A comparative study of music and soft tissue massage on heart rate variability.

James Morgan

A research project submitted in partial fulfilment of the requirements for the degree of Master of Osteopathy at Unitec New Zealand 2010
Abstract

**Background and Objectives:** Music and massage are considered by many to promote relaxation. However there appears to be insufficient supporting evidence to validate physiological changes that may reflect relaxation. This study examined the physiological effects of music and massage as indicated by heart rate variability. The low to high frequency (LF/HF) ratio component of heart rate variability was used to indicate changes in the autonomic nervous system. The autonomic nervous system controls many functions of health in the body and is divided into the sympathetic nervous system (responsible for increasing heart rate), and the parasympathetic nervous system (responsible for decreasing heart rate). An increase in LF/HF is believed to indicate sympathetic nervous system activity, or sympathetic dominance, while a decrease indicates parasympathetic activity. Therefore a decrease in LF/HF would be expected should physiological relaxation occur.

**Methods:** 16 male subjects attended four sessions in random order involving music, massage, music and massage combined, and a control. Each intervention lasted 10 minutes with a 10 minute pre- and post-intervention washout time. Heart rate data were recorded for all sessions, and heart rate variability was analysed.

**Results:** There was no statistically significant difference in LF/HF between the four intervention studies. Additionally for each of the intervention studies, there was no statistically significant difference between pre-intervention, intervention and post-intervention.

However, in a sub-group of subjects that had low heart rates (less than 60 beats per minute) an increase in LF/HF was found (p<0.05) in response to massage, though not in response to music, music and massage combined or the control intervention.

**Conclusion:** These results suggest no physiological effect of music or massage interventions occurs in the autonomic nervous system as indicated by heart rate variability. In the sub-group of low heart rate responders (n = 5), an increase in LF/HF indicating sympathetic dominance in response to massage was found. This finding may suggest that massage in fact has a stimulating (sympathetic) rather than relaxing (parasympathetic) effect on subjects with low heart rates. This same effect however was not shown by these subjects when massage was combined with music.
Declaration

Name of candidate: James Morgan

This Research Project entitled “A comparative study of music and soft tissue massage on heart rate variability” is submitted in partial fulfilment for the requirements for the Unitec degree of Master of Osteopathy.

CANDIDATE’S DECLARATION

I confirm that:

• This Research Project represents my own work;
• The contribution of supervisors and others to this work was consistent with the Unitec Regulations and Policies.
• Research for this work has been conducted in accordance with the Unitec Research Ethics Committee Policy and Procedures, and has fulfilled any requirements set for this project by the Unitec Research Ethics Committee.

Research Ethics Committee Approval Number: 2009-936

Candidate Signature: …………………………………………………..Date: …………………

Student number: 1109705
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<tbody>
<tr>
<td>Ach</td>
<td>Acetylcholine</td>
</tr>
<tr>
<td>ANOVA</td>
<td>Analysis of variance</td>
</tr>
<tr>
<td>AOA</td>
<td>American Osteopathic Association</td>
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<tr>
<td>BP</td>
<td>Blood pressure</td>
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<td>bpm</td>
<td>Beats per minute</td>
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<tr>
<td>DBP</td>
<td>Diastolic blood pressure</td>
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<tr>
<td>ECG</td>
<td>Electrocardiogram</td>
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<tr>
<td>EEG</td>
<td>Electroencephalogram</td>
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<td>GSR</td>
<td>Galvanic skin response</td>
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<td>HF</td>
<td>High frequency</td>
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<tr>
<td>HRV</td>
<td>Heart rate variability</td>
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<td>Hz</td>
<td>Hertz</td>
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<td>LF</td>
<td>Low frequency</td>
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<td>LF/HF</td>
<td>Low frequency to high frequency ratio</td>
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<td>MT</td>
<td>Music therapy</td>
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<td>OMT</td>
<td>Osteopathic manipulative treatment</td>
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<td>R-R</td>
<td>R to R</td>
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<td>RR</td>
<td>Respiratory rate</td>
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<td>RSA</td>
<td>Respiratory sinus arrhythmia</td>
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<tr>
<td>SBP</td>
<td>Systolic blood pressure</td>
</tr>
<tr>
<td>TES</td>
<td>Thoracic erector spinae</td>
</tr>
<tr>
<td>UREC</td>
<td>Unitec Research Ethics Committee</td>
</tr>
<tr>
<td>VAS</td>
<td>Visual analogue scale</td>
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<td>VLF</td>
<td>Very low frequency</td>
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1.0 Aim

The aim of this research was to perform a quantitative study to compare the physiological effects of self-selected music and Osteopathic soft tissue massage technique (massage) on Heart Rate Variability (HRV), and thus by extension on the Autonomic Nervous System (ANS) in healthy young adult males at rest.

1.1 The Objectives

The objectives of the study were:
- To measure the effect of self-selected music on HRV.
- To measure the effect of massage on HRV.
- To measure the effect of music and massage combined on HRV.
- To measure the effect of no intervention (control) on HRV

1.1.1 If an Effect Found

- To compare any measured effects on HRV of music and massage.
- To determine if there is an additive effect of music and soft tissue massage on HRV.

1.2 Hypothesis

It was believed that the effect of interventions would produce a physiological relaxation response as indicated by an increase in parasympathetic activity and a decrease in sympathetic activity.

It was hypothesised that the effect of soft tissue massage on HRV would be a greater reduction in sympathetic activity than the effect of music, and that the combined effects of both interventions together may be additive.
2.0 Literature Review

2.1 Introduction (Overview)

Certain types of music are considered to promote relaxation (Chlan, 1998; Okada, Kurita, Takase, Otsuka, Kodani, Kusama, Atarashi & Mizuno, 2009) however the primary focus of research appears to relate to the subjective responses of the subject, such as pleasure and changes in mood (Urakawa & Yokoyama, 2005) rather than objective physiological responses. There are conflicting reports as to the effect of music in the literature, relating to the exact circumstances and variables which affect the body’s response to music, such as the type of music (McCraty, Barrios-Choplin, Atkinson & Tomasino, 1998; Nater, Abbruzzese, Krebs & Ehlert, 2006; Umerura & Honda, 1998) and the subject’s involvement in the music (Sudheesh & Joseph, 2000). Music was found to decrease sympathetic nervous system (SNS) and increase parasympathetic nervous system (PNS) activity as measured by heart rate (HR), blood pressure (BP), and heart rate variability (HRV), indicating physiological relaxation (Larsen & Galletly, 2005; Okada et al., 2009; Smolen, Topp & Singer, 2002; Walsh, 2000). However SNS activity and HR were also found to be increased after exercise in subjects who listened to preferred music (Urakawa & Yokoyama, 2005). Additional variables found to affect the human physiological response to music are respiratory rhythm (Larsen & Galletly, 2005) and gender (Nater et al., 2006).

A feeling of relaxation is often induced by massage (Moyer, Rounds & Hannum, 2004), however there appears to be proportionately more qualitative research on how a subject feels than quantitative research on the physiological response of the body. Further, studies which incorporate both quantitative measures as well as qualitative measures tend to focus more on the qualitative and/or non-physiological measures, such as pain and anxiety visual analogue scales (VAS), which although are quantitatively analysed, are subjective and not physiological measures (Jane, Wilkie, Gallucci, Beaton & Huang, 2009). Of studies which related to physiological findings, a meta-analysis reported significant reductions in HR and respiratory rate (RR) promoting biological relaxation (Labyak & Metzger, 1997), and further results indicated massage had a relaxing effect as measured by decreased systolic blood pressure (SBP), diastolic blood pressure (DBP), and HR (Mok & Woo, 2004). These findings however are in conflict with studies which reported both increased and decreased arousal as indicated by HR and electroencephalogram (EEG) activity depending on the type (light, moderate or
vibratory) of massage (Diego, Field, Sanders & Hernandez-Reif, 2004), and increased SNS activity following massage (Hatayama, Kitamura, Tamura, Nagano & Ohnuki, 2008). Although subjective relaxation seems to be a common finding, physiological relaxation responses appear to be dependent on the type of massage.

The use of Osteopathic manipulative treatment techniques (OMT), such as high velocity low amplitude manipulation and soft tissue, cranial, articulation and muscle energy techniques, is one of the defining factors which distinguishes Osteopaths from other medical professions (Johnson & Kurtz, 2003; Ray, Cohen & Buser, 2004). These techniques are thus an important foundation to Osteopathic practice.

Osteopathy utilises soft tissue massage as a treatment therapy, to alter muscle tonicity and improve blood flow, and generally to improve physical health. Osteopathic soft tissue massage technique is one of the most commonly used techniques in Osteopathic practice. In a national mail survey of 3000 randomly selected Osteopaths from the American Osteopathic Association (AOA), from which 955 questionnaires were used for analysis, soft tissue was rated the most used technique from a list of eleven different OMT (Johnson & Kurtz, 2003). These techniques are all designed to improve the physiological function of the patient by treating somatic dysfunction and impaired function through the musculoskeletal system, as well as associated vascular, lymphatic and neural structures. Improvement of function is generally measured through quality of life measures such as pain reduction and increased range of motion, as well as other measures such as increased blood flow and improved neurovascular and lymphatic function (Johnson & Kurtz, 2003). In another study on the use of OMT in emergency medicine in which over 4000 surveys were sent out to members and non-members of the AOA, and over 900 returned, it was reported that soft tissue treatment was one of the top three techniques most often used, with over 80% of respondents indicating frequent (daily/weekly) use of soft tissue treatment (Ray et al., 2004).

One example of the use of Osteopathic soft tissue techniques is in the treatment of piriformis syndrome, a condition involving inflammation of the piriformis usually due to micro or macro trauma to the buttocks (Boyajian-O’Neill, McClain, Coleman & Thomas, 2008). In this condition treatment is aimed to stretch the piriformis muscle to restore normal range of motion, decrease pain and eliminate symptoms. OMT thus aims to reduce tension in muscles. Studies
outside of the field of Osteopathy also concur that soft tissue massage improves blood flow and physical health (Durkin, Harvey, Hughson & Callaghan, 2006; Mars, Maharaj & Tufts, 2005; Weerapong, Hume & Kolt, 2005).

