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Pressure pain mappings - development and applications

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This dissertation is based on the following peer-reviewed articles referred to by their Roman number in the text.

Study I: Binderup AT, Arendt-Nielsen L, Madeleine P. Pressure pain threshold mapping – A new imaging modality of muscle sensitivity to pain. 2008 Annual IEEE Student Paper Conference 2008; 1:126-129. DOI: 10.1109/AISPC.2008.4460549.

Study II: Binderup AT, Arendt-Nielsen L, Madeleine P. Cluster analysis of pressure pain threshold maps from the trapezius muscle. Comput Methods Biomech Biomed Engin 2010; 8:1. DOI: 10.1080/10255840903446979.

Study III: Binderup AT, Arendt-Nielsen L, Madeleine P. Pressure pain sensitivity maps of the neck-shoulder and the low back regions in men and women. BMC Musculoskelet Disord 2010; 11:234. DOI:10.1186/1471-2474-11-234.

Study IV: Binderup AT, Arendt-Nielsen L, Madeleine P. Pressure pain threshold mapping of the trapezius muscle reveals heterogeneity in the distribution of muscular hyperalgesia after eccentric exercise. Eur J Pain 2010; 14(7):705-712. DOI:10.1016/j.ejpain.2009.11.001.

Study V: Binderup AT, Holtermann A, Søgaard K, Madeleine P. Pressure pain sensitivity maps, musculoskeletal disorders and long term sickness-absence among cleaners. Int Arch Occup Environ Health 2011; 84:647-654. DOI: 10.1007/s00420-011-0627-6.

Contents

Preface

The present studies were carried out at Centre for Sensory-Motor Interaction (SMI), Aalborg University, in the period from 2007 to 2010. Current PhD stipend was funded by Aalborg University.

I am grateful to all co-authors for their contributions and for a fruitful collaboration. Especially, I wish to express my gratitude to Professor Pascal Madeleine and Lars Arendt-Nielsen for their supervision and guidance to my projects. I would also like to thank all my colleagues at Centre for Sensory-Motor Interaction for providing a friendly, inspiring and competitive research environment. Particularly, I thank Afshin Samani for his collaboration during the data collection for study II and IV. A further thank to all the volunteers who participated in the experiments of the current PhD study.

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Lastly I would like to thank my parents and my dear Xixi who gave me support and encouragement through this ordeal.

Introduction

Musculoskeletal disorders

Recent European surveys still report substantial evidence that musculoskeletal disorders (MSD) located in the back, neck-shoulder regions are a significant ill health and cost problem (Schneider and Irastorza, 2010). Every year millions of Europeans employed in all kind of sectors are affected by MSD. These disorders cover a broad range of health problems such as back pain/injuries and work-related upper limb disorders. Moreover, lower limb region can also be affected. The treatment and recovery from MSD are often problematic due to the chronicity of the symptoms. MSD can thus, result in permanent disability. According to the latest European Survey on Working Conditions, 24.7% of the European workers complain of backache, 22.8% of muscular pains, 45.5% report working in painful or tiring positions while 35% are required to handle heavy loads in their work. Backache and neck-shoulder pain are often reported to be the most prevalent work-related health problem.

MSD are a cause of major concern due to their health effects on individuals and their economic impact on businesses and the social costs. As an example, the cost of work-related upper-limb MSD in Europe is estimated between 0.5 and 2% of Gross National Product and their impact is still increasing. Interestingly, there are also gender differences in the type and frequency of MSD occurrence. Thus, as expected MSD account for a significant proportion of absenteeism at work. This is in fact confirmed by a number of studies reporting a huge impact of MSD on work-related absence and a high proportion of days lost in Europe.

MSDs have a multifactorial aetiology and in most cases, it is difficult to point out the exact cause of an individual case of disease. Further, these disorders are not commonly accepted as occupational diseases in the national compensation or reporting systems. There is to date, little evidence of the use of standardised diagnostic criteria for MSDs across countries (Buckle and David, 2000). This variation is reflected in the nationally reported data and makes comparisons difficult. Low back and neck-shoulder region disorders are accepted as occupational diseases by only a few EU-countries and only for specific forms of diseases (Biosca de Sagastuy and Skaliotis, 2000). Based on the current knowledge, it can nevertheless be concluded that MSD are also the most common occupational disease.

MSD are a group of disorders often accompanied by pain from muscles, tendons, joints and nerves (Madeleine, 2008). All parts of the body can be affected, although upper limb and back are the most common areas. Even if MSD and their implications represent an important challenge, our basic knowledge of pain from deep structures in human is still limited. The quantification of the deep structures sensitivity to pain can provide a better understanding of underlying mechanisms behind MSD.

Visualizing pain

Over the recent years, a lot of effort has been put into developing tools and methods to assess and visualize painful conditions in muscle and skeletal tissue in attempt to uncover localization and spread and to prevent development of chronic pain. Pain is defined as an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage (Merskey and Bogduk, 1994). Chronic musculoskeletal pain causes reduced quality of life with loss of work and social networks for the individual and is an economical burden for the society (Punnett and Wegman, 2004). Tools such as ultrasound scans (O'Sullivan et al., 2009) and magnetic resonance imaging (Horrigan et al., 1999) are frequently used to visualize the physical state of the muscle, in attempt to find a physical source for the pain. While rich in image detail and resolution, these finding do not always correspond well with the felt pain sensation and can provide misleading evidence to the cause of the pain (Manchikanti et al., 2000). Similarly, multi-channel surface electromyography (EMG) has further been used to give insight into how muscle activation patterns changes with the presence of pain

(Madeleine et al., 2006). However, none of these methods are a measurement of the pain itself, only possible cause or derivatives, and do not enable visualization of the actual musculoskeletal pain sensitivity and its spatial distribution throughout the tissue.

A different kind of approach more suited for evaluating the actual pain that the subject is experiencing is known as quantitative sensory testing. These methods are all based on giving sensory input to the test subject and have a quantifiable parameter that defines different levels or thresholds of perceived pain intensity. The pain threshold is defined as the least experience of pain which a subject can recognize (Merskey and Bogduk, 1994). These sensory inputs can be temperature related (warm, cold) or mechanical as light touch, brushing or pressure force against the muscle (Hansson et al., 2007).

Pressure pain algometry

Assessing mechanical sensitivity is usually performed by palpating sensitive body areas. However, manual palpation does not provide quantitative value reflecting deep structure sensitivity. To circumvent this, pressure pain algometry (PA) has been used for many years as a method to assess conditions in the muscle, like level of tenderness related to inflammation, myofascial pain and fibromyalgia. PA is also widely used to study the effects of anesthetics and drugs in human tissue (Kumar et al., 2006, Hsieh et al., 2010, Lemming et al., 2007).

PA is usually performed by using a handhold pressure algometer that is pressed vertically at a constant speed against the skin of the subject. When the subject feels that the sensation of pressure shifts to a sensation of pain he or she informs the examiner, who immediately stops applying more pressure. This pressure level is defined as the pressure pain threshold (PPT). This threshold is often used in studies as the pressure does not cause any damage to the skin/muscle tissue. As such PPT is based on a subjective evaluation of pain (like the VAS where subjects rate perceived pain intensity on a scale), but has showed great reliance in reproducibility and allows clinicians and researchers to quantitative compare findings (Fischer et al., 1998).

The receptor fibers that are invoked by the pressure towards the muscle are nociceptors belonging to group III (thin myelinated) and group IV (unmyelinated). Nociceptors react to noxious (tissue-threatening) stimuli, though the threshold level is lower than the actual level required to damage the tissue. As such, their purpose is to warn the central nervous system about potential as well as actual damage to the skin/muscle tissue (Mense et al., 2001). As the increase in pressure force is stopped the moment the nociceptors starts to the discharge the force level applied by PPT are too low to inflict permanent damage to the tissue (Fischer et al., 1998). To prevent temporal summation, which means increased sensitivity towards pain due to repetitive stimulation within a short time frame, the time interval between two consecutive presses at the same location should be at least 30 seconds (Nie et al., 2006). Further, the probe should be of a size smaller than the receptive field of the muscle nociceptors (<3 cm^2)(Simone et al., 1994). A probe size of 1 cm^2 has proven effective to activate the deep tissue afferents (Nie et al., 2009) and has been chosen as an appropriate size for this study.

PPT recordings have already been used to assess spatial sensitivity differences within the same muscle, though there are contradicting results to which parts of the muscle that are the most sensitive in general, and which are affected most by different conditionings. The musculotendinous junction has been found more sensitized towards pain by strenuous eccentric exercise compared with the muscle belly (Fridén and Lieber, 2001, Newham et al., 1983) while other studies have reported the opposite (Andersen et al., 2006, Nie et al., 2005, Weerakkody et al., 2003). What is known though is that difference between two adjacent PPT recording sites can be vastly different depending on the muscle at distance between sites (Andersen et al., 2006, Nie et al., 2005, Weerakkody et al., 2003). Females have been observed to have generally lower PPT than males even in healthy subject (Chesterton et al., 2003). The cause of this difference is not fully understood (Greenspan et al., 2007) but physiological (Cairns,

2007), cultural (Dawson and List, 2009) and psychological factors (Miller and Newton, 2006) have been found to play a role. PPT has also been used as a semi-objective measurement of work related MSD and the conditions of chronic these leads to (Gold et al., 2006, Schenk et al., 2007).

Muscles of interest

MSD have the highest prevalence in the low back and neck-shoulder regions of the body (Breivik et al., 2006, Bernard, 1997), making mapping of the major muscles in these regions particular interesting. For the neck-shoulder region, the trapezius muscle is an excellent muscle to try to map due to its size and accessibility. It connects at the acromion bone on the shoulder and all the way from the neck to the twelfth thoracic vertebrae of the spine. The trapezius is divided into three subdivisions, and each division is defined based on the muscle fiber direction and functional involvement in different shoulder and neck movements (Inman et al., 1996). The fibers in the upper part attach from the ligamentum nuchae to the firth cervical vertebrae. These fibers elevate the shoulder and are used in bending of the neck and rotation of the head. The fibers in the middle part attach from the sixth cervical to the third thoracic vertebrae. This part is involved in movements of scapula. The fibers in the lower part attach from the third thoracic to the twelfth thoracic vertebrae and are also involved in movements of scapula (Travell et al., 1999).

