les résultats des tests des patients pourront être évalués en regard des percentiles spécifiques de l'âge estimés dans cette étude, ce qui permettra d'évaluer le degré d'anomalité.
Statement of Originality

This thesis is composed of two parts. The first is a review of the literature on quantitative sensory testing of peripheral nerve fibers. Although a great deal of research has been conducted in this field, there is a lack of adequate reference norms for a healthy population. Based on a review of the literature, I identified an appropriate methodology for establishing reference norms for quantitative sensory testing. To my knowledge, this methodology has not been used for testing of peripheral nerve function in a healthy population.

The second part of the thesis consists of an original study undertaken to establish reference norms for quantitative sensory testing of thermal and vibration sensory thresholds. I recruited and tested 148 healthy subjects in order to estimate the distributions of sensory thresholds while controlling for important covariates. Emphasis was placed on ensuring a uniform distribution of age and gender, with particular attention to older subjects. Nonparametric statistical methods were used in order to avoid bias due to a priori assumptions regarding the distributions' shape. To my knowledge, these statistical methods have not previously been used in the published literature in the field of quantitative sensory testing.
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First and foremost, I would like to thank my thesis supervisor, Dr. Michal Abrahamowicz, who has acted as a mentor and inspiration for my work in this field. The successful completion of this thesis was due in large part to his patience, encouragement, dedicated teaching, availability and valuable input during the entire MSc training program. I would also like to thank my co-supervisor, Dr. John Stewart, who has always made room for me in his busy schedule and contributed a great deal of his time, energy and resources to help me complete my thesis research project. Without the support and advice of Dr. Stewart, this study would not have been possible. Last, but certainly not least, I would like to thank Dr. Robyn Tamblyn for acting as a member of my thesis committee. Her interest, time and advice was very beneficial and greatly appreciated.

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CHAPTER 1. INTRODUCTION

Background

The human nervous system is a complex organization of structures. Information from internal and external environments is interpreted and integrated by the nervous system. A response to this information is then initiated. For descriptive purposes, the nervous system is categorized into the central and the peripheral nervous systems. The former consists of the brain and spinal cord while the latter includes the peripheral nerves and ganglia (Watson, 1991).

Peripheral nerve fibers innervating human skin are responsible for conveying cutaneous sensations. These primary afferent nerve fibers range in size, conduction velocity, and may or may not be ensheathed in myelin. The type of sensory modality conveyed depends upon the characteristics of the peripheral nerve mentioned above (Dyck, Thomas, Griffin, Low, & Poduslo, 1993, chap. 8). Peripheral nerve fiber impairment, referred to as peripheral neuropathy, can have serious and debilitating consequences due to reduced or amplified responses to sensory stimuli. Common manifestations of peripheral neuropathy can be roughly divided into two groups. Negative sensory symptoms are loss of temperature sensation; loss of vibration, touch-pressure and joint position sense (e.g., numbness); and muscle weakness. Positive sensory symptoms are paresthesia, painful sensations, and excessive response to natural stimuli: hyperalgesia, allodynia, or dysesthesia (Brown & Asbury, 1984; Dyck, Thomas, et al, 1993, chap. 39; Vinik, Holland, Le Beau, Liuzzi, Stansberry & Colen, 1992). Patients may be unaware of
reduced pain and temperature sensitivity that can lead to the development of neuropathic ulcers: plantar and other foot ulcers due to recurrent trauma; and/or Charcot's arthropathy (Brown & Asbury, 1984; Guy, Clark, Malcolm & Watkins, 1985; Vinik et al, 1992).

The impact of peripheral neuropathy in the general population is hard to estimate (Dyck, Thomas, et al, 1993, chap. 40). Assessment of neurological function is difficult, time consuming and often relies heavily on patient report of symptoms. Despite these methodological challenges, a great deal of progress has been made with epidemiological studies aimed at understanding etiology and identifying risk factors for peripheral neuropathy. Impairment of peripheral nerves is seen in patients with infective, inflammatory and immune diseases; systemic diseases; and cancer (Dyck, Thomas, et al, 1993, chaps. 64 - 83; Lipton et al, 1991). The Rochester Diabetic Neuropathy Study estimated the cumulative incidence for neuropathy in diabetic patients to be 20% after 20 years of follow-up (Dyck, Kratz et al, 1993). In another study, Lindblom and Tegnér (1985) estimated that 30% of patients with chronic renal failure suffer from small fiber neuropathy. Other research has shown that peripheral neuropathy may be inherited or caused by ischemia and physical agents. Industrial agents, metals, drugs and neoplasm are also known to associated with the disease (Dyck, Thomas, et al, 1993, chaps. 84 - 91). In order to increase our understanding of the impact and cost attributable to peripheral neuropathy, measurement and methodological issues must be addressed.

Peripheral nerve function is traditionally evaluated in nerve conduction studies that rely on measures of nerves' ability to conduct electrical impulses (Gerr, Letz, Hershman, Farraye & Simpson, 1991). However, nerve conduction studies only estimate the function of the
largest myelinated sensory and motor nerve fibers. These nerve fibers account for only 25 - 33% of the nerve fibers in the peripheral nerve (Jacobs & Love, 1985; Ochoa & Mair, 1969). Since nerve conduction studies are unable to measure total peripheral nerve function, they have limited diagnostic value in assessing disease affecting smaller fibers. This has practical relevance since many peripheral neuropathy patients, the most common being diabetics, have predominant or selective damage to smaller fibers (Brown & Asbury, 1984; Guy et al. 1985; Said, Goulon-Goeau, Slama & Tchobroutsky, 1992). Studies of these fibers have shown that cutaneous thermal sensations of cooling are conveyed in the small, slow, thinly myelinated fibers and sensations of warming in the small, very slow, unmyelinated fibers. Sensations of vibration are conveyed in large, fast, myelinated fibers (Lindblom, 1980; Pertovaara & Kojo, 1985). Thus testing the ability of patients to detect sensations of warmth, coolness and vibration helps assess the function of several types of fibers in the peripheral nerve.

Currently accepted methods of testing sensations of warmth, cooling and vibration consist of psychophysical techniques generally referred to as quantitative sensory testing (QST). A consensus report from the Peripheral Neuropathy Association defines QST as "techniques used to measure the intensity of stimuli needed to produce specific sensory perceptions" (Peripheral Neuropathy Association, 1993). Applications of QST have become important for many reasons.