Massage therapists also use massage to promote relaxation, and some use music as an aid to induce relaxation (Eidlitz, 2006). As highlighted by the lack of certainty and inconsistencies in music and massage research, both these interventions require further research to explore their use in the medical field for altering physiological states to promote relaxation and improve health.

HRV is a well known and widely used measurement in health research as it is easily and non-invasively measured and assessed and through analysis and interpretation provides information about an individual’s state of health (Kobayashi, Ishibashi & Noguchi, 1999; MacArthur & MacArthur, 2000; Malik, 1996; Rosenwinkel, Bloomfield, Arwady & Goldsmith, 2001). HRV is believed to provide information on the level of stimulation and changes to a subject’s ANS (Berntson, Bigger, Eckberg, Grossman, Kaufmann, Malik, Nagaraja, Porges, Saul, Stone & VanDerMolen, 1997; Malik, 1996).

This research paper explores the use HRV as a quantitative measure of physiological change in subjects who undergo music and massage interventions.

### 2.2 The Autonomic Nervous System

The autonomic nervous system (ANS), one of the two main divisions of the nervous system, regulates the activity of cardiac muscle, smooth muscle and glands (Andreoli, 2000). The ANS is divided up into the sympathetic nervous system (SNS) and parasympathetic nervous system (PNS) (Standring, 2005). The two systems generally have opposing actions with the SNS stimulating the heart and inhibiting the digestive system, while the PNS increases digestion and inhibits the heart, promoting relaxation (Berntson et al., 1997).

It is well recognised that these two systems have complex interactions, and that the SNS and PNS cannot be viewed in isolation. An example of this is that PNS influences on HR were found to be so much greater than SNS effects that even a complete sympathetic blockade
may not affect HR (Levy, 1971). For this reason the PNS and SNS cannot be examined separately, as major effects on one may have a greater or lesser effect on the other, depending on the state of activation or resting tone in the system. Thus exploration of the ANS must consider the balance between the PNS and SNS as well as the individual changes.

The parasympathetic influence on HR is mediated through the release of the neurotransmitter acetylcholine (ACh) by the vagus nerve (Malik, 1996), and as such the terms vagus and vagal are synonymous with the parasympathetic influence on the heart. Vagal tone is dominant on the heart at the atrial level during rest (Ingemansson, Holm & Olsson, 1998).

Fibres from the ANS innervate involuntary (smooth) muscle throughout the body, as well as the heart muscle, thus affecting the lungs and respiration, the heart – including HR and BP, digestion and sweating among other systems (Moore & Dalley, 1999). The effects of the ANS are so far reaching as to make the ANS an important determinant of health, as any influence which affects the ANS will in turn affect the systems innervated by the ANS, causing changes in HR, sweating, RR and so on. In turn, these easily measured physiological variables give insight as to the state of an individual’s ANS.

### 2.3 Heart Rate Variability

#### 2.3.1 Definition, Origins and Use of HRV

Whereas HR is a simple and truthful measure of load on the body (López, Terrados, García & Pérez, 2008) and is generally measured on a per minute basis, the time interval between beats is not necessarily consistent, varying between 10-30% even when the HR remains the same (Kobayashi et al., 1999). HRV is the term used to describe this beat-to-beat alteration in HR (MacArthur & MacArthur, 2000). HRV can also be described as oscillations in consecutive cardiac cycles (Malik, 1996). Other terms such as cycle length variability, heart period variability, R-R (the time between heart beats) variability, and R-R interval tachogram (Malik, 1996) are synonymous with HRV.

HRV first came into clinical use in 1965 when it was noticed that foetal distress was preceded by changes in R-R intervals before any change was noticed in HR itself (Malik, 1996), and in the 1980’s certain changes in HRV were found to be a strong predictor of mortality after an
acute myocardial infarction (Malik, 1996). Today measurement of HRV is an important research tool as it is known that duration and type of activity have effects on various measures of HRV. Therefore understanding which activities affect HRV may indicate benefits to the heart and health (Tulppo & Huikuri, 2004).

It has been shown that low HRV has negative prognostic implications in a variety of clinical situations (Goldberger, Challapalli, Tung, Parker & Kadish, 2001). Reduction of HRV has been reported in several cardiological and noncardiological diseases, and depressed HRV can be used as a predictor of risk after acute myocardial infarction and as an early warning sign of diabetic neuropathy (Marek, 1996). This suggests that interventions that affect HRV will also affect the health of the individual, especially the heart. Generally the higher the average HR the lower the HRV (Tulppo & Huikuri, 2004). A rapidly beating heart correlates with stress (physical or emotional) and a lower HRV, and is shown to be linked to decreased health and higher mortality. Conversely, a lower HR with higher HRV (corresponding to states of relaxation) is healthier. Put another way, a high HRV is a sign of good adaptability of the ANS, while a low HRV can indicate poor adaptability (Pumptra, Howorka, Groves, Chester & Nolan, 2002).

Under resting conditions healthy individuals display periodic variations in their HR both from minute to minute as well as from day to day. The minute to minute rhythmic phenomenon is known as respiratory sinus arrhythmia (RSA), and results in cardio-acceleration during inspiration and cardio-deceleration during exhalation (MacArthur & MacArthur, 2000), while the day to day changes are known as circadian rhythm. Environmental factors are of primary importance in defining the circadian pattern of HR (Mulcahy, Keegan, Fingret, Wright, Park, Sparrow, Curcher & Fox, 1990). Thus when considering HRV it is important to remember the effects time of day has on HR.

As with HR, there is substantial variance in HRV in normal individuals, (Goldberger et al., 2001) which makes it potentially difficult to draw comparisons between individuals.

There are many different ways of expressing HRV data, however as long as the data being measured are collected over a window of at least five minutes the various methods are for the most part the same (MacArthur & MacArthur, 2000). A commonly used device used for data
collection is the Polar S810 Heart Rate Monitor which has been shown to be valid (Gamelin, Berthoin & Bosquet, 2006). An alternate way of measuring HR is with the use of a high quality electrocardiogram (ECG). Gamelin et al. (2006) compared the Polar S810 Heart Rate Monitor to an ECG recorder. The results show that in the supine position there is good correlation between the results, supporting the use of the Polar S810 Monitor to measure HR. The comparative ease of the Polar monitor compared to an ECG makes it the preferred choice of measuring device.

2.3.2 HRV and the Autonomic Nervous System

There is a strong link in the literature between HRV and the ANS as it is believed that HRV is an indicator of ANS activity (Niskanen, Tarvainen, Ranta-aho & Karjalainen, 2002; Urakawa & Yokoyama, 2005). HRV analysis is a technique that uses beat to beat variations to indicate the level of activity of the ANS (Lippman, Stein & Lerman, 1993). HR is mainly determined by SNS and PNS activity, and individual differences in HRV partly relate to individual differences in the level of parasympathetic and sympathetic stimulation of the heart (Kristal-Boneh, Raifel, Froom, & Ribak, 1995). Changes in SNS or PNS activity will change HR and also affect HRV. Although there is no simple connection between HRV and HR, in general increased SNS activity (or decreased parasympathetic activity) will increase HR and lower HRV, while increased PNS activity (or reduced sympathetic activity) will lower HR and increase HRV (Clifford, 2002; Tsuji, Venditti, Manders, Evans, Larson, Feldman & Levy, 1996). Reduced HRV is thus used as a marker of reduced vagal activity (MacArthur & MacArthur, 2000).

The most established use of HRV relates diseases and conditions associated with the heart (Pumprla et al., 2002). A reduction of HRV has been reported in myocardial infarction, cardiac transplantation and myocardial dysfunction (Malik, 1996). HRV has also been studied to explore the effect of various stressors on ischemic heart disease patients, as well as asymptomatic workers (Kristal-Boneh, Raifel, Froom & Ribak, 1995). Also some anti-arrhythmic drugs are known to decrease HRV, while beta-blockers cause HRV to increase (Clifford, 2002).

Additionally HRV is affected by exercise training and mental stress (Keenan, 2005; Malik, 1996), lowering under mental load and increasing with regular exercise by increasing vagal
tone (Keenan, 2005; MacArthur & MacArthur, 2000). There is also a link between emotions such as anxiety and hostility and a reduced HRV, demonstrated in a meta-analysis of several studies (MacArthur & MacArthur, 2000). The results from these studies are consistent with the findings that stress causing an increased HR correlates with a reduced HRV and potentially reduced health (Pumprla et al., 2002).

There is also some indication of the use of HRV in other areas of health research such as diabetic neuropathy, and tetraplegia (Malik, 1996) indicating the wide diversity of usefulness of HRV as a research tool in the exploration of human health.

HRV is assessed by either time or frequency domain methods. The time domain method is a direct measure of the normal-to-normal intervals (i.e. intervals between QRS complexes caused by sinus depolarisation) or instantaneous HR (Okada et al., 2009). Although simple, time domain methods do not give any indication of the different parts of the ANS (Malik, 1996), and so is a less useful method than the frequency domain method. The frequency domain method gives information on how power distributes as a function of frequency, and data are divided into nonparametric and parametric spectra, which in most instances give comparable results (Malik, 1996). Within these spectra measurements are divided into several subcategories, including high and low frequencies (HF and LF) as well as very low and ultra low frequencies (VLF and ULF).

The HF (0.18-0.4 Hz) component represents parasympathetic nervous activity (Clifford, 2006; Hatayama et al., 2008; Keenan, 2005; Tochikubo, Ri & Kura, 2006; Urakawa & Yokoyama, 2005). Efferent vagal activity is the major contributor to the HF component of HRV (Keenan, 2005), and as respiration is mediated through the vagal nerve (Clifford, 2006) HF is synchronous with respiration and is identical to RSA (Keenan, 2005; MacArthur & MacArthur, 2000).