Previous studies on the muscle have shown non-uniform morphological and histological properties of the trapezius muscle fibers has been reported (Lindman et al., 1990) supporting an independent control of the three trapezius subdivisions (Madeleine et al., 2006, Mathiassen and Aminoff, 1997). It has also been shown that the PPT is significantly different in different parts of the trapezius muscle (Nie et al., 2005), but a broad overview of the spatial distribution of PPT in the whole muscle in healthy subjects and how/if this distribution changes with muscle pain is missing.

In the low back region, the erector spinae is an important muscle due to its involvement in supporting the spine in daily bending and lifting and as source to low back pain (Breivik et al., 2006, Bernard, 1997). The muscle is a bundle of three other muscles: The longissimus, the spinalis and the iliocostalis. The muscle connects both to the neck at processus mastoideus and to the pelvis at os sacrum as well to a number of rib bones and runs all the way along the spine. A focus on the region from the first to the fifth lumbar cervical vertebrae is sound as this part of the muscle is not covered by other more superficial muscles. Though the number of studies on the spatial pain distribution of the low back has been limited, there has been found significance between PPT measurements closer to the spine compared to those on the lateral side of the muscle (Hirayama et al., 2006).

Aims of this Ph.D. Project

Most of the previous PPT studies mentioned have only focused on a very small number of PPT recording sites at different locations on the muscle, either deliberately or due to the small muscle size. This study will instead use a high number of sites to increase spatial sensitivity of PPT measurements on the muscle by using a geometrical grid based on a few anatomical landmarks to set the PPT recording sites placement on muscle.

As such research involved finding an optimal interpolation method to interpolate the PPT values between the recording points. The most optimal method will be the one that can best predict the value of a “missing” recording point based on the data from the remaining recording points. A method for automated grouping of the recorded values should also be developed, to identify zones of approximate equal sensitivity. This grouping can either be done based on the recorded values themselves or on the properties of the PPT map. It is important to

have a grouping method that can strongly discriminate between regions of “equal” PPT levels, and to have a method that ends up with the same groups when applied multiple times to the same data.

This provides the overall aim of the presented PhD. project: To develop a new mapping modality for pressure pain assessment of the trapezius and erector spinae muscles through a high resolution spatial map of PPT recordings.

This requires: 1) methodological studies for an efficient mapping, that both visually and statistically separate the map into zones of pain sensitivity, and 2) experimental and clinical studies, where the methods are applied in an experimental and clinical setting to assess conditions in the trapezius or erector spinae muscle.

Five studies were performed to fulfill these objectives. Two studies were primarily aimed at developing a new PPT mapping technique while the remaining three aimed at applying the mapping technique on subjects to observe differences in pain perception and localization between groups, see Figure 1. Each study resulted in a peer reviewed article for publication in an international scientific journal.

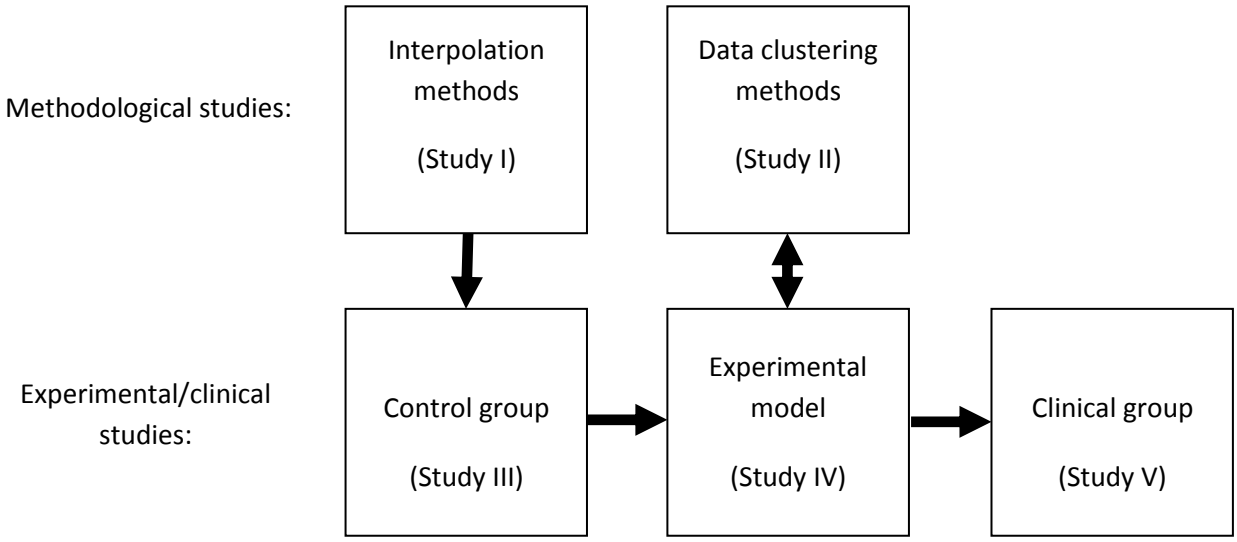

Figure 1. Conducted studies. The arrows show interrelation between studies and development of the mapping method.

Methodology

Establishing a recording grid

The first step was to define a grid setup for the PPT measurements. This grid was desired to be systematically defined so it could be applied with ease on any subject. Other considerations included having a high spatial resolution while at the same time not having points so close that the recording sites would overlap (due to the size of the algometer probe), which would increase the possibility of spatial summation of pain. Spatial summation is increased sensitivity towards pain due to invoking stimulation in a larger amount of receptive fields (Nie et al., 2009). Also the number of points had to be adjusted with regard to the amount of time the measurements would take. It was desired to have more than one PPT recording per point, but not to have the whole recording session last more than half an hour.

For the trapezius muscle it was decided to have the grid based on only two anatomical landmarks; the seventh cervical vertebrae (C7) and the acromion bone. Both easily identifiable by manual palpation. The distance between these two landmarks would be the basis for the distances between points in the grid both in the medial/lateral and the cranial/caudal direction. A reasonable distance between points when taking all previous mentioned considerations into account was found to be one sixth of the C7-acromion distance. The only exception to this rule was for the points (1, 2, 3 and 4) located in the neck part of the muscle where the muscle width is small. The horizontal distance here was set to one seventh of the C7-acromion distance to make certain that all points would be placed on the muscle. The adjacent point to acromion was placed on twelfth of the distance which also were the distance between the points in the spinal processes and the muscle (see Figure 2). A total number of 36 points were used to cover all parts of the trapezius muscle while 12 were used to cover the spinal processes next to the muscle, giving a total of 48 points in the neck-shoulder region. A further 36 were needed to cover the trapezius on the contra lateral side in cases where we wanted to cover the whole back for a grand total of 84 points (study III and V).

For the low back, the assessable part of the erector spinae muscle was smaller than for the trapezius. As such, fewer points could be placed on the muscle. This was again desirable not only to prevent spatial summation of pain but also to make the spatial resolution of the PPT maps similar between muscles. Due the low amount of point, it was decided to always record on both side of the spine and with five points on the spine itself. The distance between the first (L1) and fifth lumbar vertebrae (L5) was used as the baseline for the distance between adjacent points in this region, see Figure 3. Five points were placed on the spinal processes with one fourth of this distance between them. That would ensure that each point were located on each of the five lumbar vertebrae. The first column on the muscle on each side of the spine was also placed one fourth of the L1-L5 distance from the baseline to be clear of the spine. The points in these two columns were placed with only one twelfth of the L1-L5 distance to increase spatial resolution. One column more one each muscle was placed further out. There as only space for two points in each column due to the shape of the muscle. The low back was only measured in study III and IV.

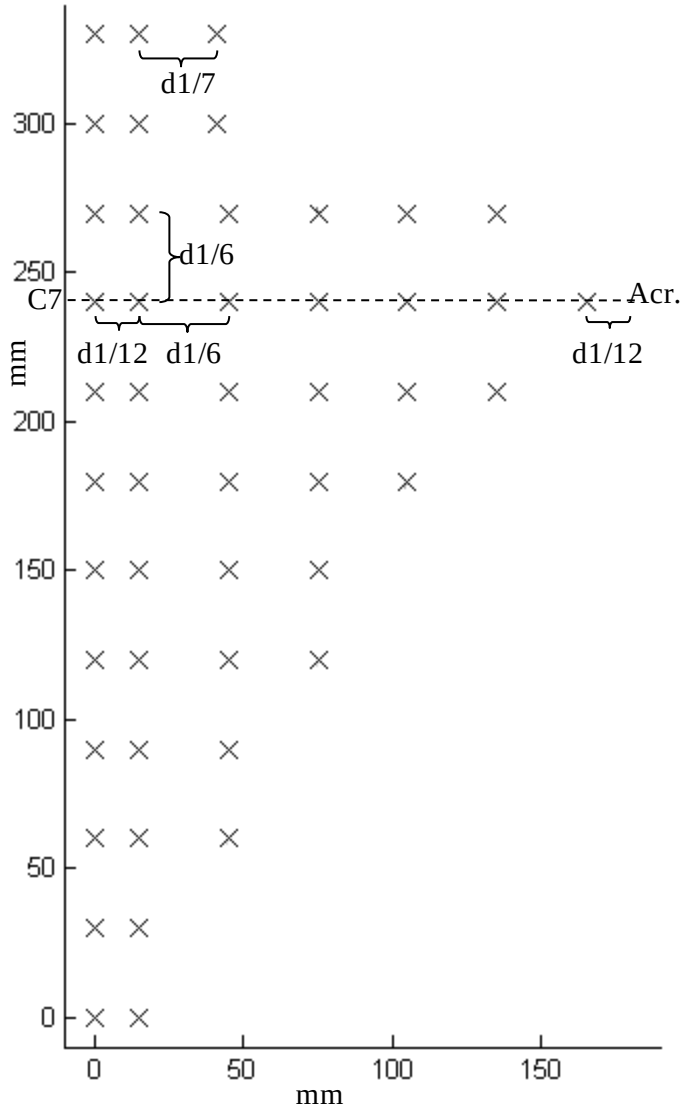

Figure 2. Schematic representation of the PPT recording grid over the neck-shoulder region (48 points). All distances between adjacent points were based on an anthropometric measure, i.e. the distance (d1) between the seventh cervical vertebrae (C7) and acromion (Acr.).