First, there is evidence that sensory impairment progresses at different rates for different patients (Sosensko, Kato, Soto & Bild, 1992). The level of impairment may place patients in higher risk categories for serious outcomes such as foot ulceration leading to amputation.
(Vinik et al. 1992; Young, Breddy, Veves & Boulton, 1994). Quantifying the level of sensory dysfunction allows the progression of the impairment to be monitored over time and alert clinicians to new risk categories (Griffiths & Wieman, 1992; Hilz, Claus & Neundörfer, 1988). Secondly, QST results may be used to verify the efficacy of any new treatments developed for peripheral neuropathy. Finally, the tests may be used to implement screening programs for high risk populations. This would be particularly useful for populations exposed to neurotoxic compounds (Bleecker, 1985).

There are several aspects of QST methodology which have helped to make it a popular clinical and research tool. Mainly, the quantitative tests are very sensitive, acceptably reproducible and noninvasive (American Diabetes Association, 1988). These characteristics play an important role in monitoring sensory function. The other advantages of QST, compared to classical nerve conduction studies, are the portability of the testing equipment, the quickness of the testing procedure, and the lack of pain normally associated with the tests.

Although QST is now recognized as an established methodology in human and animal research, several issues remain unresolved. The report from the Peripheral Neuropathy Association (1993) states that in order for a specific sensory test to be developed and validated several requirements must be met. The criteria for a valid QST include: a well defined stimulus suitable to the type of sensation being tested, a stimulus that does not vary over a range of magnitudes, and a stimulus that is quantifiable. Also, The algorithm used to present the stimulus and the method used to estimate the threshold of sensation must be validated on healthy and diseased populations. Furthermore, the results of the testing must
be compared to a group of healthy subjects in order to establish abnormality criteria according to modality, site, age, gender, and other possible covariates (Gerr & Letz, 1993; Gruener & Dyck, 1994; Peripheral Neuropathy Association, 1993).

In this thesis, I attempt to meet these requirements in order to establish normal limits for QST of vibration and thermal sensation.

Objectives

The primary objective of the study contained within this thesis is to estimate the distributions of vibration and sensory thresholds in healthy subjects using QST. The objective of the literature review is to present an assessment of the different methodologies associated with QST and currently used to measure sensory thresholds. Based on this review of the literature, I intend to construct an appropriate methodology for assessing vibration and thermal sensory thresholds and will include covariates that may independently influence threshold levels.

These distributions will be made available to clinicians and researchers to assess the probability of normality for thermal and vibration sensory test results.
CHAPTER 2: LITERATURE REVIEW

Testing Algorithms for Thermal and Vibration Sensory Thresholds

A large part of the research in the field of QST has focused on the evaluation of testing algorithms for establishing thermal and vibration sensory thresholds. Several types of machines are available to perform the testing and most equipment currently uses well defined stimuli that are quantifiable and allow unlimited repetitions. Although accurate and appropriate equipment considerably reduces measurement error, testing algorithms still have a major impact on threshold values (Dyck, Karnes, Gillen, O’Brien, Zimmerman & Johnson, 1990; Jamal, Hansen, Weir & Ballantyne, 1985; Yarnitsky & Ochoa, 1991).

Among several methods employed for measuring sensory thresholds, three have received the most attention: the method of limits, the method of levels and the two-alternative forced choice method. In what follows, I will discuss each of these methods in more detail.

The method of limits is one of the most widely used but most criticized testing algorithm for determining thresholds with QST. This technique consists of presenting a subject with a stimulus which is either gradually increasing or decreasing in intensity. The subjects are asked to flip a switch when they detect the stimulus. At this point the stimulus direction is reversed until the baseline level of intensity is attained. Thresholds are usually determined as a mean of the “first detected” stimuli from several successive stimuli presentations.

One of the main attractions of the method of limits is the relatively short time it takes to test subjects. Studies comparing the method of limits with the two-alternative forced choice
estimate that the latter takes twice as long for testing vibration (Gerr & Letz, 1988) and almost six times as long for testing thermal sensation (Claus, Hilz & Neundörfer, 1990). Proponents of the method of limits argue that the quickness of the testing improves subject cooperation and concentration: key factors in any type of psychophysical testing. Unfortunately, there are several methodological drawbacks that outweigh these perceived benefits. First of all, the subjects press a switch to alter the stimulus the moment they detect the sensation; therefore, differences in individual reaction times may confound comparisons of observed threshold values (Claus et al. 1990; Fagius & Wahren, 1981; Yarnitsky & Ochoa, 1991; Yarnitsky & Sprecher, 1994). Since attentional factors play a large role in reaction time, there may be a substantial amount of within subject variance resulting in artificially high values for some subjects’ thresholds (Fruhstorfer, Lindblom & Schmidt, 1976). A second concern is that the threshold values may vary with rate of stimulus change. A rapid rate of stimulus increase usually results in overestimating thresholds (Dyck, O’Brien, Kosanke, Gillen & Karnes, 1993; Dyck, Zimmerman, Gillen, Johnson, Karnes & O’Brien, 1993). Adjusting the method of limits in order to eliminate these problems would result in lengthy and cumbersome test procedures, thereby eliminating the time advantage (Dyck, Zimmerman et al., 1993).

In order to address the problems associated with the method of limits, the two-alternative forced choice testing algorithm was developed (Hansen, Jamal, Weir, Ballantyne & Bissessar, 1987). The exact methodology depends on the set-up of equipment used for testing. Usually, two identical plates or posts are available to the subject. The subject is asked to make contact with each plate and choose the one that she/he believes to contain the stimulus (Halar, Hammond, LaCava, Camann & Ward, 1987). The stimulus is present in
only one plate each the time the subject is asked to make a choice. Stimulus intensity is changed in discrete steps and the direction of the step is dependent on the accuracy of the subject's response. The thresholds are calculated as a mean of the stimulus levels at which errors are made and the levels at which the lowest correct responses are made. This method can also be applied to equipment with only one stimulus surface. In this case, the subject is continually in contact with the stimulating surface and is asked to choose between two time periods. The stimulus is present during one of the two periods. The change in stimulus intensity and calculation of thresholds follows the paradigm described above (Jamal, Hansen, Weir & Ballantyne, 1985). While the forced choice method eliminates confounding by reaction time and has acceptable reproducibility (Gerr & Letz, 1988), as indicated previously, it is a lengthier procedure. Due to the time disadvantage, many researchers and clinicians still opt to employ the method of limits (Bove, Letz & Baker, 1989; Claus, Mustafa, Vogel, Herz & Neundörfer, 1993; Muijser, Hooisma, Hoogendijk & Twisk, 1986).