The LF (0.04 to 0.15 Hz) component reflects sympathetic activity on the heart (Clifford, 2006; Keenan, 2005) though does appear to be mediated by both the vagus and cardiac sympathetic nerves (MacArthur & MacArthur, 2000; Malik, 1996). An increase in LF has been found in mental stress and similar activities indicating sympathetic activation, meaning that an increase in LF can be taken as a marker of SNS activation (Clifford, 2002).
The VLF and ULF bands are less well understood and may relate to physical activity, circadian patterns and long term regulatory mechanisms (Clifford, 2006).

The balance or ratio between LF and HF (LF/HF) in normalized units represents SNS activity (Hatayama et al., 2008; Tochikubo et al., 2006) and can be used as an index of parasympathetic-sympathetic (sympatho-vagal) balance (Clifford, 2002; Keenan, 2005; Okada et al., 2009), that is the controlled and balanced behaviour of the two branches of the ANS (Malik, 1996). However the exact proportions of contribution to the LF component from the SNS and PNS are still not clearly understood (Clifford, 2002; MacArthur & MacArthur, 2000; Malik, 1996; Urakawa & Yokoyama, 2005) making it difficult to know for certain which system (PNS or SNS) predominately contributes to the change, when a change in LF/HF is found (Clifford, 2002). Despite this uncertainty it is generally considered that an increase in LF/HF represents an increase in SNS activity (Hatayama et al., 2008; Tochikubo et al., 2006; Urakawa & Yokoyama, 2005). Thus changes in a person’s ANS can be measured and analysed quantitatively through the different components of HRV.

It has been noted in the literature that as the parasympathetic effect increases so does HRV until it plateaus, then HRV decreases as the parasympathetic effect continues to increase, (Goldberger et al., 2001). In other words it appears that increased parasympathetic effect can saturate the HRV response.

There are natural changes in the ANS with age, with a progressive decline in both sympathetic and parasympathetic activity (Pumpria et al., 2002). In the same way HRV shows a decrease with age (Aubert & Beckers, 2003; Tsuji et al., 1996), which is attributed to decreased efferent vagal tone (Keenan, 2005) and reduced beta-adrenergic responsiveness (MacArthur & MacArthur, 2000). This means that in an elderly population the usefulness of HRV is decreased, as it is less able to change due to a decreased response in the elderly heart to ANS signals.

Due to these connections between HRV, the heart and the ANS, measurement of HRV can be a strong indication of physiological changes, but also has limitations as mentioned, due to the
number of known factors which affect HRV, including age, activity (physical and mental) and medication.

2.4 Physiological Effects of Music

Music has long been known to affect the human body, with certain sounds influencing health, character, mood and consciousness (Eidlitz, 2006). Listening to music involves complex psychological, emotional, neurological and cardiovascular changes, as well as behavioural modifications of breathing (Bernardi, Porta & Sleight, 2006). Many studies have indicated that music affects the ANS, primarily through the study of HR and HRV, as well as other measures which indicate ANS activity such as galvanic skin response (GSR), RR and BP. Additionally it is proposed that music may affect healing, through vibration and sound (Eidlitz, 2006).

In 1918 it was reported that different types of music could alter HR and BP (Larsen & Galletly, 2005), a finding which has continued to be researched and confirmed, though there is some debate as to whether music causes an increase or decrease in HR (Iñesta et al., 2008; Walsh, 2000). Several studies reported that under various conditions music decreased HR, implying increased PNS activity (Bernardi et al., 2006; Hamel, 2001; Smolen et al., 2002), however an increase in HR was also reported (Bernardi et al., 2006; Urakawa & Yokoyama, 2005) implying increased sympathetic activity.

In general an effect which involves a reduced HR is considered desirable, so research into the effect of music predominantly focuses around the aim to reduce HR. For example, a reduction in HR is desirable in procedures which tend to cause an increase in anxiety and HR, such as catheterisation and colonoscopy. In Hamel's study (2001) music was used in order to try and reduce HR before catheterisation. It was found that subjects who listened to a piece of music with 70-80 beats per minute (bpm) for 20 minutes before catheterisation displayed altered autonomic, immune, endocrine and neuropeptide states compared to the control group that received treatment as usual. The altered systems resulted in decreased HR, lower metabolism and reduced RR (Hamel, 2001). These findings indicated that music caused a physiological relaxation state. Smolen et al. (2002) used similar measures with colonoscopy as the intervention, with similar findings. Their study looked at the effects of music therapy (MT) on physiological (and self reported) signs of anxiety among colonoscopy patients. HR and BP
were measured during the procedure, both of which were reported to be significantly decreased in the experimental group which listened to self-selected music. The measures of anxiety in the study (increased HR and BP) related to heightened activity of the ANS, thus showing that music decreased ANS activity as these measures were decreased in the experimental group compared to the control group (Smolen et al., 2002). Both these studies indicated that physiological measures, predominately HR, which relate to ANS activity, were reduced in subjects who listened to music compared to controls.

Chlan (1998) measured the effect of music on HR and RR and found that a single MT session was effective in promoting cardiac relaxation. An Osteopathic study on the physiological effects of music played during Osteopathic treatment found a decrease in mean and median values of HR and BP of the experimental group (Walsh, 2000).

Although there are many studies focused on reducing HR, there are some authors who have reported possible excitation effects of music. Urakawa and Yokoyama (2005) reported that listening to music resulted in excitation of the SNS as indicated by an increase in RR and HR regardless of the type of music (stimulating or calming). Further HR has been observed to increase in musicians performing in concerts (Iñesta et al., 2008). This increase in HR existed in the performer even when the music being played was known to have a relaxing effect on the listener's HR, indicating that music alone may not be enough to either increase or decrease HR.

Bernardi et al. (2006) compared 12 musicians to age matched controls listening to six different music styles (2 and 4 minute tracks with a randomly inserted 2 minute pause), and found that music had an arousal effect related to the tempo. Although it was found that music can have a relaxing effect, their findings were predominantly of excitation. In their review it was reported that some studies have found that music can lower HR, while their own study found that passive listening to music accelerates RR, increases BP, HR and LF/HF ratio (indicating sympathetic activation) in proportion to tempo, with slower rhythms inducing relaxation. A randomly inserted two minute pause in music induced relaxation greater than the pre-music relaxation silence and/or listening to music (Bernardi et al., 2006). So although the authors found that music generally caused excitation, they reported that alternating faster and slower
rhythms and pauses may be used to induce relaxation and reduce sympathetic activity (Bernardi et al., 2006).

As with the conflicting HR findings, so there is some uncertainty relating to music effects on RR, with both a decrease in RR (Hamel, 2001) and an increased RR in response to music being reported (Urakawa & Yokoyama, 2005).

A link between respiratory rhythm and music was reported as far back as 1920 (Larsen & Galletly, 2005), a phenomena known as entrainment (Clifford, 2002; Scott, 2000). Entrainment was first described in 1665 by Christiaan Huygens, as being the process whereby the pendulums in clocks in the same room gradually swing in phase (synchronise) together (Spoor & Swift, 2000). It is claimed that music affects health through this process, that is, the synchronization of the body’s natural rhythms (for example, HR or RR) with the rhythm of the music, (Chlan, 1998; Scott, 2000). The link between RR and music is also understood to be a pattern of behavioural modification – that is a change in behaviour due to an external stimulus (Eidlitz, 2006).

It is believed that the speed rather than the style of music may have a greater affect on cardiac and respiratory responses due to a finding of increased RR with increased tempo (Bernardi et al., 2006). It is for this reason that the beat per minute rate (tempo) of music chosen to induce relaxation must match the resting rate of the heart. Faster pieces of music may cause an increase in physiological measures through entrainment of the body’s rhythms to the music, promoting cardiac and respiratory stimulation, while slower pieces of music may have the opposite effect. Subjects who listened to a piece of music with 70-80bpm for 20 minutes before catheterization displayed a decreased HR and RR compared to control (Hamel, 2001). However in another study it was reported that listening to music resulted in an increase in RR and HR regardless of the style of music (stimulating or calming) (Urakawa & Yokoyama, 2005). The increase in RR corresponds with an increase in RSA which was found to be greater when subjects were concentrating on music than when they were resting (Urakawa & Yokoyama, 2005). It appears that there is still much research to be done in this area to come to a consensus as to the effect of music on HR and RR.
HRV has also been extensively studied in relation to music in order to infer an affect of the ANS with findings of decreased (Okada et al., 2009) and increased LF/HF (Bernardi et al., 2006; Urakawa & Yokoyama, 2005).

Okada et al. (2009) reported an increase in parasympathetic activity (relaxation) as measured by HRV. The authors conducted MT once per week ten times consecutively for 45 minutes to 87 elderly patients (aged 82.3 ± 9.6 years) with dementia and cardiovascular disease. The music consisted of well known Japanese nursery rhymes, folk songs, hymns and recent Japanese popular music. MT took place from 11:00 – 11:45am and was performed by licensed and experienced music therapists. Subjects were randomly assigned to MT or non-MT groups. HRV was measured during the 45 minute MT intervention, and for an hour before and after the intervention. An increase in HF by 40-190% and a decrease in LF/HF by 20% were reported. Due to their reported findings the authors concluded that MT significantly increased parasympathetic activity and decreased sympathetic activity as measured by HRV (Okada et al., 2009). It was hypothesised that this was probably due to psycho-physiologic effects in which the rhythm and tempo of the music induced relaxation and distraction responses in the limbic and hypothalamic systems of the brain, and that MT reduces both adrenalin and noradrenalin levels. In addition to their physiological findings, some longitudinal findings were also reported in relation to cardiovascular disease. The incidences and acute exacerbations of congestive heart failure (CHF) events were reported to be significantly lower in the MT group than the non-MT group (10.9% versus 34.4%, p<0.05) showing that MT is useful in helping prevent CHF events in elderly cardiovascular disease patients. The authors concluded that soothing music can decrease the severity of sympathetic activation states such as anxiety, tachycardia (increased HR) and tachypnea (increased RR) via vagal innervations (Okada et al., 2009).