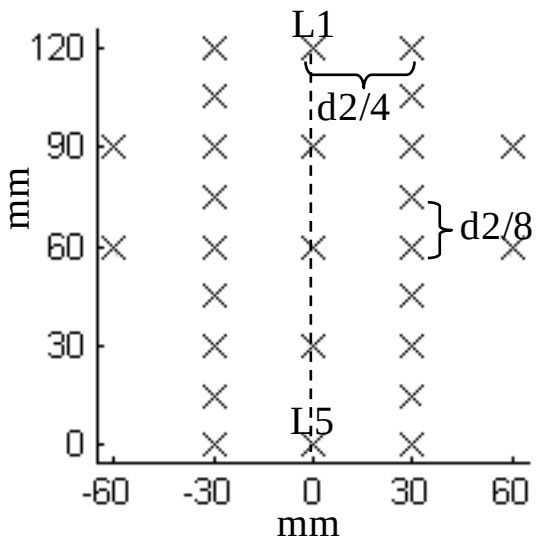

Figure 3. Schematic representation of the low back PPT recording grid (27 points). All distances between adjacent points were based on an anthropometric measure (the distance (d2) between the first (L1) and fifth (L5) lumbar vertebrae).

The recording order was chosen as either going column wise or row wise and to avoid bias a random selection between recording patterns was used. The proposed recording grid setups provide good coverage of the regions of interest with while being simple and quick to measure up and provide as such a good foundation for the PPT maps.

For every point, at least two PPT recordings were made. If these measurements were highly different (coefficient of variance of 0.2 or more), a third recording was done, and the mean value of the two recordings with the lowest mean was used as the used for that point. To test if this was a valid approach, the correlation coefficient between the two recordings were computed and found to be 0.8 or more (study III). To normalize PPT among subjects every PPT recording in each region (neck-shoulder and low back) was divided by the mean PPT for that region and subject.

Interpolation methods

It is necessary to interpolate to estimate the PPT values between the recording points to visualize PPT recordings as maps. There are different ways of interpolating data with both benefits and drawbacks. The most simple and commonly used way of interpolation between two known values is linear interpolation. It can only interpolate in two dimensions within a rectangle and is as such unusable to interpolate data using the selected grids. As such, a method that can interpolate (and to some degree extrapolate) within a scatter point setup, like the previously presented PPT recording grid, is needed.

One of these methods is known as inverse distance interpolation where the unknown value of any point can be computed as a summation of all known point values multiplied with and adjustment for the distance between every known point and the unknown. In mathematical terms that can be described like this:

$$F(x,y) = \sum_{i=1}^{n} w_i f_i$$

where n equals the number of recorded points, f_i is the value of the recorded point and w_i is the weight of the recorded point. In the classical form given by Shepard (1968) the weight value is computed as

$$w_i = \frac{h_i^{-2}}{\sum_{j=1}^{n} h_j^{-2}}$$

where h_i is the distance from the interpolated point to the recorded point given by

$$h_i = \sqrt{(x - x_i)^2 + (y - y_i)^2}$$

where (x,y) is the coordinates for the interpolated point and (x_i,y_i) is the coordinate of each recorded point. An improved method of inversed weighted interpolation was proposed by Franke and Nielson (1980) where the weighting is computed as

$$w_i = \frac{\left[\frac{R - h_i}{R h_i}\right]^2}{\sum_{j=1}^{n} \left[\frac{R - h_j}{R h_j}\right]^2}$$

where R is the distance from the interpolated point to the most distant recorded point.

Figure 4 demonstrates the non-linearity of the inversed distance interpolation method using Franke and Nielson weightings in a two point setup where each point is being defined by a single co-ordinate. This demonstrates how known point values proportionally contribute more to the estimation calculation of points laying close to them than those further away.

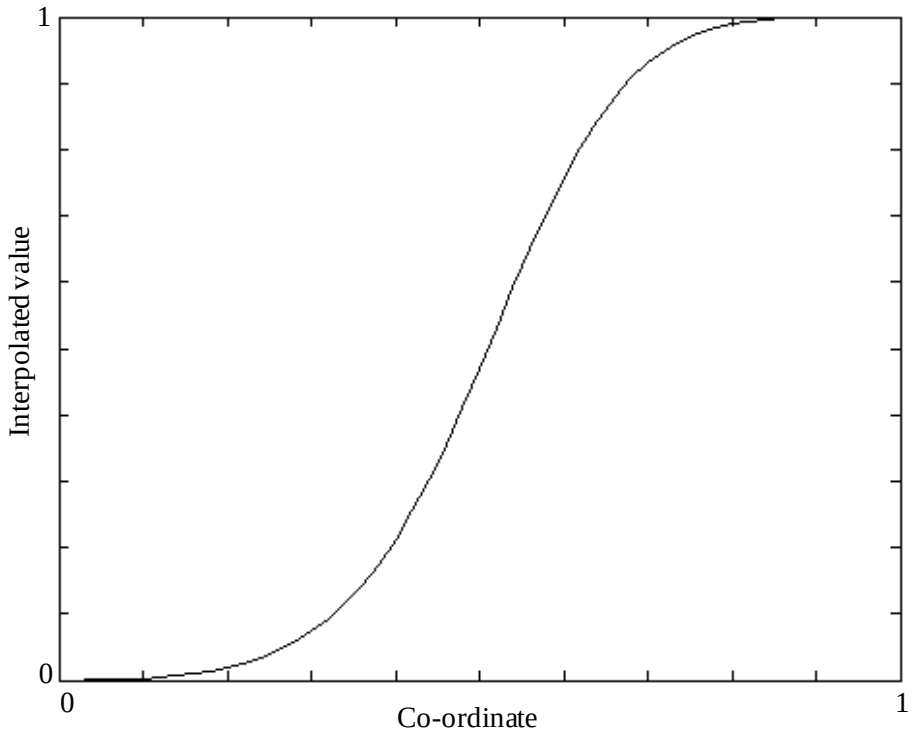

Figure 4. One co-ordinate example of inversed distance interpolation with Franke and Nielsen weightings between two known points. One point has co-ordinate and known value of 0 while the other has 1.

The interpolation method shown fulfills the needs required for generating the PPT maps in a representative way allowing for meaningful interpretation based on the visual inspection alone. With this method it is possible to show pain topography which is the first step in studying the spatial proportions between different parts of a muscle.

Designing a clustering approach

A high number of measurements points on the trapezius muscle enable a division of the map into subgroups. This is not done for the erector spinae muscle due to the relative low number of points on each muscle. The simplest approach we selected, i.e. the PPT points were assigned to three groups based on the three anatomical subdivisions (upper, middle and lower), see Figure 5. The number of points in each subdivision was not equal but that was not possible due to the different sizes of the muscle subdivisions and to keep the general symmetry of the grid.

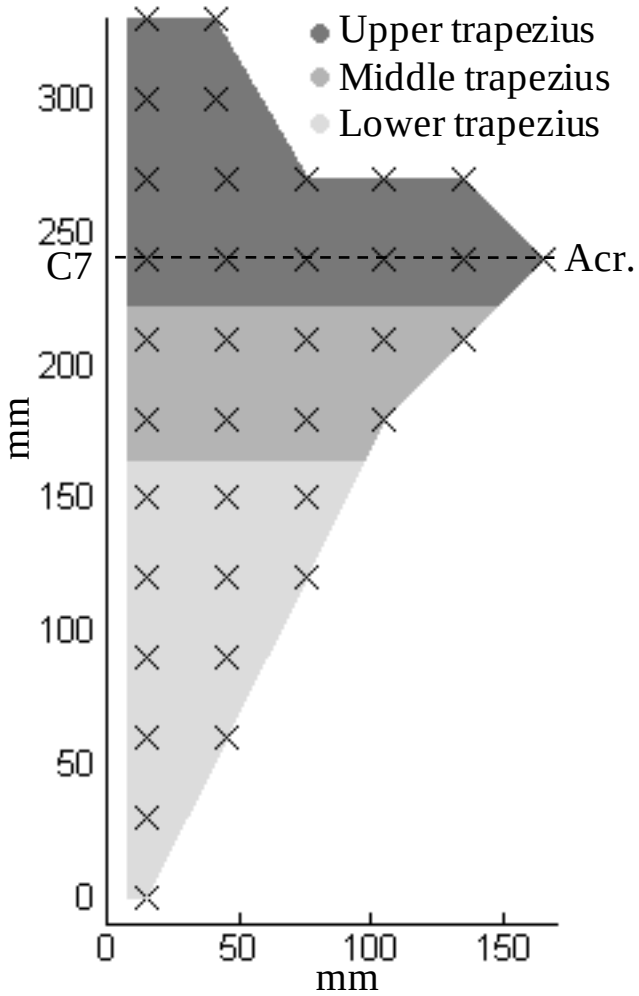

Figure 5. Trapezius muscle divided roughly according to the anatomical subdivisions

And alternatively approach is to disregard the position of the points on the muscle at first and instead group or cluster based on the PPT values recorded at each point. By comparing the position and grouping of the points after the clustering process, it would be possible to determine if there are certain areas with distinct pressure pain sensitivity. Clustering methods and algorithms can be divided into two basic types: hieratical and non-hieratical (Everitt et al., 2001). Hierarchical algorithms progress through a series of steps that build a tree-like structure by either adding individual elements to (i.e., agglomerative) or deleting them from (i.e., divisive) clusters. Non-hierarchical algorithms (also referred to as iterative methods) partition a data set into a pre-specified number of clusters. Before selecting on what method to use, we evaluated two different methods by applying them on PPT measurements (obtained from study IV), one hieratical method (Minimum value difference) and one non-hieratical method (K-means clustering).