The rate of change for the stimulus intensity significantly influences both the method of limits and the forced choice testing algorithm, especially when assessing thermal thresholds (Fruhstorfer et al., 1976; Pertovaara & Kojo, 1985; Yarnitsky & Ochoa, 1991). To address this issue, Yarnitsky and Ochoa (1991) recently developed the method of levels as a variant of the forced choice algorithm for testing thermal thresholds. Rather than asking subjects to choose which of the two plates or times contain the stimulus, participants are presented and asked each time whether they detect it. Testing is continued at graded steps of increasing stimulus magnitude from a baseline temperature to the point at which the stimulus is felt. Then the magnitude of the stimulus is decreased in a stepwise
fashion until it is not felt. This is repeated about three times. The value midway between the highest negative response and the lowest positive response is taken as the threshold value. The method of levels has the advantage of being highly reproducible without taking anymore time to administer than the method of limits (Yarnitsky and Sprecher, 1994).

In a study comparing different testing algorithms, Yarnitsky and Ochoa (1991) found the method of levels provided lower estimates of thermal thresholds than those obtained by the method of limits. A comparison of threshold values attained with the method of levels for three different rates of stimulus change showed no difference in observed thresholds for each rate (Yarnitsky & Ochoa, 1991). A similar analysis performed by these authors for the method of limits demonstrated a significant association between threshold values and rate of temperature change. Although there is no published data available for the measurement of vibration thresholds using the method of levels, the equipment has been modified to test vibration thresholds using the same technique used for thermal thresholds (Medoc Ltd., Ramat Yishai, Israel).

Based on the findings from published literature, the method of levels appears to be the preferred choice for testing thermal thresholds. However, until initial studies have been complete comparing the performance of this method for measuring vibration thresholds with other validated techniques, I would be reluctant to estimate thresholds using the method of levels. For this reason the two-alternative forced choice method would be preferred for testing vibration thresholds. Although this technique is more time consuming than the method of limits, avoiding the issues of individual differences in reaction time more than compensates for this limitation.
After determining an appropriate test algorithm, it is important to adjust for key covariates. Several studies have been published studying about factors that influence observed sensory thresholds. Since the studies use different methodology, conclusions about the influence of the relevant variables are often difficult make. This section will review the available evidence and attempt to identify variables that should be taken into account when establishing reference values from a healthy population (Peripheral Neuropathy Association, 1993).

Studies of the effect of age on vibration thresholds have consistently shown a deterioration of sensation with age (Aaserud, Juntunen & Matikainen, 1990; Era, Jokela, Suominen & Heikkinen, 1986; Gerr, Hershman & Letz, 1990; Goldberg & Lindblom, 1979; Nico, de Neeling, Bek, Bertelmann, Heine & Bouter, 1994; Wiles, Pearce, Rice & Mitchell, 1991). The increase in vibration thresholds with age is particularly pronounced in persons over 70 years old (Thomson, Masson & Boulton, 1992).

The evidence for the relationship between age and thermal thresholds is not as clear. Merchut and Toleikis (1990), used the two-alternative forced choice method to estimate thermal thresholds in 54 male and female subjects. The investigators found no significant relationship between age and thermal thresholds, however, the statistical power of the findings were limited since only 16 subjects were in the age range 60 - 80 years. Using the same QST method, Arezzo, Schaumburg, and Laudadio (1986) found an apparent increase in thresholds and variance with age but the trends were not significant for 100 normal
subjects, aged 18 - 65, who were tested on the hand and foot. An earlier study conducted by Bertelsmann, Heimans, Weber, van der Veen, and Schouten (1985) using the same methodology with 36 subjects aged 24 - 91 years, found an association for age and thermal thresholds in the foot. In two more recent studies, using similar testing algorithms, a rise in thermal thresholds with age was found in 71 subjects aged 21 - 92 years (Doeland et al. 1989) and in 216 subjects aged 50 - 76 years (Nico et al. 1994). Using a technique similar to the method of levels, Fowler, Carroll, Burns, Howe, and Robinson (1987), found a positive linear effect of age on thermal threshold in 209 subjects (mean age of 39.6). Based on the findings for vibration thresholds and the more recent findings for thermal thresholds, age must be considered as a major covariate for sensation thresholds.

In studies where height was measured, it was associated with an increase in threshold values in the lower limbs (Gerr & Letz, 1994; Nico et al., 1994). This finding was more common for vibration thresholds than for thermal thresholds (Era et al., 1986; Wiles et al., 1991).

Results concerning the effect of gender on sensation thresholds seem to be confounded by effects of height. Using a modified form of the method of limits, Wiles et al. (1991) estimated vibration thresholds for 1,365 subjects aged 10 - 91 years old. In this random sample of subjects, the investigators adjusted for height and found no overall effect for gender except on the ankle, where females had lower threshold values. Nico et al. (1994), in a study of 216 randomly sampled subjects, found that women had significantly higher vibration thresholds but concluded that this finding may have been confounded by height.
The investigators used the method of limits for the vibration thresholds and the two-alternative forced choice to measure thermal thresholds.

The same pattern of results exists for the measurement of thermal thresholds. Doeland et al (1989) estimated thermal thresholds with the two-alternative forced choice method in 71 healthy subjects. Female subjects had significantly lower thresholds when no adjustment was made for height differences. Lautenbacher and Strian (1993) examined a small sample of 20 undergraduate students using the method of limits, and found no differences between men and women after assessing effects of height. Lautenbacher and Rollman (1993) summed up the issue very neatly when they stated, “the notion that women are more responsive than men can be replaced by the notion that small people are more responsive than large individuals.” (p. 256). In order to ascertain whether women are more sensitive than men, height must be taken into account when measuring sensory thresholds.

Evidence regarding the influence of skin temperature is also not clear. Kojo and Pertovaara (1987) found an elevation in warm thresholds with an increase in adaptation skin temperature for six volunteers. The Vietnam Experience Study (Gerr & Letz, 1994) also found skin temperature to be a significant covariate for thermal thresholds. No effect of skin temperature on vibration thresholds has been found (Gerr & Letz, 1994; Gerr et al, 1991). The norm for QST is to maintain the thermode base temperature between 32 - 35°C (Lautenbacher & Strian, 1993).
Several other potential covariates for which no marked impact on sensation thresholds was found include: time of day (Strian, Lautenbacher, Galfe & Hölzl, 1989), caffeine, sweating, exaggerated local vasodilatation and skin hyperemization (Hilz, Claus, Balk & Neundörfer, 1992), smoking (Wiles et al. 1991), and alcohol consumption in nonalcoholics (Gerr & Letz, 1994). The Vietnam Experience study of 4,462 young men, found income and race to be significantly correlated with thermal thresholds estimated using the two-alternative forced choice method (Gerr & Letz, 1994). Despite the large sample base, it is difficult to imagine what mechanism may be responsible for these effects. The results may have been confounded by understanding of the testing protocol. These findings are refuted by another large study of randomly sampled men in Finland that were measured using the method of limits. The investigators found that education levels, income and living habits had no effect on thresholds of men aged 31 - 35, 51 - 55 and 71 - 75 (Era et al. 1986). Furthermore, the thresholds do not appear to show a decrease over time due to learning effects (pilot study: Aaserud et al, 1990; Salle & Verberk, 1984).