Equally the opposite effect has been reported, with an increase in SNS activity as measured by HRV. Urakawa & Yokoyama (2005) looked at the effect of music on HRV compared to no music in an exercise condition. Subjects in the study were healthy female college students (20-37 years old). Subjects were asked to bring music CDs based on their preference (which included pop songs with lyrics, jazz, nature sounds, “healing music”, and classical tracks). Subjects were required to rest without music, exercise then rest without music for one day, and at the same time on the following day (to avoid diurnal variation) rest with music, exercise
and rest with music. Music was delivered via a vibroacoustic apparatus (body sonic system) – a chair which allowed the subject to feel low pitch sounds through their body. LF/HF ratios were significantly increased (p<0.05) by exercise when subjects listened to music, while LF/HF changes were not significant (p>0.05) without music. HR was reported to be significantly increased by exercise both with and without music. These findings supported the authors’ hypothesis that SNS activity is increased after exercise in subjects who listen to music, with the music being responsible for promoting physiological excitation in an exercise induced sympathetic setting. The authors theorised that after exercise in which SNS activity is dominant, music synchronised with the stimulated SNS, resulting in further increased SNS activity. (Urakawa & Yokoyama, 2005).

Another indication of the ANS involves the measure of levels of sweat, as the ANS (specifically the SNS) controls sweating (Moore & Dalley, 1999). Galvanic skin response, or resistance (GSR) relates to a decrease of skin resistance in skin containing sweat glands as a result of increased sweating, and by implication increased SNS activity. Greene and Sutor (1971) played interrupted music, and used skin resistance to put off an interruption in the music by 30 seconds to measure ANS responses. A subject who responded to music continued to listen to music, while if there was no response the music was paused until a response occurred. Their findings suggested that ANS responses may relate to a conditioning response and raise the hypothesis that the more involved a subject is in the music the greater the effect on the ANS. A study by Sudheesh and Joseph (2000) of approximately 100 male and female subjects aged 20-25 years used GSR as an indicator of SNS activity to objectively measure the response of their subjects to music. The three classes of music used in the study were; karnatic classical songs (a type of Indian music), popular film songs, and western heavy metal songs. The findings of the study showed that people with different tastes in music responded differently, while those who were not involved in the music had no response. A subject was understood to be ‘involved’ in the music if they enjoyed or were interested in the music. Thus the physiological response of the ANS to music may be affected by how much the person likes or dislikes the piece of music being listened to.

The psychological effects, and use of music as therapy, are also explored in the literature. Many papers showed that music can be used to induce moods in experimental settings, and by extension be used for therapeutic purposes (Etzel, Johnsen, Dickerson, Tranel & Adolphs,
2005; Kreutz, Bongard, Rohrmann, Hodapp & Grebe, 2004; Lai & Good, 2005; Malinova, Malinova & Krusteva, 2004; Scott, 2000). It is claimed that music can be used to improve concentration, reduce anxiety and promote relaxation (Chlan, 1998), as well as enhance well-being, reduce stress and distract patients from unpleasant symptoms (Kemper & Danhauer, 2005). In their 2005 study, Li and Good (2005) reported that music was found to result in significantly better sleep quality as well as significantly better components of sleep quality. The study found better perceived sleep quality, longer sleep duration, greater sleep efficiency, shorter sleep latency, less sleep disturbance and less daytime dysfunction ($p<0.05$). Sleep improved weekly, indicating a cumulative dose effect (Lai & Good, 2005). It appears that music may also reduce psychological stress (Okada et al., 2009) and improve athletic performance – possibly by lowering the level of perceived exertion during physical exercise by a distraction effect (Bernardi et al., 2006), and improve relaxation compared to rest (Kemper, Hamilton, McLean & Lovato, 2008).

Panksepp and Bernatzky (2002) looked at the role of the brain in interpreting music on an emotional level, and autonomic responses (such as chills) to music. The authors emphasised the importance of remembering that individuals have distinct physiological responses to music, and that music is effective in inducing certain moods. It was also noted that females use music for mood regulation more than males and that if the subject chooses their own music there is a stronger effect from the music than if it is chosen by the experimenter. In the study 19-23 year old college students were measured for two music sessions, where they listened to 40min of “happy music” in one session and 40min of “sad music” in the other. Each participant brought a selection for each session (mostly common popular 1990s tunes). The “happy music” elevated feelings of happiness, and “sad music” elevated feelings of sadness, as measured by a self-rated seven point mood scale. Chill responses to music were shown to be triggered by a high-pitched sustained crescendo, especially relating to a solo instrument suddenly emerging from a softer orchestral background. Overall it was reported the effects lasted approximately 10min, and by 20min the changes were no longer significant (Panksepp & Bernatzky, 2002). These findings are corroborated by Urakawa and Yokoyama’s study which stated that it appeared that music that was in synchrony with a person’s mood at the time promoted a psychological response that led to physiological changes regardless of whether the person was in an activated or calm state (Urakawa & Yokoyama, 2005).
Factors that influence the effect of music include gender and personal preference of music styles. Gender has an influence on physiological measures, as men and women differ in their music preferences and emotional reactions to music (Nater et al., 2006). Personal preference may have an impact on a person’s response to the music. When given the option to choose music for relaxation, many (56%) chose classical music for promoting relaxation (Chlan, 1998). Further, significant reductions in agitation during and following an individualised compared to classical music intervention were reported (Gerdner, 2000) indicating that it was important for the subject to be free to choose the music which they listen to. This freedom in choice may avoid agitation or mental stress, which as indicated by Marek (1996) will affect HRV. Urakawa and Yokoyama (2005) also reported that preferred music decreased anxiety and increased relaxation, but also indicated that the music increased HR, vascular constriction, peripheral skin temperature and stimulated muscle activity. In contrast to Gerdner’s study (2000), Scott (2000) reported that there were significantly more positive changes to mood using researcher-selected music, instead of self-selected (or individualised) music, demonstrating that it is the components of the music, and not who chooses the music, which are important.

Finally, although Bernardi et al. (2006) reported that results were not influenced by the subject’s music preference, the type of music does seem to be significant. Classical music caused comfort compared to rock music and noise which caused discomfort (Umerura & Honda, 1998). Grunge rock (a style of music characterised by harsh guitar chords) increased negative emotions such as hostility, compared to designer music (music designed to have specific effects on the listener) which improved caring and relaxation (McCraty, Barrios-Choplin, Atkinson, & Tomasino, 1998), and heavy metal music is related to unpleasantness (Nater et al., 2006).

Overall the two major considerations for music are pieces that will not cause the subject agitation or discomfort, and which have a lower beat rhythm that entrains to the desired HR to promote relaxation.

In comparing subjects’ music sessions it was found that there was no evidence of autonomic habituation to any given music from session to session (Bernardi et al., 2006). This means
that any affect of music will occur even if the subject has listened to the music previously, so using the same music a second time shouldn’t result in a lesser effect the second time.

Despite the lack of concurrence between studies, that music exerts an effect on the ANS is not in dispute. The effect seems to be dependent on the type, tempo and volume of the music, as well as how much involvement or emotional response the subject has to the music. Further research into the effects of music on HRV could aid our understanding of the use of music as a therapeutic tool.

**2.5 Soft Tissue Massage**

**2.5.1 Physiological Effects of Massage**

Soft tissue massage is a well known technique used for reducing tonicity of hypertonic muscles and relief of tension and pain. It is a much used therapeutic technique in Osteopathy for improving fluid drainage, reducing tonicity of muscles, and promoting relaxation. It is also claimed that through touch the Osteopath can influence the neurological organisation of the individual and affect other psychological processes (such as mood and pain perception) (Lederman, 2003). Thus soft tissue massage can not only promote relaxation physically through a muscle, but also neurologically through the psychophysiological response of the patient to touch.

It seems to be common that subjects who are asked how they felt report a relaxation response to massage, however there is less research into the actual physiological response of the human body in purely objective physiological findings. For example an examination of the longitudinal effects of full body massage therapy used the subjective measures of present pain intensity VAS, anxiety-VAS and the modified Short-Form McGill Pain Questionnaire, and only mean arterial pressure as a physiological measure of the effect of the intervention (Jane et al., 2009). Although the results of the study were reported to be statistically significant for short, intermediate and long term effects, the emphasis of the study was on pain management and not the physiological response to the intervention. As this present study focuses on the objective physiological responses of the human body to music and soft tissue massage, a greater emphasis has been placed in the literature review on searching for papers specifically focused in this area rather than the papers which report subjective relaxation responses.
Diego et al. (2004) in addition to anxiety and stress scores also researched the physiological effects of massage therapy by recording EEG activity for alpha, beta and delta wave changes, and ECG for HR. Thirty-six healthy adults were randomly assigned to a moderate massage, light massage or vibratory stimulation group. The moderate massage group showed a decrease in HR as well as changes in EEG activity indicating relaxation (increased delta and decreased alpha and beta activity). The light massage group on the other hand showed the opposite indicating arousal, with increased HR and beta activity and decreased delta activity. The vibratory stimulation group also showed indications of increased arousal (Diego et al., 2004).

Other studies confirm the relaxation effect of massage. A single application of massage therapy was shown to reduce anxiety, BP and HR, while multiple applications reduced delayed assessment of pain. (Moyer et al., 2004). Massage was also shown to reduce levels of pain perception and anxiety, as well as decreasing SBP, DBP and HR indicating relaxation (Mok & Woo, 2004).

Regarding the effects of soft tissue massage (effleurage) on the physiological components of relaxation, a meta-analysis of nine studies done between 1964 and 1993 on both healthy and immobile subjects reported that biological relaxation is associated with significant reductions in HR and RR for both genders, and that effleurage massage of at least three minutes promotes biological and subjective relaxation (Labyak & Metzger, 1997). Effleurage massage involves smooth gliding strokes (Kutner, Smith, Corbin, Hemphill, Benton, Mellis, Beaty, Felton, Yamashita, Bryant & Fairclough, 2008). The measures compared in the meta-analysis were SBP, DBP, HR and RR. Three minute massages were associated with a decline in SBP, DBP, HR and RR and had a continued reduction during the 5 minute post massage period. Gender related differences were noticed regardless of health status. Female subjects showed a rise in BP during the first 3-5 minutes of massage which declined but did not drop below baseline during the 10 minute post massage rest period. Male subjects however all showed a decline in BP and continued reductions during the 10 minute rest period. In both genders HR and RR declined immediately in the first 3 minutes of massage and remained below baseline throughout the massage. These changes are also consistent with subjects’ self-reports of the
relaxing effects of back rubs. Subjects reported the experience pleasant and/or relaxing regardless of the length of the massage (Labyak & Metzger, 1997).