I had set up the following goals for our desired ideal clustering method:

1. Consistency. The method should provide the same final assignment of points to clusters when provided with the same data input multiple times.
2. Unsupervised. The method initialization should be based on some fixed parameters that must not be changed between subjects.
3. Distinctive clusters. No point can be present in more than one cluster when the method has finalized.
4. No empty clusters. All clusters must contain at least one point at any time during the process.
5. Cluster size. The method should not necessarily aim at making all clusters equal size.
6. Labeling. The points should be label according to the mean value of the data in the cluster, so Cluster 1 represents the lowest values, Cluster 2 the second lowest and so on.

To fulfill goal 6, I ran a procedure in the end of all the clustering methods that sorted whatever number of final clusters that had been found according to the mean PPT value of the points assigned to the individual clusters. Cluster 1 would therefore always represent the cluster with the lowest mean absolute PPT value, cluster 2 the second lowest and so on.

For the cluster method description we use notations for different variables. Their definition can be seen in the following table:

Notation	Definition
N	Total number of data points.
p_i	Absolute PPT value of a data point.
P	Array of all data points: $P = \{p_1, p_2, \dots, p_N\}$.
$d_{i-1,i}$	Distance between neighboring point p_{i-1} and p_i. As the scale is 1-dimensional the distance is: $d_{i-1,i} = \lvert p_{i-1} - p_i \rvert$.
K	Total number of clusters.
n_j	The number of data points assigned to cluster C_j.
C_j	A cluster defined by the points assigned to that cluster: $C_j = \left\{p_{j,1}, p_{j,2}, \dots, p_{j,n_j}\right\}$.
c_j	Centroid value of cluster C_j: $c_j = \frac{1}{n_j}\sum_{i=1}^{n_j} p_{j,i}$.

Minimum value difference clustering

This primary goal is to cluster points together with low difference in PPT between them. The method did not merge clusters or reassign points when they have been assigned a cluster so it was hieratical by nature. Thus, the variances within the clusters were kept low (standard deviation below 13.7). The procedure of the method is described by the following steps:

1. Sort values in the point pool *P* according to value into a scale going from min(*P*) to max(*P*).
2. Compute difference between all neighboring points on the scale: $D = \{d_{1,2}, d_{2,3}, \dots, d_{N-1,N}\}$.
3. Select the pair with the lowest difference and assign them as the first cluster: C_1.
4. Compute the mean of the two values as the centroid value of a new cluster and remove the points from the point pool *P*.
5. Compute differences between all neighboring points and cluster centroids.
6. If the lowest difference is between two cluster centroids: ignore and find the lowest difference either between two points or a point and a centroid.
7. If the lowest difference is between two points: assign them to a new cluster and go to step 3.
8. If the lowest difference is between a point and a cluster centroid: assign the point to the existing cluster and compute a new cluster centroid based on the old centroid value and the value of the point.
9. Remove the point from the point pool *P*.

10. Repeat steps 1 to 7 until all points have been assigned a cluster.

Outcome

The method provides a high number of final clusters for the test data (mean±standard deviation: 15.0±0.8). The points assigned to each cluster were scattered over the muscle and there were no repetition between subjects. These factors made it impossible to setup some parameters for comparing the clusters between subjects and the method was therefore discarded.

K-means clustering

This is a common used non-hieratical clustering approach (Hartigan and Wong, 1979). The method is strong as it maximizes variability between the clusters while minimizing the variability within the clusters. The method works by selecting a number of initial cluster centroids through different procedures, described in detail later, within the range of the PPT data. The clustering is then run through the following steps until there is no longer change in the cluster assignment of the data points.

After having chosen initial cluster centroids:

1. Compute distance between every single point and all the cluster centroids.
2. Assign every point to the cluster of the nearest cluster centroid.
3. Compute new cluster centroid for every cluster based on the values of the points assigned to the cluster.
4. Repeat step 1 to 3 until the cluster assignment of the points is identical between two iterations.

Random value initial cluster assignment

This initialization procedure selects the value of the centroids for the initial clusters by drawing randomly from the point pool *P*. Though commonly used, this method breaks one of the important rules for our desired clustering method: the resulting outcome clusters, after the K-means clustering process, are highly dependent on the values of the initial centroids. As such, the outcome clusters will not necessarily be the same when the method is repeated on the same data. This will make it impossible to fulfill goal number 1 for our desired clustering method. Thus it was decided not to use this approach.

Range based fixed value for initial cluster assignment

When using this initialization procedure we would compute the values for our initial centroids based on the value range of the data that is about to be clustered. As such, the outcome from two runs would always be the same when applied on the same data.

We divided the value range of the data points with the desired number of clusters *K* to find the value difference between two centroids:

$$diff = \frac{\max(P) - \min(P)}{K}.$$

We did not start by placing the first centroid at the value min(*P*) as it each centroid should cover the same range of the full value range. As such we computed the value of the initial centroids using the following equation:

$$c_k = (k - 0.5) * diff + \min(P), \text{ where } k = \{1,2, \dots, K\}.$$

This approach fulfilled a lot of our desired goals (1, 3 and 5) and was guaranteed to provide the same number of clusters within subjects which made it possible to compare the clusters spatially location. We needed to test the method to see if it would fulfill goal 2 and 4 which both ties into selecting *K* on beforehand.

Choosing the number of clusters

The K-means method requires the number of initial/final clusters has to be specified before the clustering process can begin (Hartigan and Wong, 1979, Maulik and Bandyopadhyay, 2002). As there is no perfect theoretical way to determine this number, we instead had to apply a randomized (Monte Carlo) approach and investigate what the "natural" number of clusters in our PPT data was.

We did this by using the K-means clustering algorithm with random values for the initial cluster assignment while increasing the number of clusters on the PPT data for every subject. Even though, the outcome of a single run would be affected by the randomness in selecting the initial centroids, it was believed that by running a large number of runs (1000) on each data set the "natural" number of clusters in the data will show its prevalence. In fact, it is this randomness that is necessary to apply the Monte Carlo approach.

To evaluate the best number of clusters we looked at cluster repeatability. That meant how many times out of the total number of runs does the same final point to cluster assignment appear. The assignment then scored based on how many times this particular assignment reappear, best possible score being 100% meaning appearing 1000 times out of 1000 runs. It was to be expected that more than one final cluster assignment would score appearance, but only the one with the highest value was evaluated. The idea was that the number of initial clusters that scored the highest amount of appearance would indicate that this number of clusters was the best for clustering our data. The randomness factor also meant we could expect empty clusters when clustering with any number higher than 1. This probability increased with the number of initial clusters.

A second way to evaluate the number of initial cluster was to look at the ratio between the variance within the clusters (computed by sum of squares for each cluster: $SS_{cluster}$) and the total variance of the data (computed as: SS_{total}). As we wanted to know how much of the total variance that was explained by the clusters we computed the R^2 value between the two:

$R^2 = 1 - \frac{\sum_{j=1}^{K}\sum_{i=1}^{n_j}(p_{j,i}-c_j)^2}{\sum_{i=1}^{N}(p_i-\bar{\mu})^2}$, where cj is the centroid value of the cluster that pj,i gets assigned to after the clustering process and $\bar{\mu}$ is the mean of all the data point values.

We used the K-mean clustering with range based fixed value for initial cluster assignment for this evaluation as it possibly limited the number of empty clusters by forcing separation between starting points. Also, it would make the choice of number more relevant for this approach.

Cluster Repeatability and R^2

The following figures show the results of our number of initial clusters evaluation procedures. Figure 6 shows for the Cluster Repeatability for the Monte Carlo approach and Figure 7 show the R^2 results and number of empty clusters when using fixed values for the initial clusters.

It is seen on Figure 6 that there is a clear drop in the generation of similar clusters when increasing the number of initial clusters above one. The drop is most dominant after 3 initial clusters where it quickly falls to a steady state. This indicates that there is the PPT data is largely spread and clustering should not be done with more than a few (2 or 3) clusters.

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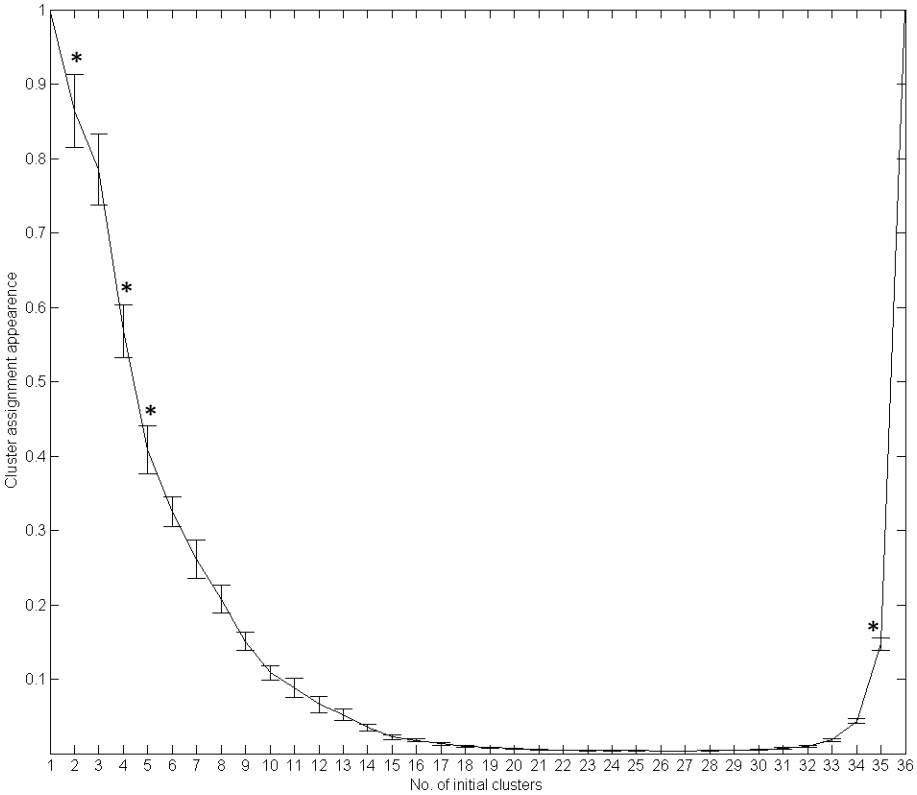

Figure 6. The cluster appearance score (mean±std. error) for the most reappearing cluster combination given a certain number of initial clusters repeated 1000 times on 20 subjects. *: Significant difference between this score value and the previous one (P<0.05).