Therefore, the covariates of potential interest include age, height, gender and skin temperature. Investigation of these covariates will allow for more powerful statistical comparisons and greater accuracy for threshold estimates (Gerr & Letz, 1993). In addition to statistical significance, the clinical importance of the covariate effect should be taken into account, especially given the limited precision of measurement instruments.
Review of Published Normative Data

Adequate reference values from healthy subjects are essential for the evaluation of patients (Peripheral Neuropathy Association, 1993). Also, the sensitivity and specificity of thermal and vibration thresholds cannot be evaluated without an appropriate reference group (Feinstein, 1985). Appropriate reference values and information about the diagnostic capabilities of the tests will allow researchers and clinicians to effectively utilize vibration and thermal thresholds as diagnostic tools.

As summarized by O'Brien and Dyck (1995) "the quality of the available reference values are far from ideal, mainly because of unrepresentative selection of healthy subjects, inadequate sample size, lack of medical evaluation of subjects, failure to also measure other variables that influence the attribute studied, and failure to rigorously apply appropriate statistical analysis." (p.17) This is true of most published research on sensory thresholds to date. The two studies with the largest sample sizes, namely the Vietnam study and the Finland study, failed to include women in their sample (Gerr & Letz, 1994; Era et al. 1986). The Vietnam study included young healthy men of a limited age range and the authors caution readers about the generalizability of their results. The study of randomly sampled men in Finland studied three specific age categories: 31 - 35, 51 - 55 and 71 - 75 making it difficult to interpolate for intermediate age groups and to draw conclusions about the continuous effect of age (Era et al, 1986).

Yarnitsky and Sprecher (1994) recently attempted to establish solid reference values for thermal thresholds using several different test algorithms, including the method of levels,
in a sample of 106 healthy volunteers. Although the authors accounted for skin temperature, gender, and age, no attempt was made to measure height. The authors also stratified their sample by age using arbitrary categories. Statistical analyses were done within each stratum. This effectively limited sample size creating unstable estimates of normative values. The numerical instability of the estimated normal limits is demonstrated by the nonmonotone relationship of the limits with age. For example, estimates of cold thresholds on the hand of females decreased for 40 - 59 year olds compared to 20 - 39 years, then increased again for the 60 - 79 year olds compared to the 40 - 59 group. The reverse of the effect observed in females is seen for cold testing on the foot among males.

Bravenboer, van Dam, Hop, v.d. Steenhoven, and Erkelens (1992) also attempted to establish normal values for thermal thresholds using a forced choice algorithm. Testing 40 females and 40 males, the authors again used arbitrary age categories. For cold threshold in the hand, the observed values show a nonsignificant decrease for the 35 - 44 year olds compared to 25 - 34 year olds. The authors gave no explanation for why the older group may be more sensitive that the younger group. Differences between groups were evaluated using independent-groups Student's t-test with no correction of the significance levels for the multiple testing. Also, many potentially important covariates were not measured.

This review of the methodology and results of the most current attempts to establish normal values for thermal and vibration thresholds confirm O'Brien and Dyck's (1995) conclusions on the state of published norms. The design of the study has taken into account the criticisms of the published research on reference values.
Conclusion

In order to avoid some of the drawbacks identified above, I have ensured an adequate distribution of age with particular emphasis on the older groups. Other covariates which have been identified as potentially important have been measured and their effects evaluated from a statistical and clinical perspective. Only the test procedures that have been well standardized and identified in this review have been employed. Statistical methods were flexible and not dependent on assumptions regarding the distribution of the observed thresholds, in order to enhance the quality of reference values.

The article in Chapter 3 describes research conducted in order to establish reference values for thermal and vibration thresholds in a healthy population.
CHAPTER 3: Manuscript of article to be submitted for publication

ESTIMATED DISTRIBUTIONS OF THERMAL AND VIBRATION SENSORY THRESHOLDS

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Abstract

Quantitative sensory testing was used to measure thermal and vibration thresholds in the hand and foot of 148 healthy subjects. Age was uniformly distributed between 20 and 86 years. Independent effects of age, gender, height, and skin temperature were estimated using multiple linear regression. Parametric and nonparametric methods were used to estimate the distributions of interest. Significant age-related increases were observed for vibration thresholds ($p<0.0001$) and in the foot for thermal thresholds ($p<0.0002$). Height was significantly associated with vibration thresholds in the foot ($p<0.003$). Selected percentiles are estimated for thermal trends in the hand. Age-adjusted continuous distributions for the remaining thresholds are presented with corrections for height where appropriate. Our results provide reference values for thermal and vibration sensory thresholds in a healthy population. Future patient test results may be compared to estimated age-specific percentiles in order to assess levels of abnormality.

Key words: quantitative sensory testing, thermal sensation, vibration sensation, normal subjects, age
Introduction

For a number of well-described reasons, quantitative sensory testing (QST) is emerging as a practical measure of nerve fiber damage in patients with peripheral neuropathies.\textsuperscript{1-26} In particular, QST of the ability of patients to detect sensations of warmth, cold and vibration is useful for evaluating the function of a wide spectrum of fibers in the peripheral nerve.\textsuperscript{27,28} In spite of this, use of QST is limited by the lack of appropriate normative data.\textsuperscript{29}

In order to establish reference values for a healthy population, variables that may influence results from QST must be taken into account.\textsuperscript{30} Due to differences in methodology and study samples, there is a lack of consensus on key covariates. Age seems to have the biggest impact on thresholds. Studies of vibration thresholds have shown a deterioration of sensation with age,\textsuperscript{1,31-35} an effect that is particularly pronounced in persons over 70 years old.\textsuperscript{36} A somewhat weaker but similar effect has been seen for thermal thresholds.\textsuperscript{4,9,34,37-39} Frequently, analysis are stratified by gender, but the effect of gender on sensation thresholds may confounded by height differences.\textsuperscript{40} In studies where height was measured, it was associated with an increase in threshold values in the lower limbs.\textsuperscript{34,41} This was more common for vibration thresholds than for thermal thresholds.\textsuperscript{31,35} No effect of skin temperature on vibration thresholds has been found,\textsuperscript{32,41} although there are some findings that indicate skin temperature may influence thermal thresholds.\textsuperscript{41-42} Investigation of these covariates will allow for more powerful statistical comparisons and greater accuracy for threshold estimates.\textsuperscript{30,43}
A further limitation of published normative values is the lack of appropriate statistical analysis. Characteristics of sensory thresholds indicate that the observed threshold values are unlikely to follow a normal distribution. However, many researchers rely on this assumption when calculating percentiles and this may induce bias. Adjusting for continuous covariates such as age is often based on arbitrary categorization which leads to a low sample size within each category. For example, Yarnitsky and Sprecher recently attempted to establish solid reference values for thermal thresholds using several different test algorithms in a sample of 106 healthy volunteers. Stratified statistical analysis was done within each age category. This effectively limited the sample size, creating unstable estimates of normative values. The numerical instability of the estimated normal limits is demonstrated by the nonmonotone relationship of the limits with age. Estimates of cold thresholds on the hand of females decreased for 40 - 59 year olds compared to 20 - 39 years, then increased again for the 60 - 79 year olds compared to the 40 - 59 group. The reverse of the effect observed in females is seen for cold testing on the foot among males.