In addition to its physiological (and subjective) relaxation effect, massage has also been shown to be an effective treatment for both short and long-term persistent low back pain sufferers (Melchart & Eisenberg, 2000). Massage decreases anxiety and depression, increases attentiveness, relieves stress and pain and reduces BP (Hatayama et al., 2008). Massage has also been shown to be useful to enhance sleep. For example, back massage is useful for promoting sleep in critically ill older men (Richards, 1998). Overall, some of massage therapy's largest effects are reductions in anxiety and depression of a similar magnitude to psychotherapy (Moyer et al., 2004).

In general, although there is some indication in the literature of an arousal response to massage (Diego et al., 2004), it is generally accepted that massage has a relaxing effect (Labyak & Metzger, 1997; Mok & Woo, 2004; Moyer et al., 2004).

2.5.2 Massage and the ANS

Although the quantity of research on the effects of massage on the ANS is small, there are a few papers with differing reports as to the effect of massage on the ANS. Both an increase in parasympathetic (Tochikubo et al., 2006) and sympathetic (Hatayama et al., 2008) activity have been reported. These differences may relate to the type, duration or location of massage, and further research is needed in this area before any solid conclusions can be made.

Tochikubo et al. (2006) reported a parasympathetic response to pulse-synchronized air-massage to the lower extremities. The major findings reported were significantly increased peripheral vascular blood flow in the lower extremities, and an increase the HF component of HRV. It was suggested that the intervention may increase venous return and affect cardiac PNS activity, possibly as a consequence of baroreflex control (i.e. as a response to the change in pressure caused by the pressure of the air-massage cuffs). It was also reported that LF/HF decreased significantly. Weerapong (2005) also found changes in PNS activity indicated by a cardiac relaxation response (decreased HR) following a massage intervention.
A recent study into the effects of cosmetic facial massage on the ANS using HRV found that subjects were activated physically as shown by an increase in sympathetic nervous activity following massage. The increase in sympathetic activity was indicated by a significantly enhanced LF/HF ratio after the massage. The LF/HF ratio increased from 0.81 to 1.22 ($p = 0.035$) indicating a decrease of parasympathetic proportional to an increase in sympathetic activity. The facial massage involved cleansing with cream by hand, removal of keratin by steaming, ultrasound facial treatment with gel, and application of lotion for moisture retention. The total time of facial massage was 45 minutes (Hatayama et al., 2008). Additionally the authors expressed surprise at the results, which were contrary to their prediction, due to the amount of anecdotal evidence that massage enhances relaxation. It appears that though subjects feel relaxed, their physiological markers indicate stimulation.

Regarding the effects of OMT on muscle contraction headache, 22 subjects were researched with Osteopathic soft tissue procedures (such as kneading, deep pressure and stretching) as well as high velocity low amplitude procedures to release restriction (Hoyt, Shaffer, Bard, Benesler, Blankenhorn, Gray, Hartman & Hughes, 1979). A one way ANOVA of changes in EMG levels of the frontalis muscle and dominant hand temperature showed no systematic changes among the three treatment groups. A repetition of the study on non-symptomatic subjects also showed no systematic changes in EMG levels. There was however a significant reduction in rated levels of pain ($p<0.0003$). These results raised questions as to the appropriateness of EMG levels of the frontalis muscle, and dominant hand temperature as measures of reduction of severity of headaches. The article commented on the apparent independence of ratings of headache severity from EMG levels in the frontalis muscle, citing other studies with similar results. A hypothesis was raised that there is some intervening process between EMG levels of frontalis contraction and presence of headache. Hypothesised was that pain in muscle contraction headache is mediated by cortical interpretation of sensory data and resultant changes in autonomic reactivity and thus pain may be independent of EMG levels. In other words cortical activation influences the degree of ANS activation, so actions which influence the cortex such as instructing a subject to relax may reduce the subjective level of pain without affecting EMG levels. It was speculated that the effect of Osteopathic manipulation (soft tissue techniques) was through the level of cortical response, reducing autonomic reactivity and thus subjective pain. The suggestion for further study was the
measurement of skin conductance as an indication of autonomic reactivity to test this hypothesis (Hoyt et al., 1979).

Massage appears to have a different effect on the sympathetic and/or parasympathetic response, depending on factors such as the therapist’s intent, the scope of other treatments involved, and the way in which the person being massaged interprets the intervention (Lederman, 2003). These would appear to be the pathways which are hinted at by Hoyt et al. (1979) in which a cortical response, such as the subject’s interpretation of the massage, causes an ANS change. So depending on the cortical response massage may reduce arousal, implying reduced SNS activity, or may cause an increase in SNS activity as manifested by a slight increase in HR and RR (Lederman, 2003). This response may also depend on the circumstances involved, such as the rate or rhythm of the massage being applied, the force, and the subject’s interpretation of the massage they are receiving.

2.6 Combined Effects of Music and Massage on HRV

There is sparse literature available relating to music, massage and HRV. From what is available it would appear that music and massage used together do not produce a greater effect than the two individually. Calming music and hand massage were used in a nursing home to reduce agitation and create a calm structured surrounding, but no additive benefit was found by combining the two together (Remington, 2002). However there is insufficient literature on any additive effects of music and massage to make statements as to whether any cumulative effect may or may not present.

In regards to measuring HRV in participants under different experimental conditions, further research is needed. A better understanding of the relationship between HRV and ANS effect is needed to better understand the effects of interventions that are designed to alter or affect autonomic tone and HRV (Goldberger et al., 2001).
2.7 Ectopic Beats

As ectopic beats are an important consideration in any research relating to HRV, it was considered vital to include a description of ectopic beats, what they are and how they affect the data, and the processes required to remove them.

2.7.1 Definition of Ectopic Beats

Raw data frequently contained ectopic (or outlying) datum points. The term ectopic, from the Greek meaning ‘out of place’ (Clifford, 2006) in HRV analysis refers to datum points which are believed for one reason or another, to be incorrect or out of place in the data set.

Ectopic beats can be identified by abnormal morphology (such as a very large or small beat), but also by abnormal timing if the beat arrives significantly earlier or later than the beat after or before (Clifford, 2002). Ectopic beats with abnormal morphology can be caused by various artefacts of instrumentation as well as by a ventricular arrhythmia (Clifford, 2006), while those with abnormal timing tend to be caused by atrial arrhythmias (Clifford, 2006). About one third of healthy men exhibit one or more ventricular ectopic beat per hour, and up to six per hour associated with ventricular arrhythmias (Clifford, 2002).

2.7.2 How Ectopic Beats Occur

Outlying datum points result due to a combination of factors, including as artefacts of the instrumentation and due to certain extreme factors such as a sudden loud noise. Ectopic beats can also be caused by premature ventricular contractions (Keenan, 2005).

Artefacts of instrumentation relate to such factors as the accidental contamination of the Polar recording device by another nearby electronic device known as electrosurgical noise (especially devices operating at frequencies between 100kHz and 1MHz (Clifford, 2002)), poor adjustment of the chest strap resulting in errors in data collection (known as contact noise or motion artefacts when the electrode is loose and begins to lose contact with the subject (Clifford, 2002) usually causing sharp changes in the ECG because of the variations in impedance between the electrode and subject’s skin), and poor sensitivity of the instrument to
the participant’s heart beat (usually caused by insufficient dampening of the chest strap electrode points resulting in insufficient conductivity for a clear reading), resulting either a merging of two beats together, a skipping of one beat entirely, or repeating a measurement a second time unnecessarily. Further sources of ectopic beats are muscle contractions which can produce electromyographic noise and relating to respiration, a phenomena known as baseline drift (Clifford, 2002).

The data collecting device can also produce noise and create artefacts (Clifford, 2002). Generally artefacts, such as those caused by movement of the electrode on the subject’s skin, and abnormal beats, tend to have a much larger amplitude than a normal beat (Clifford, 2006). It was found that in instances where the Polar monitor is completely unable to detect the subject’s heart beat, the instrument will record a repeat of the last measure, resulting in a ‘flat-line’ on the HR chart.

2.7.3 How Ectopic Beats Affect the Data

Normal heart beats (sinus rhythm) are generated in the sinoatrial node of the heart, which is richly innervated by both the SNS and PNS branches of the ANS. Thus beats which are generated from outside of these normal conduction mechanisms are not considered to represent the ANS (Clifford, 2002). Thus beats which fall outside of the timing of a normal sinus rhythm are considered ectopic and should be excluded as they do not represent autonomic activity and are not believed to contribute to HRV (Clifford, 2002). Additionally the prolonged pause created by the unusual timing of an ectopic beat alter the proportion of frequency components (usually causing a falsely higher representation of HF component to HRV) (Clifford, 2002).

Due to the abnormal timing of an ectopic beat, it appears in the data as a beat much larger (or smaller) than it should be in reality, forming a spike in the R-R tachogram (Clifford, 2002), which results in an increase in the randomness, or variability of the data (Peltola, Seppänen, Mäkikallio & Huikuri, 2004; Storck, Ericson, Lindblad & Jensen-Urstad, 2001), and introduces mathematical artefact to the data resulting in an incorrect HRV extrapolation (Lippman et al., 1993). Essentially ectopic beats corrupt the recorded signal, thus making it necessary to correct ectopic beats before analysis (Mateo & Laguna, 2003). Not removing ectopic beats
results in alterations in HRV (Lippman, Stein & Lerman, 1994) as the increased randomness created by ectopic beats creates a greater indication of variability than is actually present in the data. Typically this will show as an increase in HF power and a decrease in LF power, resulting in a distortion of the LF/HF ratio (Clifford, 2002). This means that ectopic beats can interfere with analysis of HRV and give wrong results (Storck et al., 2001).