Only the results for 1 to 10 initial number of clusters are shown in Figure 7 as higher number produces an increasingly amount of empty cluster without improving the R^2 result. At seven initial clusters we saw empty clusters appearing in the final cluster outcome so the initial cluster number should be lower than that. Interestingly, the R^2 value passes 95% of the total variance explained at 3 initial clusters (95.5 ± 0.4%).

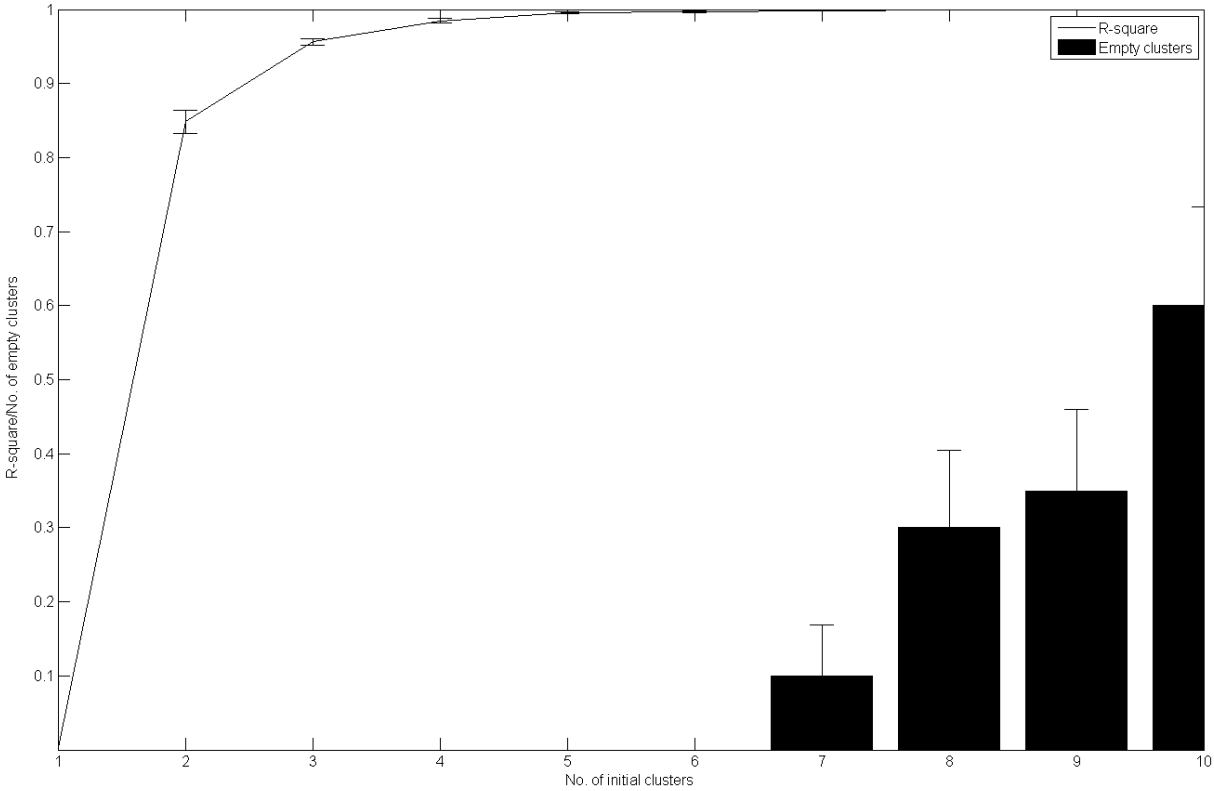

Figure 7. R^2 results and number of empty clusters (mean±std. error) for different number of initial clusters.

These findings showed that there is a natural conglomeration of the data when using 3 initial clusters, and that with the proposed method of selecting the values for the initial clusters based on the PPT data range of the subject there is no chance of ending up with empty clusters. Also, as the clustering process now can be done with fixed parameters that are independent of the individual subject it makes this clustering method fulfill all the goals for our desired clustering method.

Parameters for spatial evaluation

Given that the major goal of the clustering algorithm was to enable separation of groups of people based on overall spatial differences in their PPT distribution it was necessary to select parameters that could provide this information. The first parameter was the R^2 result of the clustering analysis. Having established that among a healthy population and giving three initial clusters R^2 should be around 95%, a significantly lower R^2 value at three initial clusters would strongly indicate that any given subject would have at least one cluster more. This could indicate an area with abnormal sensitivity is present in the muscle.

The other spatial parameters we found useful for evaluation were the final cluster centroid locations on the muscle. We computed these by taking the mean co-ordinate value for all points assigned to each cluster in both the cranial-caudal and medial-lateral direction. Position of these centroids would relate directly to pain sensitivity in certain area of the muscle.

Final clustering outcome

When combining the use of the K-means clustering method and of the parameters for spatial evaluation of the PPT data, it appeared that three clusters positions are differently located in the cranial-caudal direction but not in the medial-lateral, see Table 1. Note that for this comparison of centroid positions, all subjects are set to have the same C7-arcmoin distance (180 mm). Looking at the overall cluster distribution (Figure 8) it is apparent that the points are majorly divided into three separate areas. These areas generally corresponded to the upper, middle and lower sub-divisions of the trapezius muscle. The cluster of the points with lowest mean PPT value were majorly located in the upper part of the muscle, intermediate mean PPT value in middle part of the muscle while

the cluster with highest mean PPT value were located mainly in the lower part of the muscle. Though, due to being located on musculo-tendinous tissue, which is known to have lower pain sensitivity than muscle tissue (Andersen et al., 2006), points 15, 20 and 24 were assignment to the cluster with highest mean PPT.

In general, the topographical distribution is in line with findings showing clear physical differences between muscle fiber attachment and direction throughout the trapezius muscle separating it into 3 sub-divisions (Johnson et al., 1994); an upper part (from fascicle SNL to C6), a middle part (from fascicle C7 to T1) and a lower part (from fascicle T2 to T12). Electromyography studies have shown independent motor control activation of the trapezius sub-divisions (Mathiassen and Aminoff, 1997, Holtermann et al., 2009), which combined with our findings suggest a sensory and neuromuscular partitioning of the muscle (Windhorst et al., 1989). When clustering the PPT data based on the PPT values alone, we clearly find the upper part of the muscle being the most sensitive to pressure pain stimulation. This is interesting as clinical findings have reported that MSD are most frequently located in the upper part of the shoulder girdle (Punnett and Wegman, 2004, Larsson et al., 2008, Rosendal et al., 2004) and pointing on a connection between areas of low pain threshold in healthy persons being the most prone for MSD.

Cluster	Medial/lateral direction (mm)	Cranial/caudal direction (mm)	Size
1	56±2	265±5 †,Δ	11.4±0.8 •,□
2	58±2	192±4 †,*	16.3±0.8 •,‡
3	62±5	149±8 *,Δ	8.2±0.8 ‡,□

Table 1. Comparison of cluster position in the medial/lateral and cranial/caudal direction as well as cluster size. Values are represented in mean±Standard Error. Cluster 1 is the cluster with lowest mean PPT, cluster 2 has intermediate mean PPT and cluster 3 has the highest mean PPT value. †, *, Δ, •, ‡, □: Significant difference between respective pair of values ($P < 0.001$).

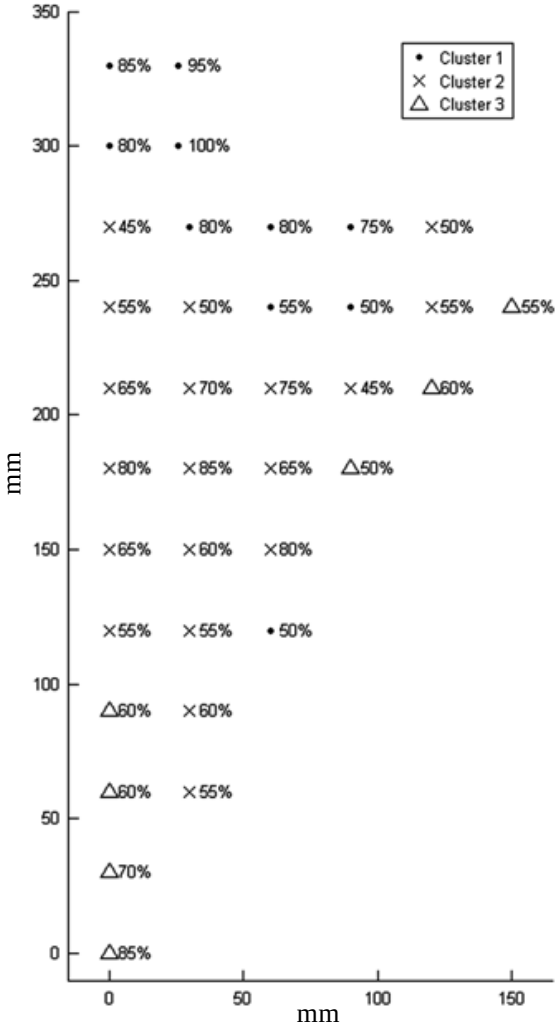

Figure 8. Overall cluster distributions and number of match in % for the dominant trapezius muscle in healthy subjects. The recording points have been marked with a symbol according to which cluster the point most often has been assigned to. •: Cluster of low pressure pain threshold (PPT) values. ×: Cluster of intermediate PPT values. Δ: Cluster of high PPT values.

The method presented clustering method is a new way to enable the identification of abnormal topographical PPT distribution in the shoulder region. Provided a set of PPT measurements from a new subject it is possible to compare the position of the three clusters with those of healthy subject and determine possible difference in the pain distribution and where the reason for this might be located. This can be used diagnostically as a quantitative tool to study PPT maps and to monitor the effect of treatment on musculoskeletal pain or changes.