The aim of this study was to make available reference values from a healthy population for thermal and vibration sensory thresholds. Covariates which have been identified as potentially important have been systematically evaluated. To avoid bias in estimating percentiles, we have used flexible statistical methods that are not dependent on a priori assumptions regarding the distribution of the observed thresholds.
Materials and Methods

Subjects. Adults without evidence of neuropathic disease were eligible for this study. An effort was made to obtain an approximately uniform distribution of gender and age, with a good representation of elderly subjects. Healthy volunteers were recruited. Subjects with known diabetes mellitus were excluded. All subjects signed an informed consent, completed the Michigan Neuropathy Screening Instrument (MNSI)\textsuperscript{46-47} and were given a standardized clinical examination for response to light touch, vibration and pinprick in the foot. A score of $\geq 2$ points on the MNSI or one absent response for the clinical exam excluded the subject.

Apparatus. Testing for vibration sensitivity was performed on the Vibratron II (Sensortek Inc., Clifton, NJ). The instrument consists of two posts that are set to vibrate at different amplitude levels. The maximum possible amplitude corresponded to approximately 200 microns. Thermal tests were conducted with a Medoc TSA 2001 thermal sensory analyzer (Medoc Ltd., Minneapolis, MN). This employs a micro-computer driven 46 x 29 mm Peltier thermode with a temperature range was 0 - 50°C.

Testing algorithms. For thermal thresholds, the method of levels was used. Baseline temperature was kept at 32°C with rates of temperature change held constant at 1°C per second. This base temperature and rate of change are used by most researchers and clinicians.\textsuperscript{40,44} Stimuli were presented at an initial step of 4°C with the thermode returning to 32°C immediately after completion of the temperature step. Subjects were asked to state each time they perceived a change from baseline temperature. This was recorded as a
YES response. No indication that a stimulus was perceived was recorded as a NO response. Stimuli continued to be increased by steps of 4°C until the first YES response. Stimuli were then decreased by 2°C until the first NO was recorded. After that, the step was halved for each successive stimulus with the direction changing according to response: increasing for NO and decreasing for YES. The test was completed when the step reached 0.2°C which corresponded to the detection threshold of the apparatus. The mean of the stimulus temperature for the last YES and the last NO was used for the threshold estimate. The subject was not asked about the stimulus after each presentation. This modification of the testing paradigm developed by Yarnitsky and Ochoa was implemented to avoid cuing subjects for a response.

Vibration thresholds were estimated using the two-alternative forced choice procedure described by Arezzo et al. Participants were required to touch the posts with one digit for approximately 2 seconds with enough force to blanch the nail. After touching each post, the participant indicated which of the posts contained the stimulus. Only one post was active for each trial. The active post was determined by the investigator using a pseudo-random procedure. If the response was correct, the stimulus intensity was reduced by 10% for each subsequent trial until the participant made the first error. Then the same stimulus was repeated twice. If the stimulus was correctly identified on two of three trials, a further reduction of 10% was made. If errors were made on two of three trials, the intensity was raised by 10%. Testing was completed when five errors had been made. The vibration threshold was determined by identifying the five errors and the five lowest correct scores, eliminating the highest and lowest values and calculating the mean for the remaining eight scores.
Procedure. Birth date, gender and height were recorded for each subject. Skin temperature was measured using a temperature probe applied to the thenar eminence of the right hand. All tests were performed by the primary investigator (GB). Distractions were minimized and participants were allowed 10 minutes to adapt to the room temperature. Visual access to the equipment was restricted. Vibration tests were conducted first on the index finger then on the large toe of the right side. Thermal tests were conducted after the completion of the vibration tests. The thermode was attached with a Velcro® strap that was adjusted to obtain maximum skin contact. Warm perception was tested on the thenar eminence. After obtaining the warm sensory threshold, cold perception was tested at the same site. The two thermal sensations were then tested on the dorsum of the foot.

Statistical Analysis. Independent effects of covariates on threshold values were estimated using multiple linear regression. Departures from linearity were assessed by testing the significance of a quadratic model. Significance levels were adjusted for multiple testing using the Bonferroni correction. For thresholds with no significant covariates and a limited number of observed values, the probability of a randomly selected subject exceeding each value observed in the sample was calculated. Ninety-five percent confidence limits were calculated for the probabilities using Fleiss' method for single proportions, that accounts for the asymmetry of the confidence interval in the case of very low proportions. Data were stored on Quattro Pro 6.0 for Windows®, and regression analyses were performed using SAS 6.0 for Unix®. Nonparametric statistical methods and density estimations were completed using S-Plus for Unix®.
Adjusting for the Effect of Age. For estimating the distributions of the thresholds, a priori selected covariates (age, gender, height and skin temperature) were taken into account if and only if these respective effects were found to be statistically significant and clinically relevant in the multiple regression analysis described above. In the case of the significant difference between two genders, separate distributions were estimated for males and females using appropriate subsets of data. However, stratified analysis was not adequate in accounting for the effects of continuous covariates such as age since the sample sizes for each age category would be low, leading to unstable estimates. Therefore, in the case of a significant effect of age, the first step of the analysis involved calculating residuals from the linear model regressing the threshold on the subjects' age. The numerical value of the residual represented the discrepancy between the actual threshold of an individual subject and the threshold that would be expected given this subject's age. Thus, the variation of the residuals represented the between-subject variation after removing the systematic effect of age. Accordingly, the distribution of residuals represented the generic shape and spread of the distribution of individual scores, around the unknown mean, in a hypothetical population of subjects who all have the same age. To reconstruct the distribution of the scores for a specific age value, the entire distribution of residuals, which by definition have the mean of 0, was then shifted by the value corresponding to the expected mean value for this age. This value was estimated from the regression model by summing the intercept (correspond to mean score at age 0), the product of the regression coefficient for age (representing the effect of increasing age by 1 year), and the age value in years.