One study found that less than 1% of ectopic beats was enough to seriously affect HRV analysis, and so even a small number of ectopic beats should be filtered before analysis (Storck et al., 2001). It is believed that HRV is unaffected by isolated ectopic beats once they have been removed (Clifford, 2002). Various methods, such as a filter algorithm to detect and replace ectopic beats, is an effective correction method to reduce the heightened variability caused by ectopic beats (Storck et al., 2001).

### 2.7.4 How to Avoid Ectopic Contamination

One study went to particular lengths to avoid potential artefact contamination of their measurements, such as operating sensor testing of their instrument battery level under active load, testing of impedance levels of their EMG sensor-skin interface, as well as testing of instrument performance to assess accuracy of physiological measurement (Hoyt et al., 1979). They also monitored room temperature and tried a single blind procedure (where the measurement team were unaware of the subject’s treatment condition, and the investigator who was administering treatment was unaware of what the measurements being obtained were) (Hoyt et al., 1979). In retrospect of the current study it was considered possible that the battery in the Polar device used may have been gradually depleted over the course of the study enough to begin to cause more outliers to be produced, although no actual evidence of this was seen. However for future studies the regular changing of batteries in the Polar device should be considered in order to reduce any lowered battery level artefact contamination of data.

### 2.7.5 Setting Limits / Defining Unusable Data

Only data which is free of ectopic beats and other anomalies (i.e. arrhythmic events, missing data and noise effects) should be used for analysis of HRV, though preferential selection of
such data can introduce selection bias (Malik, 1996). Interpolation (replacement of ectopic beats) can reduce the error caused by these abnormal beats to allow data containing ectopic beats to still be usable for analysis.

There is some discrepancy as to the exact number of ectopic beats which make a data set unusable. Data containing a high incidence of ectopic beats should be excluded (Clifford, 2002), however the definition of ‘high’ varies from author to author. One very conservative approach involves excluding data that has more than 20 ectopic beats per hour of data recorded (approximately 0.5% of the total) (Malik & Camm, 1995), while a more complicated method allows for the removal of as much as 20% of the data points without introducing error into the HRV analysis (Clifford, 2006). These limits are however largely arbitrary and have not necessarily been tested beyond the disease specific criteria of the studies in which they were employed.

For the purposes of this current study it was considered that a data set containing greater than 5% ectopic beats was unusable. Additionally any sections of data showing indications of arrhythmic events (a series of abnormal beats together is classified as an arrhythmia (Clifford, 2006)) or noise effects (such as a flat-line of unchanging HR, or fluctuating from a high to low HR) were also considered unusable, even if the individual data points were not considered outliers. One data set contained such abnormalities, believed to be caused by an unknown cardiac condition, undiagnosed and not reported before the participant began the study, and was excluded from data analysis.

As ectopic beats often appear either early or late with respect to the timing of a sinus beat (Clifford, 2002) they can be defined according to the timing of cardiac depolarisation by identifying points with an R-R interval less than or equal to 80% of the previous interval (Clifford & Tarassenko, 2005), or less than 80% of the average R-R interval (Malik & Camm, 1995). Both of these definitions are not free of problems however. The Clifford and Tarassenko definition is prone to failure if the previous interval is also considered an ectopic, as the definition may lead to a failure to identify the second ectopic beat, while the Malik and Camm definition requires the removal of ectopic R-R intervals to ascertain the average R-R interval which was needed to identify the ectopic R-R intervals in the first place. An alternative definition sets the limits for timing at 1.325 times greater or 0.755 times smaller than the
previous R-R interval (Clifford, 2002), though this definition is also as susceptible to the same flaws as described above. Additionally these methods exclude the possibility of a normal individual being able to adapt their HR more or less than the definition allows for (and the reverse, meaning that legitimate ectopic beats may not be detected). Normal variability may be anything between 10-30% (Kobayashi et al., 1999). A further more aggressive model identifies ectopic beats if the beat interval changes by more than 12.5% compared to the previous interval (Clifford, 2002). Using these methods runs the risk of both removing legitimate sinus beats as well as failing to remove ectopic beats in proportion to the percentage limits which are set (Clifford, 2002). Additionally it has to be remembered when it comes to editing ectopic beats that the change in timing of an ectopic beat will affect the interval before and after the actual beat and so both may need to be edited (Clifford, 2002).

2.7.6 How Ectopic Beats are Removed / Ectopy Correction

In general sections of data containing ectopic beats should be excluded from the study (Malik, Bigger, Camm, Kleiger, Malliani, Moss & al., 1996), however this is not always possible, for example in the current study a single ectopic beat in the middle of one ten minute section of data would render all the subject’s data unusable. Further it has been shown that this kind of selective use of data results in loss of data and can bias results (Lippman et al., 1994), because if ectopic beats relate to changes in autonomic tone, for example a set of data points showing a gradual increase which peaks with one or more ectopic beats, and this data is excluded, it will bias the HRV results by removing this true example of variability (Lippman et al., 1993).

As entire sections with ectopic beats cannot be excluded without potentially excluding the subject’s entire data, it is necessary to exclude only the individual ectopic beats. This can be done in many various ways, which fall into two general categories; deletion of the ectopic beat, or replacement (interpolation). Interpolation of ectopic beats was compared with deletion without replacement, and it was reported that interpolation worked well while deletion resulted in significant error (Salo, Huikuri & Seppänen, 2001). However Lippman et al. (1994) showed that deletion of ectopic beats was acceptable for short samples of data, being just as good as or better than more complicated methods of ectopic removal.
In order to decide which method – deletion or interpolation – is most appropriate, it is helpful to understand that ectopic beats may occur either instead of or in addition to a normal sinus rhythm. If the ectopic beat occurs as well as a normal sinus rhythm, it is appropriate to remove the beat entirely from between the two normal beats (Clifford, 2002), thus restoring the R-R interval between the two normal beats back to normal. If however the ectopic beats occurs instead of a normal beat, the ectopic beat cannot be removed (without leaving a very large interval) and so it needs to be replaced with a ‘phantom’ beat, a beat which is normally taken as being an average of the interval before and after the ectopic beat, where a sinus beat would have been expected to occur (Clifford, 2002).

The process of removal of outlying datum points is a potentially involved and complicated one, due to the vast numbers of datum points under consideration, making automatic removal methods desirable (Acar, Savelieva, Hemingway & Malik, 2000). Due to this, an Excel spreadsheet was created to partially automate the process of manual removal of outlying datum points. Several formulas were created to identify outlying points, whether three standard deviations greater or lesser than the mean of the data set. These outliers were then replaced by a formula with an average of the two proceeding points (preceding rather than following, as the following points being as yet unchanged by the formula may be outliers themselves – thus using preceding points ensured that replaced data was not still an outlying point).

The correction technique of using a certain number of beats preceding the ectopic beat was found in one study to be similar to other methods of ectopic correction (such as taking an average of the intervals before and after the ectopic beat), and may even be more accurate (Solem, Laguna & Sörnmo, 2006). An alternative replacement method involves replacing ectopic data with non-ectopic data from the same data set which most closely approximates the ectopic data (Lippman et al., 1993). This method is complicated, and no real evidence is given to indicate its superiority over other simpler methods of ectopic removal.

Although the method employed to detect ectopic beats was successful in identifying (and replacing) largely outlying datum points, it was considered possible that setting the limits of identifying ectopic beats at purely three standard deviations of the data may not have sufficiently identified ectopic beats according to the timing definitions described above.
Consequently some ectopic beats may have been missed in the data, potentially affecting the results.
3.0 Method

3.1 Overview of Experiment

Twenty (N=20) male participants aged 18-35 were recruited for the study to attend four 30 minute sessions of data collection. After data from drop-outs were removed, 16 participants of mean (standard deviation) age 25.9 (5.5) years were analysed. A heart rate (HR) monitor was worn for all 30 minute sessions to collect HR data for the entire session length. The four interventions involved music, massage, music and massage combined and a control with no music or massage.

The experiment took the form of a randomised, controlled cross-over trial. The reason for a cross over trial was to eliminate any cumulative effect from interventions. Interventions were randomised to eliminate any potential bias relating to the researcher, participant or selection process (Lachin, 1988).

The study was not blinded as it was not possible to blind the subject to the intervention they were receiving. However subjects were relatively blinded to the results/outcomes of the study as heart rate variability (HRV) data can only be obtained after analysing HR data.

3.2 Ethics

Before research could begin a proposal for ethical consideration was submitted to the Unitec Research Ethics Committee (UREC). Points of ethical consideration were age of participants, gender of participants, ethnicity and nationality of participants, safety of participants in the study including both physical safety and protection from coercion or emotional harm due to the study. The study was approved by the UREC from 30-4-2009 to 30-4-2010.

Participants in the study were strictly volunteers and were free to participate or decline participation at their own discretion, as well as withdraw their consent for participation at any time up until two weeks after their final data were collected. It was considered that as many potential subjects turned down the researcher’s approach to ask for interest in participation that there was no coercion into participating. An example of the participant consent form and
information sheet outlying the specific issues and frequently asked questions about this research project is attached in section 10.1 and 10.2.

### 3.3 Selection and Recruitment

In order to recruit participants into the study, posters/fliers were put up on notice boards around Unitec (Carrington Campus) in the Health and Sports departments, with details of the study and the researcher’s contact details, so that potential participants could get in contact with the researcher to register their interest in participating in the study. Fliers were also handed out by one of the researcher’s supervisors to students at Unitec.

Participant recruitment was also done by general and personal word of mouth. Subjects ranged from well known to completely unknown to the researcher.

Through these above methods, 20 male subjects were recruited by convenience sampling to participate in the study from within and outside of the Unitec population. Twenty participants were chosen as it was considered that this number would provide sufficient data to validate the study, based on a personal communication in November 2008 with Associate Professor Andy M Stewart PhD, member of the UREC, and based on the recommendations of the UREC. For these reasons a power calculation was not performed.