Spatial pain findings in humans

Men and women

Women have been reported to have generally lower PPTs than men (Chesterton et al., 2003) and while not fully understood (Greenspan et al., 2007) the cause is suggested to involve physical (Cairns, 2007)as well as cultural(Dawson and List, 2009) and physiological factors (Miller and Newton, 2006). Further, the prevalence of MSD accompanied by pain in the back is higher among women than men (Leboeuf-Yde et al., 2009) it is of interest to investigate if there is any spatial difference in PPT between the two genders. Studying healthy (no history of previous neck-shoulder or low back disorders) and young populations (men: 23.4±2.5 years, women: 23.9±3.4 years) would make the findings of this study a good base (obtaining of normal values) or control for other populations.

Visual inspection of the normalized PPT data showed similarities in the pain topography between the two genders for both regions, see Figure 9 and 10, but women showed a general lower tolerance towards pain than men in both the neck-shoulder (328.9±121.6 vs. 357.1±101 kPa) and the low back region (428.2±136.9 vs. 506.1±322.8 kPa). Still, a PPT study in the masseter muscle indicates no difference between genders suggestion that these differences may be muscle specific (Svensson et al., 2003). The relative difference in muscle size to the PPT prope might have had an effect as it has been shown that larger propes invoke a smaller pain repsonce (Nie et al., 2009). As women in general have smaller muscles than men this could be a contributing factor. Further, a higher degree of temporal summation has been reported in women (Ge et al., 2005), which could cause increased pain integration during the pressure stimulus resulting in lower PPTs. It is most likely that peripheral and central mechanisms are responsible for gender differences in PPT.

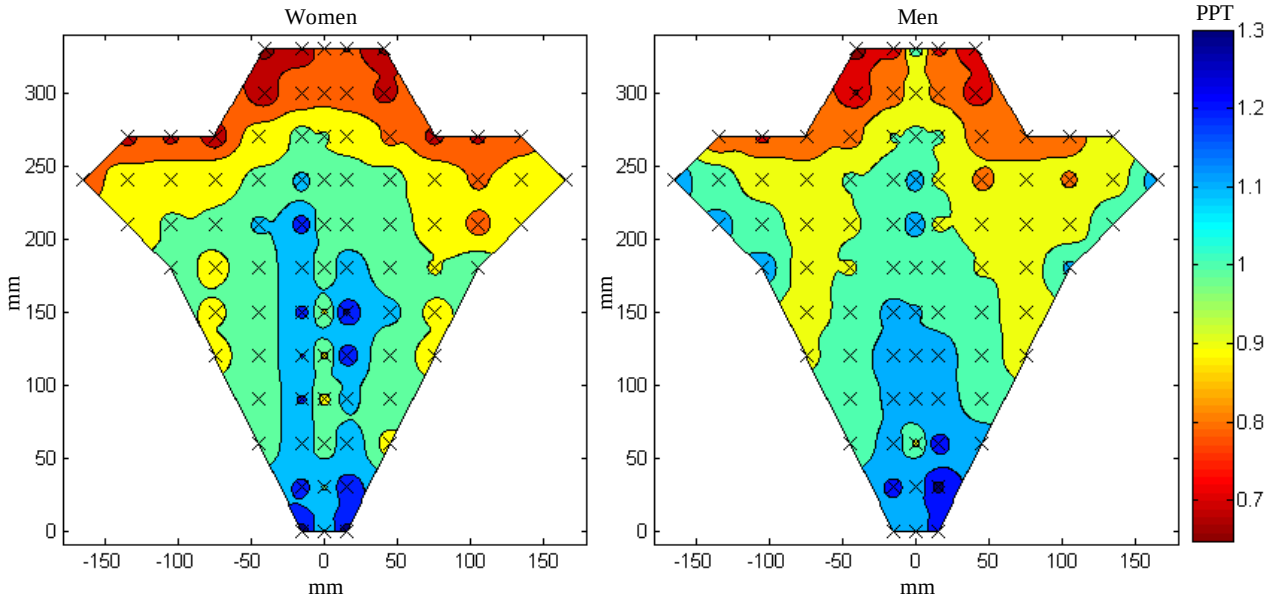

Figure 9. Normalized pressure pain threshold maps for the neck-shoulder region for healthy women (N=11) and men (N=11).

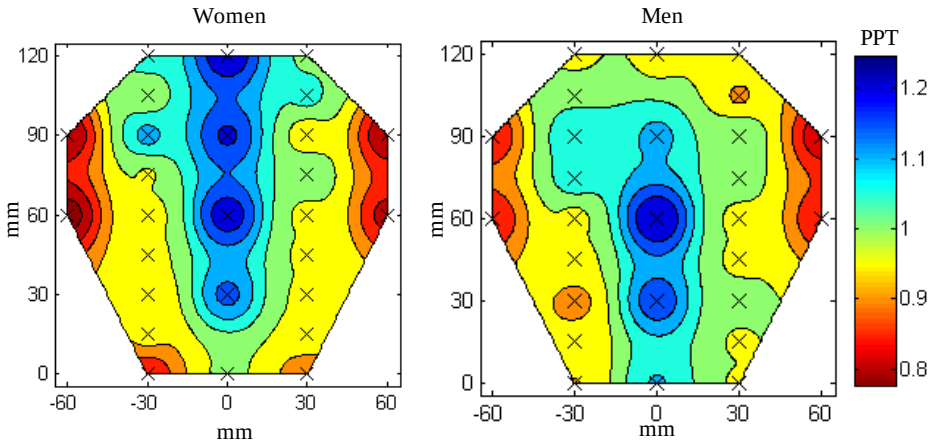

Figure 10. Normalized pressure pain threshold maps for the low back region for healthy women (N=11) and men (N=11).

The spatial proportions of the maps for both regions were equal between genders and symmetrical along the spine. For both, the upper part of trapezius was found to be the most sensitive (295.2±95.9 kPa) and the lower part the least (373.0±121.1 kPa). This difference between subdivisions could be related to the number of points located on muscle belly and musculo-tendinous tissue in each division as the latter type of tissue in general is less sensitive to pressure pain than the former (Andersen et al., 2006, Nie et al., 2005). In the neck-shoulder region, the spine does not show distinct difference in sensitivity compared with the muscle but a gradual increase in PPT in the caudal direction is observed (O'Neill et al., 2009). In the low back region, the spinal processes showed to be far less sensitive than the muscle parts and, in accordance with previous studies in this region (Hirayama et al., 2006), the edge of the erector spina muscles were found to be the most sensitive locations.

The presented findings show for the first time the spatial distribution of pressure pain sensitivity in cervico-thoracic and lumbar regions among both healthy men and women. This investigation provides the basis for further clinical studies on e.g. chronic low back pain. The study also confirmed that women are generally more sensitive than men to pressure pain stimulation in both the cervico-thoracic and the lumbar regions with no gender or side differences in normalized pressure pain maps.

Delayed onset muscle soreness

An endogenous temporary model of muscle hyperalgesia and allodynia that can be used to mimic chronic pain condition in a controlled fashion is to use delayed onset muscle soreness (DOMS) in healthy subjects (Svensson et al., 1997, Frey Law et al., 2008). Muscle hyperalgesia is defined as an increased response to a stimulus which is normally painful, while allodynia is defined as pain due to a stimulus which does not normally provoke pain (Merskey and Bogduk, 1994). Due to the overlap of these two terms in regard to soreness developed due to eccentric exercise the term hyperalgesia is used to describe increased sensitivity towards pressure pain stimulation. DOMS is characterized by mechanical muscle hyperalgesia, occasional resting pain, and altered motor control (Nie et al., 2005, Bajaj et al., 2002, Kawczynski et al., 2007, Samani et al., 2009). There are contradictory results with respect to the location of the most sensitive part of the muscle. Some have found the musculotendinous junction to be more sensitized towards pain by strenuous eccentric exercise compared with the muscle belly (Fridén and Lieber, 2001, Newham et al., 1983, Cleak and Eston, 1992) while other studies have reported the opposite (Andersen et al., 2006, Nie et al., 2005, Weerakkody et al., 2003, Gibson et al., 2006, Weerakkody et al., 2001). With these conflicting findings we conducted a study aiming at investigating the spatial distribution of hyperalgesia within an affected muscle after DOMS using PPT mapping. The focus was only on the trapezius muscle due to the ease of exercising this muscle and its involvement in many neck-shoulder chronic pain conditions (Bernard, 1997). The PPT mapping was done on two groups of subjects; one who did the eccentric exercise and one that did not. This was done to provide a comparable control group for the DOMS group while at the same time making it possible to further validate the repeatability of PPT measurements by comparing measurements taken with half an hour interval and 24 hour interval in a group of healthy subjects.

The decrease of pressure pain sensitivity from before the exercise, to immediately after, to 24 hour after was widely different between the two groups, see Figure 11, with the group doing exercise decreasing it significantly from before (234.3±3.5 kPa) to 24 hour after the exercise (174.2±3.5 kPa). This was a general development of hyperalgesia as the spatial proportions in the PPT maps were equal before and after the exercise for both groups. The upper part of trapezius was found to be the most sensitive subdivision and the lower part the least for both groups at all three measurements. This is could explain why clinical findings report that MSD in the neck-shoulder region are most frequently located in the upper part of the trapezius muscle (Punnett and Wegman, 2004, Kadi et al., 1998). The topographical extent of hyperalgesia in the trapezius muscle following eccentric exercise was in ad equation with its subdivisions. Together with findings on differentiated electromyographic activity of the trapezius muscle subdivisions showing localized control of fresh and painful muscle (Madeleine et al., 2006, Mathiassen and Aminoff, 1997, Holtermann et al., 2009, Samani et al., 2009)this suggests a correlation between sensory and neuromuscular partitioning (Windhorst et al., 1989).

Changes in PPT due to DOMS over a muscle have been shown to be heterogeneous between sites positioned relatively close to each other (Andersen et al., 2006, Weerakkody et al., 2003, Weerakkody et al., 2001, Hedayatpour et al., 2008). This lack of uniformity in pressure pain sensitivity may be related to the mechanical and metabolic capacity of muscle fibers in producing tension, temperature (Nadel et al., 1972), activation of phospholipase A2 (Palmer et al., 1983), and lipid peroxidation from oxygen radicals (Li and Sakamoto, 1996).