Estimating the Shape and Quantiles of Thresholds' Distribution. Once an appropriate distribution (either original values or residuals, possibly stratified by gender) was obtained,
it was first analyzed using conventional descriptive statistical methods. All data were transformed so that low values corresponded to better sensation and high values to more "abnormal" results. Histograms were created to assess the shape of distributions and the empirical quantiles were calculated. Particular interest was in high quantiles (such as 90th or 95th) corresponding to tentative cut-off points to define normal range. However, due to limited sample size, the empirical quantiles would be influenced too much by individual observations and, therefore, not reliable estimates of the true quantiles in the population. Therefore, the population distributions were estimated from the data and the quantiles of the estimated distributions were obtained.

Three models were considered when estimating the distribution of residuals: two conventional models (normal and log-normal) and a nonparametric density estimation approach. The lognormal model, based on the assumptions that logarithms of the values of the variable of interest are normally distributed, is a standard model for positively skewed distributions. To estimate this model the residuals were first transformed by adding a constant so that the minimum value would correspond to 1.0, then the logarithms of the transformed values were used to estimate the mean and standard deviation. The motivation for using a nonparametric approach was that it does not require any a priori assumptions about the shape of the true distribution, except that it is smooth and continuous. The shape is estimated directly from the data by making use of the flexibility of regression splines (e.g., piecewise polynomials) which have proved useful in estimating distributions of diagnostic variables in other clinical studies. The fit of the three models to the data was compared based on log likelihoods as well as on Akaike
Information Criterion (AIC) that takes into account the difference in degrees of freedom between parametric and more complex nonparametric models.53

In some analyses, the effect of height was found to be statistically significant and of potential clinical importance. In that case, the estimated distributions and their percentiles should be considered as corresponding to a "typical height", arbitrarily fixed at 170 cm. For different heights, the quantiles could be obtained by adding a correction factor based on the linear regression model.

**Results**

Sixty-seven men and 81 women participated in the study. Age of the participants ranged from 20 to 86 years. Table 1 shows the distribution of participants by age and gender. Average height for the sample was 170 cm (s.d.=10). Initial skin temperature varied from 29°C to 36°C with a mean value of 33°C (s.d.=1.8). Vibration units were expressed in microns and thermal thresholds as an absolute difference in temperature from the base temperature.

*(insert Table 1 here)*

Warm and cold thresholds in the hand showed very little variation. Height and age were significantly associated with warm thresholds in the hand (p<0.01) but their effects were not clinically important as the maximum possible differences in predicted warm thresholds across the range of height and age observed in the sample was below the level detectable by
the testing apparatus (0.2°C). Initial skin temperature was significantly associated with a
decrease in cold thresholds for the hand (p<0.005) but this effect was again below the
detectable level of the equipment.

*insert Table 2 here*

Table 2 reports the estimated percentage of the study population that exceeds each possible
temperature value for warm and cold thresholds in the hand with the calculated 95%-
confidence intervals for each percentage (the division by a factor of 2 reflects the accuracy
limit of the equipment). Based on the 95% confidence intervals reported in Table 2, 99.1 -
92.8% of the reported threshold values fall below 1.2°C for warm thresholds in the hand
and the same percentage fall below 0.6°C for cold thresholds in the hand. Using a
conservative approach, based on these percentages, the values of 1.2°C and 0.6°C can be
definitely established as at least the cut-offs below which fall 92% of the values,
respectively, for the warm and cold thresholds in the hand for a normal population aged 20
- 86 years. In our sample, the maximum observed values were 1.4°C and 1.0°C for warm
and cold thresholds in the hand, respectively. Based on these results we can reject the
hypothesis that the proportion of normal subjects exceeding the respective cut-offs is 5% or
more (p<0.0006). Thus, 1.4°C for warm sensation and 1.0°C for cold sensation can be
firmly established as cut-offs below which falls more than 95% of the values.

In the case of the remaining observed thresholds, there was considerable between-subjects
variation so that continuous data methods were used to analyze these thresholds. Age was
the only covariate that was statistically significant with a clinically important effect on both
the warm and cold thresholds in the foot \( p \leq 0.0002 \). After the residuals from the regression of thresholds on age were calculated, an outlier was identified for the cold thresholds. This 71 year old male subject's threshold exceeded the thresholds expected at his age by 80% more than any other thresholds among 148 subjects. Although it was not possible to establish whether this outlier actually represented a subject with neuropathy who had not been detected by clinical exam, the residual value was removed from further analyses in order to reduce the impact of the outlier on the models. The distributions of the remaining 147 residuals for cold thresholds and 148 residuals for warm thresholds were analyzed using normal and lognormal parametric models as well as the nonparametric density estimation methods. Table 3 shows that for both warm and cold thresholds in the foot, the flexible nonparametric method provides the best fit to the data as indicated by the lowest absolute values of the log likelihoods and the minimum values of the AIC. These results suggest that parametric models would provide systematically biased estimates of the distributions of the thresholds. Accordingly, nonparametric estimates were used to characterize the age-adjusted distributions and to determine their 90th and 95th percentiles.

*insert Table 3 here*

Age also had a statistically significant and clinically important effect on vibration thresholds in the hand and foot \( p < 0.0001 \). However, a closer investigation of the data indicated that while the effects of age across thresholds in the hand were linear, for the foot the linear relationship only held until the age of 65 years. As shown in Figure 1, beyond that age the variance of thresholds increases dramatically making it difficult to assess the effect of age. The continuous methods described for thermal thresholds in the foot are used to analyze
vibration thresholds as well, however their use for vibration thresholds in the foot is limited to the age range between 20 and 65 years.

Data in Table 3 indicate that for vibration thresholds the flexible nonparametric model also fitted the data better than either of the parametric models.

The estimated 90th and 95th percentiles of the distributions of warm and cold thresholds for the foot and vibration thresholds in the hand and foot in the normal population are shown in Table 4 for selected age groups.

Among 29 subjects, aged 65 years or more, the maximum value of the vibration threshold was 55.13 microns. Accordingly, it is unlikely that more than 10% of the normal subjects in that age range exceed this value (p<0.05). Hence, the value of 55.13 microns can be proposed as a tentative 90% cut-off for vibration thresholds for the foot in the elderly aged 65 years or more. More precise estimation of the distribution in this age group would, however, require testing a larger number of subjects over the age of 65 years.

Vibration thresholds in the foot were also significantly affected by the subjects’ height (p<0.003). This was clinically important since the thresholds for subjects of the same age increased, on average, by about 0.24 microns for each 10 cm increase in height. Therefore,
the cut-offs reported in the relevant column of Table 4 should be considered as valid only for the height of approximately 170 cm (corresponding to the mean value in our sample). Table 5 shows the corrections that have to be added to these cut-offs values for other relevant intervals of height.

(insert Table 5 here)

Discussion

This study attempted to provide normative data for thermal and vibration sensory thresholds while controlling for factors expected to influence observed values. We found age to be the most influential covariate when measuring threshold values. This finding is consistent with the physiological impact of the aging process and with the findings of various other investigators. The results from our study emphasize the necessity of including an adequate representation of older subjects especially when investigating the discriminative properties of the test.