In order to remove any gender effect all participants in the study were male. The literature indicated differing HRV and responses to music and massage between males and females (Labyak & Metzger, 1997), and that genders should be researched and studied separately (Tochikubo et al., 2006). Males only were selected as it was considered the female menstrual cycle would affect HRV findings.

The age criteria for participants was 18-35 inclusive. As HRV is shown to change with age (Aubert & Beckers, 2003), this age group was chosen in order to make the subjects’ baseline HRV measurements as similar as possible. Children (under the age of 18) were not used in the study as their physiological response to either intervention may have been different to the adult population, and also to eliminate any consent and ethical issues of recruiting and using children for research.
3.4 Selection Criteria

The following selection criteria were employed in the recruitment of participants into this study. Participants were required to meet all the inclusion criteria, and to not meet any of the exclusion criteria to be considered valid candidates for this study.

The list of inclusion criteria was determined by the researcher based on information gathered in the literature review. The list of exclusion criteria was formed primarily from common sense concepts, as well as from previous research projects using similar methods. For example as participants were required to listen to music, any participant with hearing impairment may have difficulty or altered results compared to people with unimpaired hearing. This list of exclusion criteria thus included all the major factors which were considered at the time to be potential complicating factors to a participant’s results.

3.4.1 Inclusion Criteria:

- Male
- Aged between 18 – 35 (inclusive)
- Gave informed consent to participate

3.4.2 Exclusion Criteria:

- Hearing impaired
- Any person who found music caused agitation (or anyone who disliked all of the choices of music from the pre-selected selection)
- Using sedative medications at the time of the study
- Could not comfortably lie prone for the duration of the study
- Respiratory disease (e.g. asthma)
- Any kind of cardiac disease
- Any skin or sensation defect that may have changed the effects of massage (e.g. stroke)
- Any person who was taking mood enhancers or mood altering drugs (other than tea, coffee and alcohol) at the time of the study
3.5 Distribution of Participants into Random Groups

A Latin Square formula was used to distribute 20 participants into four groups, to determine the order in which the participant would receive each of the four interventions. The Latin Square formula as follows was used:

ABCD
BDAC
CADB
DCBA

In this way each intervention preceded and followed all others equally, allowing for any cumulative effect to be accounted for. There were five participants in each group.

The values A to D represented the four interventions in this way:
A: music intervention.
B: massage intervention
C: music and massage combined.
D: control intervention.

The four groups determined by the Latin Square were written five times each on 20 slips of paper. Subjects chose a slip of paper randomly from this selection to determine the order in which they would receive interventions. Participants could not see the order of intervention they would receive until after they had randomly chosen a slip of paper. This is considered a valid technique for randomisation, though randomisation by computer would be more scientifically justified (Kerr, Robinson, Stevens, Braunholtz, Edwards & Lilford, 2004).

3.6 Method of Data Collection

3.6.1 Interaction with Participants

Participants had the study explained to them and were given an information sheet (section 10.2) to read explaining the research and reason for the study, the participant’s obligations, the researcher’s obligations, ethical outlines and so forth. Participants were then required to sign a consent form before beginning the study (section 10.1).
Participants in the study were required to meet the pre-determined selection criteria (and to not meet any of the exclusion criteria). These selection criteria are outlined in section 3.4. One potential participant was excluded from the study before beginning due to a known heart condition.

In order to minimise the effect of other stimulating factors on the results, participants were asked to abstain from tea, coffee, and alcohol (Gendolla & Krusken, 2001), for 2 hours prior to participation. Participants were also requested to refrain from eating and exercising for 2 hours before participating in the study so that Autonomic influences on digestion would not affect the study (Berntson et al., 1997).

At the beginning of all sessions participants were greeted in a friendly way, and endeavoured to be made to feel comfortable. This included ensuring the room was heated to a comfortable temperature, and that the participant felt safe and informed as to what was happening with regards to the research they were participating in. Additional explanation was provided in the first session which the participant attended, with the provision of an information sheet, and the signing of a consent form.

All participants were required to undress for the HR monitor chest strap to be attached, and for the massage intervention. It was explained to all participants the need for a HR monitor chest strap to be attached to bare skin to measure their HR data during the experiment, and for shirts to be off for the massage sessions. This information was verbally explained to the participant at the time of research, and was also written in the participant information sheet which was given to be read by those wishing to be involved in the study. Undressing involved removing all clothing worn on the upper body, for example jumpers, jerseys, t-shirts etc., but lower body clothing such as trousers and shorts was not required to be removed. To maintain good control protocols all subjects were asked to undress for all sessions, which also facilitated putting on the HR chest strap. However a few subjects remained clothed (i.e. re-clothed after the chest strap was put on) for some sessions due to cold temperatures / sensitivity to cold.

All subjects were required to wear the Polar S810 chest strap to monitor their HR. The Polar chest strap was required to be moistened on both electrode points to facilitate conductivity in
accordance with the operating instructions for the device. The chest strap was moistened with clean fresh water (either under a running tap or immersed in a cup of fresh water) immediately before every session. The temperature of the water was generally cold, however warm water was used for some sessions for participant comfort (some participants reported mild discomfort to the cold temperature of the chest strap after it was dampened with cold water). The chest strap was attached across the middle of the chest at the bottom of the sternum (approximately an inch below the nipple line) and was adjusted for size between subjects to fit comfortably around the chest. The Polar chest strap is elastic allowing it to stretch for breathing, and was large enough to be able to be adjusted to fit all subjects.

All participants lay face down for all sessions. It was necessary for subjects to lie in this position to allow access their back and shoulders for the massage interventions. As there is a connection between HRV and posture (see effect of posture on HRV section 6.4.2) it was considered important for all subjects to lie face down for all sessions to control posture as much as possible.

After lying face down participants were asked if comfortable. Pillows and/or cushions were available for comfort if required (for under the participant’s head, stomach and/or feet) as was a rolled towel for around the face. Where sessions were done on an Osteopathic or massage table with a face-hole, a rolled towel for around the face was important to make the face hole comfortable for the subject. Subjects were aware of the importance of relaxation in the study and so were conscious of the importance of being comfortable for the study and reporting any discomfort.

It was endeavoured to control room temperature where possible to expedite participant comfort. As data collection was primarily done over the winter months, the ambient temperature was considered to be lower than normal comfortable levels at most times of the day. To raise the temperature to comfortable levels a heater was on in the room where available. It is to be noted that some of the locations of data collection did not have access to a heater. An electric fan heater was the most commonly used form of heating. In some sessions a bar heater, gas heater or wood fireplace were used for heating. Heating was adjusted to a comfortable temperature for participant and researcher. This was done by asking
subjects if they were warm enough and temperature was adjusted according to the response if appropriate.

At all times participants were treated fairly and respectfully, and all participants were gratefully thanked for their participation in the researcher’s project.

### 3.6.2 Data Collection Process

Data were collected by the Polar S810 Heart Rate Monitor (Polar Electro, 2010), a device the shape and size of a wrist watch. A stopwatch on the Polar monitor indicated the time since recording began in minutes and seconds. The data were later downloaded and interpreted by computer software designed for HRV analysis (Niskanen et al., 2002).

The initial 10 minute washout period allowed the collection of a baseline reading, as well as allowed sufficient adjustment time for any variables which may potentially have been affecting the participant’s baseline HR, such as the act of lying down for the study, walking to the room where the study was conducted, and other previous activities just prior to the study. The final 10 minute post-intervention reading was gathered to determine if there was a lasting or continuing effect of the intervention.

A 10 minute intervention time period was used by several similar studies (Hatayama et al., 2008; Hoyt et al., 1979; Labyak & Metzger, 1997) and so was chosen to allow sufficient data to be collected for analysis, being twice the minimum window required for HRV analysis (MacArthur & MacArthur, 2000). Some studies collected intervention data for less than ten minutes (three minutes, five minutes and six minutes of massage in various studies) (Labyak & Metzger, 1997) or for 15 minutes (Tochikubo et al., 2006), however overall it was considered 10 minutes was a suitable duration.

Of each 10 minute segment it was considered that some data would be unusable, namely the first and last minutes of each segment collected. The first two minutes of each segment of data were excluded in case of any transition or adjustment effect. For example most subjects showed a slightly higher HR for the first part of each session, probably due to the initial action of lying down from a standing position. Additionally when the intervention began, subjects may have been focusing or concentrating on something else and thus may have required an initial
adjustment to the new stimulus to relax. Further, some selections of music overran the 10 minute period by a few seconds to half a minute, meaning that the first few seconds of post-intervention silence still had some music stimulus and thus was required to be excluded. The last minute of each segment of data was excluded from analysis in case of any anticipation response relating to the upcoming intervention, anticipating the end of the intervention, or anticipating the end of the session. In this way the data used for analysis represented as much as possible purely the condition of interest without contamination from other conditions or extraneous factors.

Thirty minutes of continuous HR data was recorded for each session with each participant. A note was made of the specific time as recorded by the Polar stopwatch when interventions began and finished, for reference in the data analysis stage.

In the interventions involving music, the first music track began after the 10 minute baseline reading was completed, with a brief check with the participant that the volume was satisfactory, and an adjustment of volume if necessary. In the case of the massage interventions, massage was preceded with a brief comment that massage was about to begin, to ensure the participant was fully informed throughout the study. In the control intervention, no comment was made to distinguish between the 10 minute intervals.

A control group was used in order to determine if any effects of the interventions was due to the intervention, or simply due to the time spent lying down (Hoyt et al., 1979). Although one study did not consider the use of a control group important at the time, it was noted in their discussion that their lack of a control group limited their study (Hatayama et al., 2008).

In order to control for any diurnal variation (changes in HR and HRV due to the time of day or night (Iñesta et al., 2008; Urakawa & Yokoyama, 2005)), it was intended that participants attend sessions at similar times of the day on successive days. However as collection of data was limited to participant availability this was not necessarily possible for all participants. Whenever possible participants were rebooked for their next session at a similar time of the day on the next mutually suitable available day, however few participants were able to attend all four of their sessions at similar times of the day, and globally different participants attended sessions at all different times of the day.
It was also desirable to have at least a 24 hour washout period between sessions in order to control for any potential cumulative effect of interventions. In most cases this washout period was at least a day, though for a few due to time restraints of collecting data this washout period was less.