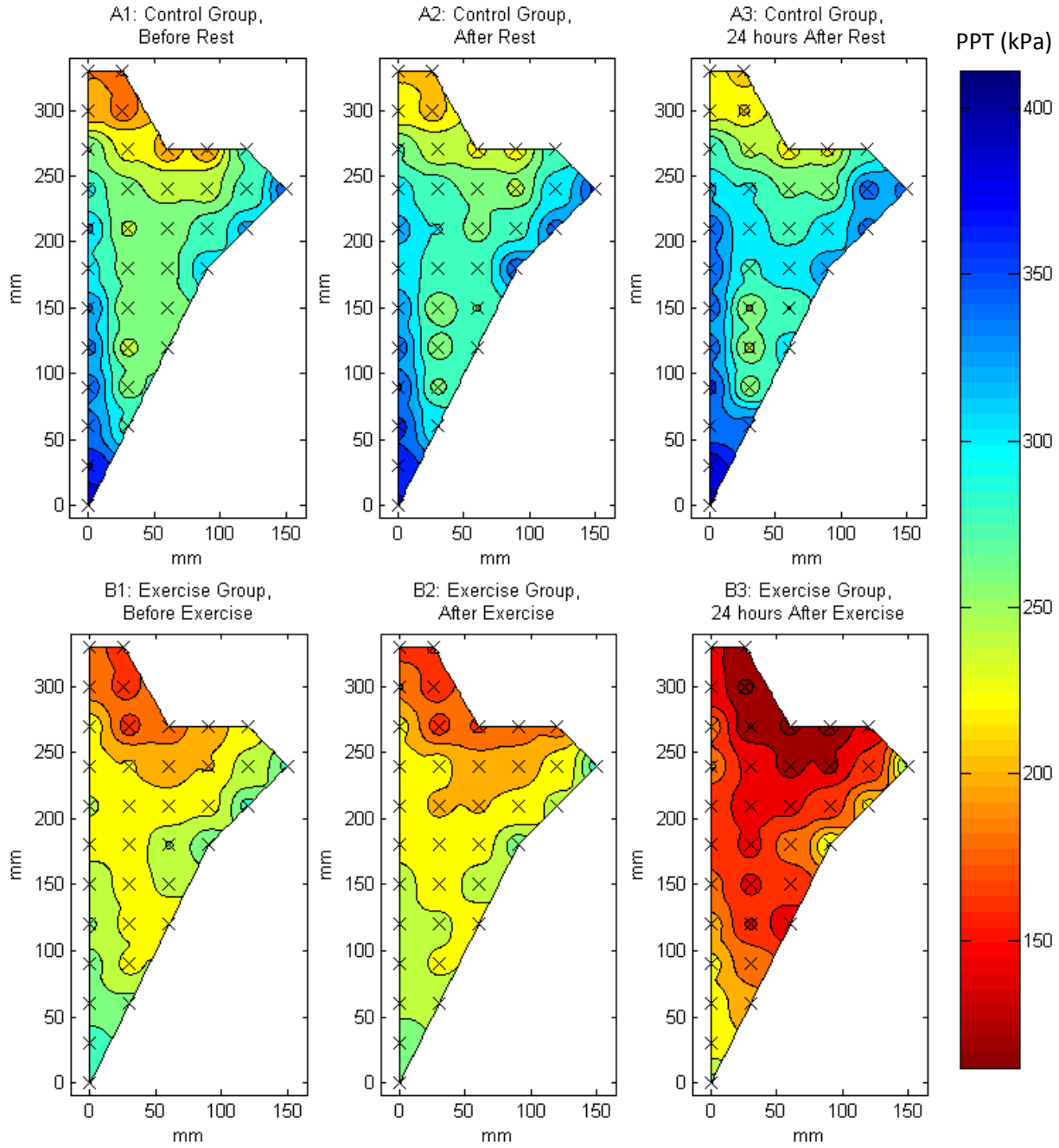

Figure 11. Absolute pressure pain threshold (PPT) maps from the trapezius muscle before (A1), immediately after (A2) and 24 hours after (A3) the rest period for the control group (N=10) and from before (B1), immediately after (B2) and 24 hours after (B3) the eccentric exercise for the exercise group (N=10). Units on axis are in millimeters.

Another interesting issue was the development in musculotendinous located points contra muscle belly points. Before the eccentric exercise, we observed that points located on muscle belly sites when grouped together were more sensitive to pressure pain that points located on musculotendinous sites. Further, the mechanical hyperalgesia elicited by DOMS was heterogeneously distributed over the trapezius muscle, i.e. muscle belly sites became even more sensitive compared with musculotendinous sites. This finding is in agreement with a number of previous studies (Andersen et al., 2006, Nie et al., 2005, Baker et al., 1997, Slater et al., 2003) but contrary to the findings by Newham et al. (1983). The topographical pattern of hyperalgesia is most likely explained by the different extent of discrete damage of eccentric exercise caused to muscle belly and musculotendinous sites. This is corroborated by studies showing heterogeneous distribution of hyperalgesia at muscle level in response to DOMS (Andersen et al., 2006, Nie et al., 2005, Weerakkody et al., 2001, Slater et al., 2003). Spatial difference in sensitivity can also be explained by the thickness of the tissue tested, belly sites having greater thickness than

musculotendinous ones as suggested by Andersen et al. (2006). In parallel, underlying bone structures in the tendon areas can provide increased tissue hardness resulting in general higher PPT scores than muscle belly areas. Further, differences in the density of sensory afferents e.g. groups III and IV afferents among muscle tendon and muscle belly can also explain our results (Andres et al., 1985). The present differences among muscle sites is in line with a study suggesting that the muscle soreness following eccentric exercise is located in the fascia (Malm et al., 2004) in which free nerve endings are found (Yahia et al., 1992). These arguments put together most likely explained the various degree of hyperalgesia observed in muscle belly and musculotendinous sites of the shoulder region.

As demonstrated, the use of high density pressure pain topographical maps of the trapezius muscle enables spatial investigation of muscle hyperalgesia after e.g. DOMS. The areas which initially were the most sensitive to pressure pain were also the areas with the highest development of hyperalgesia. As such, a general heterogeneous development with most pronounced hyperalgesia at muscle belly sites compared with musculotendinous sites. The upper part of the trapezius was more sensitive to pressure than the middle and the lower subdivision both before and after the induced muscle hyperalgesia. For the control group no change between the three measuring session were discovered validating the repeatability of PPT. This study shows how high density pressure pain topographical mappings can be helpful to visualize and track development of hyperalgesia.

Cleaning personnel

Cleaners are well known for having a high prevalence of MSD and sickness absence (Søgaard et al., 2006) and as such be prone to long term sickness absence (LTSA). MSD of the neck-shoulder and low back regions are well acknowledged as the underlying cause of a major fraction of LTSA (Holtermann et al., 2010, Steenstra et al., 2005, Burdorf et al., 1998). It is therefore likely that workers with LTSA may experience more musculoskeletal pain than workers without LTSA. PPT measurements are previously proven relevant in working populations suffering from MSD (Gold et al., 2006, Schenk et al., 2007, Hägg and Åström, 1997, Madeleine et al., 2003, Madeleine et al., 1998).

The PPT mapping showed a clear difference in general PPT levels between cleaners with a history of LTSA (more than ten consecutive sick days reported within the last year) and those without, see Figure 12 and 13. The findings of lower PPT in the neck-shoulder region and the similar tendency in the low back among cleaners with LTSA than among those without lend support to the profound role of MSD as a predictor for future LTSA, previously shown by self-reported pain (Holtermann et al., 2010, Steenstra et al., 2005, Burdorf et al., 1998). The observed lower PPT among cleaners with previous LTSA is most likely a factor increasing the risk for future MSD, as the observed hyperalgesia may expand to other remote areas as often reported in patients with neck-shoulder or low back pain (Schenk et al., 2007, Madeleine et al., 1998). Moreover, neck-shoulder pain intensity and physically heavy work are documented to enhance the risk for LTSA among workers with MSD (Holtermann et al., 2010). The cleaners with low PPT and MSD having high physical work demands are therefore likely to be at high risk also for future LTSA and to develop work-related MSD (Madeleine et al., 2003).

Like for the studies III and IV, there were no apparent differences in the spatial proportions of the PPT map in the neck-shoulder and low back between the groups. In both groups, the upper part of trapezius was found to the most sensitive area and the lower part of trapezius the least sensitive to pressure. The sensitivity of mechanical pressure was therefore generally higher in the entire trapezius muscle among cleaners with than without LTSA. Interestingly, hyperalgesia was most likely not due to a central sensitization mechanism as seen in patient populations like fibromyalgia and whiplash (e.g. Banic et al 2004) as no changes were found at a control location on the tibialis anterior. Thus, initiatives aiming at reducing the intensity of MSD among workers may be a good prevention strategy for reducing the high prevalence of LTSA in this group of workers.