Height and skin temperature were statistically associated with warm and cold thresholds respectively, assessed on the hand. However, the effects were not clinically important as they were below the detectable level for the testing apparatus. We concluded that thermal thresholds in the hand of healthy subjects are not significantly influenced by these factors.

With respect to gender, many investigators decide a priori to provide separate reference values for males and females. This often limits the precision of the results as the sample
sizes are halved if stratified analysis is used. The possibility remains that the observed effects of gender are, at least partly, due to confounding by height. In our study, height was significantly correlated to gender \((r=0.69, p<0.0001)\). When the effects of height were controlled for, there was no systematic difference between observed threshold values for men and women. There also was no difference in variance of observed values for men and women.

Height was significantly associated with an increase in vibration thresholds measured in the foot. The scope of this study does not allow us to postulate what mechanism may be responsible for this effect, however, we noted that these effects are only apparent in the lower limb for extreme heights and do not appear to be important for thermal thresholds. Further physiological research is needed to clarify the role of height in sensory perceptions. As noted by previous investigators, vibration thresholds become increasingly variable for subjects over the age of 65 years. Some authors have concluded that vibration perception may not be a reliable test for distinguishing healthy subjects from elderly neuropathic patients.\(^3\)\(^6\) The results from our study seem to support this conclusion.

Our study also demonstrated the advantages of using more elaborate statistical methods. As expected, the distribution of the threshold values did not follow a normal model. A log transformation, a standard way of handling positively skewed data failed to substantially improve the fit to the empirical data. The use of flexible nonparametric distributions allowed for a more accurate estimation and for avoiding bias due to a priori assumptions. This method was extensively studied via statistical simulations that showed lack of bias and satisfactorily low variance of the estimates.\(^5\)\(^1\) The method has been previously used in
analyzing sensory testing data and cardiovascular diagnostic indices. Experience with this method has shown that a sample size of approximately 150 is sufficient to ensure adequate precision of the estimates in the range of the 5th and 95th percentiles (M. Abrahamowicz, personal communication). Moreover, by combining results of regression analysis with nonparametric density estimation, we were able to estimate age-specific percentiles while ensuring their monotonicity. This avoided rather implausible nonmonotone relationships with age that were reported in a study where age was categorized and each age category was analyzed separately.

We have also attempted to optimize the testing algorithm employed for measuring threshold values. The more common method of limits has been shown to be influenced by subject reaction time and the rate of stimulus change. In our study, the testing methodology was restricted to reaction-time exclusive algorithms. The method of levels that was used to evaluate thermal sensation is quick to administer and is not influenced by the rate of stimulus change. However, this method has not been validated for the measurement of vibration thresholds. Therefore, the two-alternative forced choice method as developed by Arezzo was employed to measure vibration sensation. The 4, 2, and 1 stepping algorithm proposed by Dyck et al to measure vibration and thermal thresholds is a variant of the forced choice method. The method is quick to administer and excludes effects of reaction time; however, a specially-programmed computer is required (Computer-Assisted Sensory Examination System IV). Although a programmed personal computer developed by Medoc Ltd. was used to administer the method of levels for thermal testing, this algorithm may also be used with equipment that is not driven by computer software.
As emphasized by the consensus report of the Peripheral Neuropathy Association, adequate reference values from healthy subjects are essential for evaluation of patients.\textsuperscript{30} The sensitivity and specificity of thermal and vibration thresholds cannot be evaluated without an appropriate reference group.\textsuperscript{56} Without this information about the discriminative abilities of QST for thermal and vibration thresholds, the application of QST is limited. Our results provide useful data on the range of thresholds to be expected in a reference population of subjects free of clinical neuropathy, for a wide range of ages. To establish the diagnostic capabilities of these tests in a variety of patient populations, further studies are required.
References


Table 1. Distribution of age and gender in the study sample.

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Males</th>
<th>Females</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 - 29</td>
<td>14</td>
<td>15</td>
<td>29 (19.6)</td>
</tr>
<tr>
<td>30 - 39</td>
<td>12</td>
<td>15</td>
<td>27 (18.2)</td>
</tr>
<tr>
<td>40 - 49</td>
<td>14</td>
<td>15</td>
<td>29 (19.6)</td>
</tr>
<tr>
<td>50 - 59</td>
<td>11</td>
<td>13</td>
<td>24 (16.2)</td>
</tr>
<tr>
<td>60 - 69</td>
<td>9</td>
<td>8</td>
<td>17 (11.5)</td>
</tr>
<tr>
<td>70+</td>
<td>7</td>
<td>15</td>
<td>22 (14.9)</td>
</tr>
<tr>
<td><strong>Total (%)</strong></td>
<td><strong>67 (45.3)</strong></td>
<td><strong>81 (54.7)</strong></td>
<td><strong>148 (100.0)</strong></td>
</tr>
</tbody>
</table>
Table 2. Estimated percentage of study population exceeding cut-off temperatures.

<table>
<thead>
<tr>
<th>Cut-off in Excess of Baseline Temperature (°C)</th>
<th>Estimated % of Population Exceeding the Cut-off (with 95% Confidence Intervals)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Warm Thresholds in Hand</td>
</tr>
<tr>
<td>0.2</td>
<td>*</td>
</tr>
<tr>
<td>0.4</td>
<td>95.3 (90.1, 97.9)</td>
</tr>
<tr>
<td>0.6</td>
<td>10.1 (6.0, 16.4)</td>
</tr>
<tr>
<td>0.8</td>
<td>8.8 (5.0, 14.9)</td>
</tr>
<tr>
<td>1.0</td>
<td>3.4 (1.3, 8.1)</td>
</tr>
<tr>
<td>1.2</td>
<td>2.7 (0.9, 7.2)</td>
</tr>
<tr>
<td>1.4</td>
<td>0.0 (0.1, 3.2)</td>
</tr>
</tbody>
</table>

* Indicates that there were no observations for this value.
Table 3. Comparison of the fit to the data of the three models for thresholds' distributions.

Log Likelihood (AIC)* of the Age-Adjusted Residuals Under:

<table>
<thead>
<tr>
<th>Threshold/ Site</th>
<th>Normal Model</th>
<th>Log-Normal Model</th>
<th>Non-Parametric Model</th>
</tr>
</thead>
<tbody>
<tr>
<td>Warmth/ Foot</td>
<td>-208.08</td>
<td>-218.55</td>
<td>-183.83</td>
</tr>
<tr>
<td></td>
<td>(420.16)</td>
<td>(441.10)</td>
<td>(381.7)</td>
</tr>
<tr>
<td>Cold/ Foot</td>
<td>-177.41</td>
<td>-202.44</td>
<td>-141.01</td>
</tr>
<tr>
<td></td>
<td>(358.82)</td>
<td>(408.88)</td>
<td>(296.02)</td>
</tr>
<tr>
<td>Vibration/ Hand</td>
<td>-172.53</td>
<td>-201.13</td>
<td>-148.59</td>
</tr>
<tr>
<td></td>
<td>(349.06)</td>
<td>(402.26)</td>
<td>(307.17)</td>
</tr>
<tr>
<td>Vibration/ Foot</td>
<td>-152.8391</td>
<td>-179.2091</td>
<td>-141.81</td>
</tr>
<tr>
<td></td>
<td>(309.6782)</td>
<td>(362.4182)</td>
<td>(297.63)</td>
</tr>
</tbody>
</table>

* The AIC values (in brackets) provide a more conservative measure of goodness-of-fit which accounts for differences in the degrees of freedom between the three models.
Table 4. Selected percentiles for thermal and vibration thresholds by age category and site.

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Warmth - Foot (°C)</th>
<th>Cold - Foot (°C)</th>
<th>Vibration - Hand (microns)</th>
<th>Vibration - Foot (microns)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>90%</td>
<td>95%</td>
<td>90%</td>
<td>95%</td>
</tr>
<tr>
<td>20 - 29</td>
<td>3.5</td>
<td>4.9</td>
<td>1.5</td>
<td>1.9</td>
</tr>
<tr>
<td>30 - 39</td>
<td>3.8</td>
<td>5.2</td>
<td>1.6</td>
<td>2.0</td>
</tr>
<tr>
<td>40 - 49</td>
<td>4.1</td>
<td>5.5</td>
<td>1.8</td>
<td>2.2</td>
</tr>
<tr>
<td>50 - 59</td>
<td>4.4</td>
<td>5.8</td>
<td>1.9</td>
<td>2.3</td>
</tr>
<tr>
<td>60 - 69</td>
<td>4.7</td>
<td>6.1</td>
<td>2.0</td>
<td>2.5</td>
</tr>
<tr>
<td>70 +</td>
<td>5.0</td>
<td>6.4</td>
<td>2.2</td>
<td>2.7</td>
</tr>
</tbody>
</table>

* A different approach has been used to estimate these percentiles. See text below.
Table 5. Height-related corrections for vibration thresholds in the foot.

<table>
<thead>
<tr>
<th>Height (cm)</th>
<th>Correction of Values* Reported in Table 4 (microns)</th>
</tr>
</thead>
<tbody>
<tr>
<td>148 - 152</td>
<td>-0.5</td>
</tr>
<tr>
<td>153 - 157</td>
<td>-0.4</td>
</tr>
<tr>
<td>158 - 162</td>
<td>-0.2</td>
</tr>
<tr>
<td>163 - 167</td>
<td>-0.1</td>
</tr>
<tr>
<td>168 - 172</td>
<td>0</td>
</tr>
<tr>
<td>173 - 177</td>
<td>+0.1</td>
</tr>
<tr>
<td>178 - 182</td>
<td>+0.2</td>
</tr>
<tr>
<td>183 - 187</td>
<td>+0.4</td>
</tr>
<tr>
<td>188 - 192</td>
<td>+0.5</td>
</tr>
</tbody>
</table>

* Regression-based corrections are rounded-off to 0.1 microns.
Figure 1. Plot of age and observed vibration thresholds measured on the foot of 148 healthy subjects. After 65 years of age, threshold values increase in variance and no longer have a linear relation to age.
CHAPTER 4. CONCLUSION

Summary

The development of adequate reference values for sensory thresholds is essential for evaluation of the clinical and diagnostic capabilities of QST. I have attempted to estimate the distributions of thermal and vibration sensory thresholds for a healthy population. Although a great deal of research has been published in the field of quantitative sensory testing, the availability of adequate reference norms was limited by methodological issues. I have reviewed the literature to isolate specific issues that needed to be addressed when attempting to establish reference values. This review showed that the main problems arose from inconsistent use of testing algorithms, a failure to account for important covariates, the use of small or nonrepresentative samples and inappropriate statistical analysis. While some studies have partially addressed these issues, none of the published research utilized a methodology which provided adequate, stable estimates of reference norms for a healthy population. The literature review revealed that several factors contributed these methodological limitations. First, the choice of testing algorithm seemed to be based on equipment availability or preference rather than assessment of the validity of the technique for measuring sensory thresholds. Second, in studies that employed large sample sizes, the diversity of testing algorithms and inconstant measurement of covariates reduced the generalizability of the published norms. Third, with studies that addressed many of these important design issues, the resulting data was analyzed using inadequate statistical techniques.
In my own study, reported in the article contained as Chapter 3 of this thesis, I endeavored to address the above limitations. Based on the results of the literature review to identify appropriate test algorithms that have been well standardized and are considered a valid method of measuring sensory thresholds, I developed an appropriate methodology for assessing thermal and vibration thresholds. Using this algorithm, I then tested 148 volunteers who, by clinical criteria, were free of neuropathy. The study design included the measurement of several covariates that may have had an independent influence on the threshold levels as identified by the literature review.

Conclusions

In this study, age was identified as an important covariate for threshold estimates. Despite a priori expectations, gender was not found to be significant in my study after controlling for the possible confounding effect of height. The lack of significant effect I observed for gender may not be true of diseased populations. This hypothesis would also apply to effects for skin temperature. This needs to be investigated with more in-depth studies. It is possible that covariates that do not appear to influence the sensory thresholds of a healthy population, may differentially impact diseased populations.

The methodology I developed for estimating sensory threshold distributions will serve as a framework for studies comparing healthy and diseased populations. Ensuring a uniform distribution of age and gender with particular attention to subjects over the age of 50 years, enabled us to increase the generalizability of the results. Measurement of height allowed for correction values to be calculated for vibration thresholds in the lower extremity. Using
statistical techniques that did not impose arbitrary a priori assumptions on the distribution of thresholds helped eliminate bias in the estimation of percentile of interest whereas regression based methods allowed us to relate these percentiles to subjects' ages.

In conclusion, the findings of this thesis highlight the importance of using appropriate methodology at both the stage of measurement and the stage of data analysis, for establishing adequate reference norms. The study contained in this thesis is only a preliminary step in assessing the diagnostic capabilities of quantitative sensory testing. Research involving patients with differing degrees of peripheral neuropathy must be conducted in order to establish the discriminative capabilities of the tests. Ideally, long term follow-up studies of a cohort of subjects should be implemented to determine the ability of the tests to predict clinical implications of peripheral neuropathy. Results of this thesis provide references against which the data collected in such studies may be compared.
REFERENCES