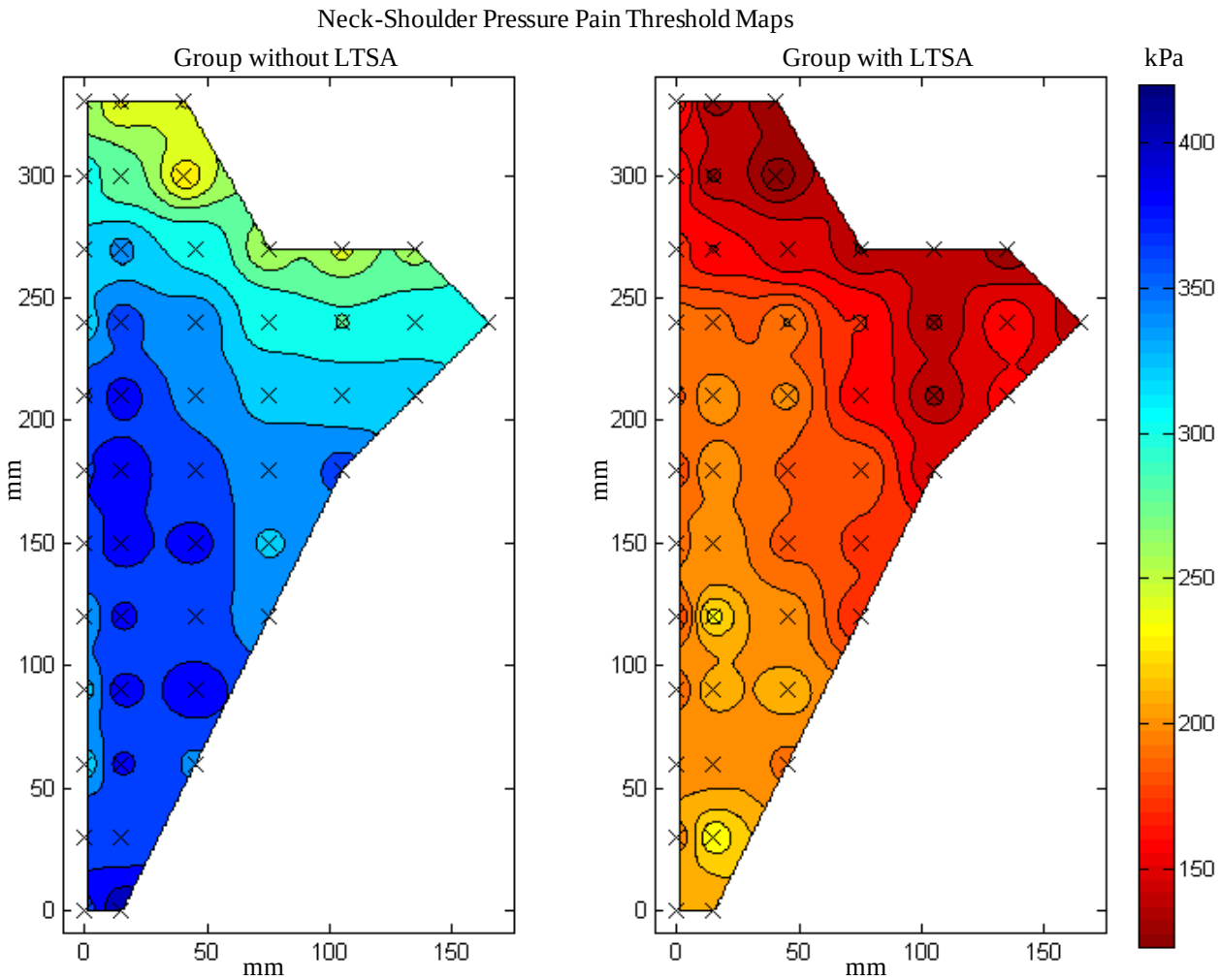

Figure 12. Absolute pressure pain threshold maps of the neck-shoulder region for cleaners with <10 or >10 consecutive sick days reported within last year (respectively, group without LTSA, N=21 and group with LTSA, N=5). Note the overall lower pressure pain threshold for cleaners with >10 consecutive sick days.

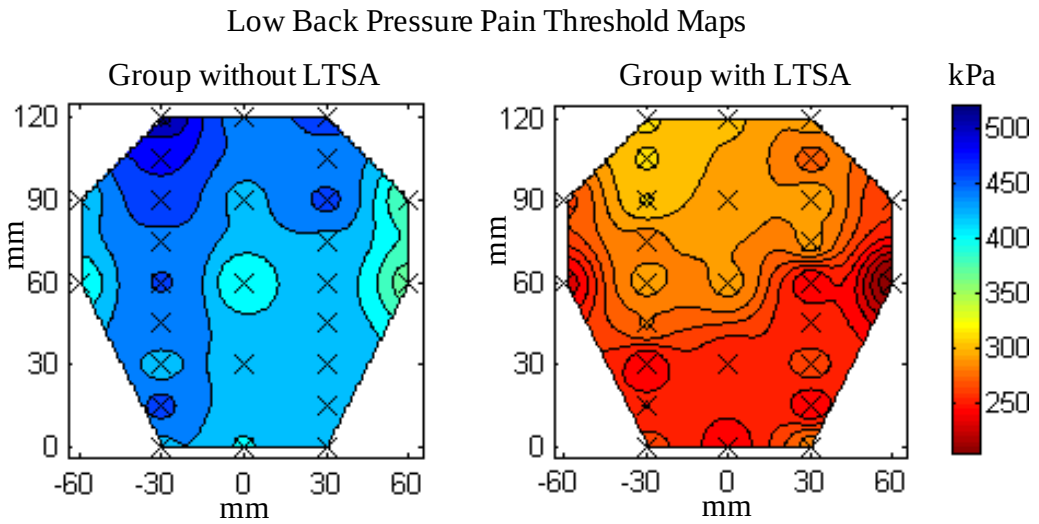

Figure 13. Absolute pressure pain threshold maps of the low back region for cleaners with <10 or >10 consecutive sick days reported within last year (respectively, group without LTSA, N=21 and group with LTSA, N=5). Note the trend towards lower pressure pain threshold for cleaners with >10 consecutive sick days.

Both groups were characterized by similar topography of the PPT in the neck-shoulder and low back. These are novel findings showing how PPT mapping reveals potential risk factors for LTSA. This demonstrates the clinical possibilities of the mapping method as diagnostic tool while at the same time demonstrating that areas of high pressure pain sensitivity in healthy subjects are those at greatest risk for developing MSD.

Conclusion

All studies using the division of the trapezius based on the three anatomical subdivisions showed the same results regarding the sensitivity order; the upper part is the most sensitive while the lower part is the least. This order does not change between sides or genders. When performing an eccentric exercise which involves movement of only a part of the muscle the soreness spreads throughout the muscle keeping the sensitivity order still. In cleaners with reported LTSA the order was the same too. These results point towards a location dependent pain perception throughout a muscle with general low PPT considered as risk zones for development of MSD. The connection between anatomical and functional subdivisions in the trapezius muscle was confirmed by the PPT maps. More studies investigating sensory-motor functional partitioning are warranted in the neck-shoulder region.

The clustering method is provided as a quantitative tool that can be used to track spatial abnormalities/changes in pain sensitivity and monitor the effect of treatment on musculoskeletal pain. Finally, the presented studies show for the first time the spatial distribution of pressure pain sensitivity in the neck-shoulder and low back regions in healthy subjects as well in experimental and clinical pain groups. PPT has been proven to be a valid measurement for visualizing pain topography which will allow for a wide range of further studies on MSD.

Abstract

Musculoskeletal disorders (MSD) are a major problem resulting in loss of work ability for the individuals and are an economical burden for society. While methods like magnetic resonance imaging and ultrasound are adequate for visualizing bones and muscles, these methods do not provide any information of the changes in deep structure sensitivity to pain. A new method aiming at directly measure and visualize pain spatial changes can be a way to localize painful areas.

The pressure pain threshold (PPT) is the minimum required mechanical pressure at which a person feels a sensation of pain from a certain location. This provides a semi-objective measurement of pain providing a quantifiable value of pain sensitivity. By covering the surface of a muscle using a high number of measurement locations it becomes possible to generate a PPT map visualizing the pain topography of the muscle. The presented studies (I-II) cover the development of methods to generate the PPT maps as well as a way to cluster the PPT measurements to investigate possible distinct areas of sensitivity within the same muscle. Further, healthy men and women have been mapped to investigate differences in PPT and topography between genders in the low back and neck-shoulder region (III). Further, delayed muscle onset soreness was induced in the shoulder region among healthy volunteers to assess spatial changes in presence of soreness (IV). PPT maps showed a lowering of PPT in presence of pain/soreness in line with anatomical (sub-divisions of the trapezius muscle) and clinical findings (upper trapezius being more sensitive). Finally, two groups of cleaners (V) were investigated with respect to sickness leave (one with reported sickness leave and one without). The PPT maps showed a clear difference in general PPT levels between cleaners with sickness leave compared with without sickness leave

The developed methods in the low back and neck-shoulder region have proved to be effective in visualizing pain topographic information (I-V). Further studies using PPT mapping are warranted in patient populations. These methods can be used as a way to assess the effectiveness of treatment or prevention of MSD.

Dansk Resumé

Smertefulde lidelser i muskler og knogler er et voksende problem resulterende i tab af arbejdsevne for den enkelte person og er en samfundsøkonomisk byrde. Metoder såsom magnetiskresonans- og ultralydsscanning er tilstrækkelige til at visualisere strukturer i knogler og muskler, men giver ikke nogen oplysninger om ændringer i følsomhed overfor smerte. En ny metode med henblik på at måle og visualisere smerteudbredelse kan være en måde at lokalisere og afgrænse smertefulde områder.

Tryksmertegrænsen er det minimum af mekanisk påført tryk hvorved en person føler den første fornemmelse af smerte. Dette giver en semi-objektiv måling af smerte og en kvantificerbar værdi af smertefølsomhed. Ved at dække overfladen af en muskel med et stort antal målinger, bliver det muligt at generere et smertekort der viser smertetopografien af musklen. De første af de præsenterede studier (I-II) omfatter udvikling af metoder til at generere smertekort samt en måde at gruppere smertemålinger. Dette gøres for at undersøge mulige separate områder af følsomhed i samme muskel. Raske mænd og kvinder er blevet undersøgt i den nedre del af ryggen og nakke-skulder regionen for forskelle i smertetærskler og topografi mellem kønnene (III). En gruppe raske frivillige fik induceret midlertidig ømhed i skuldermusklen for at se om dette ændre smertetopografien (IV). Smertekortene viste en sænkning af smertetærsklen. Desuden viste der sig separation af smertefølsomhed i overensstemmelse med de anatomiske sub-divisioner af trapezius musklen og tidligere kliniske fund (øverste del af trapezius blev fundet mest følsom). Endelig blev to grupper af rengøringsassistenter (V) undersøgt med hensyn til sygeorlov (en gruppe med rapporteret sygeorlov og en uden). Smertekortene viste en klar forskel i smertetærskler mellem de to grupper, lavest hos gruppen med sygeorlov.

De udviklede metoder har vist sig at være effektive til at vise smertetopografi (I-V). Dette kan benyttes til nye undersøgelser med tryksmertekortlægning af smerteudbredelse i patientpopulationer. Metoderne kan bruges som en måde at vurdere effektiviteten af behandling eller til forebyggelse af lidelser i muskler og knogler.

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