The massage was applied by the researcher to the back and shoulders of the subject for the purpose of reducing sympathetic excitation. The massage was applied in the same manner with comparable force to each subject, and all massage interventions were administered only by the researcher in order to minimise any chance of experimenter effect (Hoyt et al., 1979). Each massage intervention followed a similar procedure, beginning with approximately three minutes to the right thoracic erector spinae (TES) muscles, three minutes to the left TES, one and a half minutes to the right upper trapezius muscle, one and a half minutes to the left upper trapezius muscle and a final minute of bilateral longitudinal soft tissue massage to the TES muscles. It was endeavoured to make the massage intervention the same for all participants, however for some sessions the duration of massage applied to individual muscle groups was not exactly the same, though the total massage time remained the same for all participants.

The types of music selected were pieces with a rhythm of 60-80bpm (with only one or two pieces in the available selection outside of this range) which is characteristic of the resting HR (Chlan, 1998) and results in a decreased HR (Hamel, 2001). A selection of music genres, including classical, easy listening, instrumental and vocal, and all within the same beat frequency range were available for the subjects to choose from. The music was all played from the researcher’s laptop for all participants and for all music sessions. The music volume was adjusted before the session begun to a comfortable level to the participant, and readjusted if necessary at the initialisation of the music intervention period. The same pieces of music were listened to by the participant twice, once during the music session, and once during the music and massage combined session. The same music was listened to in order to better control the music intervention for the individual participant. It is believed there is no habituation effect in listening to the same piece of music for a second time (Bernardi et al., 2006). Of the pre-screened selection of music pieces, only one was later removed due to a section in the music with an increased tempo which was considered to be greater than the
beat per minute selection criteria. The participant was allowed to finish the session; however the piece of music was replaced for the following session in which it was required.

Sessions were conducted either on an Osteopathic treatment table at the student clinic at Unitec, in the participant’s home (usually on the participant’s bed) or at the researcher’s home on a bed or couch. The respective heights of these instruments were largely comparable, and the difference between subjects lying on an Osteopathic treatment table compared to a bed was not considered significant.

3.7 Data Analysis

Data were analysed using XLSTAT for Microsoft Excel.

Population data were analysed using analysis of variance (ANOVA) tests to obtain p values (significance level 5%). Additionally it was endeavoured to group participants into responder and non-responder groups for ANOVA. How responders and non-responders were defined is discussed below.

Finally participants were grouped according to their average HR during sessions and analysed using a Chi squared correlation test to determine significance of grouping into responder and non-responder groups.

3.7.1 Explanation of Grouped HRV Analysis

In analysing the data it was found that due to the inherent individual differences between subjects, group comparisons of HRV data may not be useful. The components of HRV provide an indication of autonomic changes rather than tone, which as something which is constantly changing makes it difficult to draw intra-subject comparisons, let alone inter-subject comparisons, considering the large amount of inter-subject variation (Clifford, 2002) as well as the large number of different variables which may affect HRV. Even when subjects are grouped or selected to be as similar to each other as possible, there is still significant inter-subject variation, making group HRV measures difficult (Clifford, 2002).
3.7.2 Ectopic Removal

Data collected by the Polar monitor were transferred onto a computer to be analysed. Raw data were exported to Notepad as a list of HR inter-beat intervals in milliseconds (ms). HR in ms was the form required by the Software for Advanced HRV Analysis (Niskanen et al., 2002) which uses HR data in ms to create a table of HRV parametric and non-parametric data. (Although not necessary for the study, HR data in ms can be easily converted into a HR in bpm by the formula; bpm=1000x(60/ms), where ms is substituted with the value in milliseconds to be converted to HR in bpm, for example 1000ms equates to a 60bpm HR and 750ms equates to an 80bpm HR).

A datum point was considered an ‘outlier’ if it was outside of three standard deviations of the total set of data points. This definition was set by the researcher and his supervisors in order to identify significantly erroneous data points in the data set, and was believed to be sufficient to capture the majority if not all ectopic beats, while not being so narrow as to exclude legitimate data. However a later review of literature uncovered more precise and valid methods of identifying and dealing with ectopic beats than were used in this study as described in section 2.7. Ectopic beats were replaced with an average of the two preceding datum points.

3.7.3 Drop Outs

Twenty participants were recruited for this study. All participants attended all four sessions allowing 80 sets of data to be collected. Four participants were excluded from the final results, two for medical reasons and two due to insufficient data being collected in at least one session each.

- Participants 2 and 7 both left sessions early resulting in insufficient data in their post-intervention relaxation period for analysis and so were excluded from the final results of the study.

- Participant 8 was excluded due to a medical condition (Cystic Fibrosis), which though not specifically outlined in the inclusion/exclusion criteria, was on reflection considered a significant enough medical condition to warrant exclusion from the study.
- Participant 19 was excluded due to what was believed to be an undiagnosed medical condition. After this participant’s data were examined it became obvious that the participant was undergoing periods of apparent cardiac cessation where his heart would skip one or more beats, resulting in unusable HR data and so this participant was excluded from the study.

### 3.7.4 Responders

In analysing the data it was found that subjects could be grouped into four distinct groups of responses to the intervention. Between pre-intervention and intervention it was possible for the subject to have an increase or decrease in the low frequency to high frequency (LF/HF) ratio, and the same change was possible between the intervention and post-intervention. This change in LF/HF ratio was calculated as a raw percentage (i.e. a percent change – increase or decrease – between pre-intervention and intervention) and graphed (section 10.3). The mean of all participants percent change data are displayed in section 10.6. The four possible responses a subject could have between pre-intervention to intervention, and intervention to post-intervention were:

1) an increase followed by an increase (i.e. SNS activity predominates)
2) an increase followed by a decrease
3) a decrease followed by a decrease (i.e. PNS predominates)
4) a decrease followed by an increase.

A desirable response was one which showed a decrease in LF/HF, as a decrease in the LF/HF ratio is believed to indicate an increase in the parasympathetic components of HRV, indicating physiological relaxation.

In this way it was considered that grouping subjects with like responses could give a better indication as to any possible response to the interventions. Any subject with a percent decrease either during or after the intervention was considered a possible candidate for inclusion as a positive responder. Subjects with a percent increase from pre-intervention to intervention were considered possible negative responders (though possibly late positive responders). Subjects with only positive percent changes were considered non-responders.
Not all subjects were responders in all situations however, with some showing a response to one intervention and no response to another, and no subject displayed solely a decrease in LF/HF ratio over the four interventions. One responder showed a decrease for both interventions separately, but showed a proportionally similar increase for both interventions combined.

3.7.5 Non-responders

Non-responders were considered to be those subjects which did not show indications of relaxation, as believed to be indicated by an increase in their LF/HF ratio. Although many subjects were indicated to be non-responders in one or more interventions, no subjects showed this response consistently over all the interventions.
4.0 Results

4.1 Major Findings

Data were analysed using analysis of variance (ANOVA) for pre-intervention, intervention and post-intervention data. P values were found to be greater than 0.05 strongly indicating no statistically significant change within and between interventions. This strongly suggests that contrary to popular belief, music and massage do not cause a physiological effect as measured by heart rate variability (HRV). ANOVA of responders [participants who displayed a decrease in low frequency to high frequency (LF/HF) ratio] who were grouped together also strongly showed no effect of interventions on their LF/HF response, further strengthening the conclusion that music and massage may not affect HRV.

Participants were also grouped according to their average heart rate (HR) during the study, and it was observed that participants with a HR lower than 60bpm displayed a statistically significant increase in LF/HF in response to the massage intervention. This suggests that subjects with a low HR may show an increase in sympathetic nervous system (SNS) activity in response to massage.

4.2 Control and General Results

Kruskal-Wallis ANOVA for the pre-intervention, intervention and post-intervention control data for all subjects produced a p value greater than 0.05 indicating no statistically significant change between the three portions of the control data collected.

A decrease in LF/HF ratio in the intervention period seemed visible when the mean of all subjects' LF/HF ratios were graphed (Chart 1), however when error bars (of one standard deviation) were taken into account it was visually clear that the amount of overlap between the values was too great for any true indication of difference. In general the error bars were greater than the mean values indicating a collection of data with widely different values, and with differences too large for any patterns to be apparent.
Analysis of these data is difficult as any change in one individual can be rendered insignificant when averaged with a completely opposite response by another individual. Further, as individuals’ results were found to be as much as 50 times greater or smaller than another’s, many small changes can be easily disguised. Large error bars in these data were formed due to large differences between individual responses.

As there was such a wide variety of possibilities of where the true value might lie for the control data it was impossible to establish a reasonable baseline to which comparisons could be made. The actual true values for the control could have indicated any magnitude and combination of increase and/or decrease in LF/HF, or no change at all.

**Chart 1**

With this level of uncertainty as to where the true value lies, it was concluded that the grouped control data for the LF/HF ratio was not statistically significant and not useful to establish a baseline for comparison of effects of music or massage interventions on HRV as explored using LF/HF ratios.
As with the control, the mean LF/HF graphs for the interventions indicate data too widely spread for true values to be indicated. Additionally the graphs of interventions are too similar to the control for any change due to the intervention to be indicated.

The same (non-parametric) data are presented individually below in the form of scatter graphs. As can be seen in the scatter graphs (Chart 2), the distributions of LF/HF data are very similar across the interventions. Kruskal-Wallis analysis of variance of this LF/HF non-parametric data gave p values of greater than 0.05 (the smallest p value being p=0.063 for the massage intervention).

**Chart 2**

P values for all four interventions were:
- Music intervention p=0.422
- Massage p=0.063
- Music and massage p=0.296
- Control p=0.459
A comparison of the four interventions p=0.574
Four pre-interventions p=0.784
Four post-interventions p=0.931

Music intervention compared to Control p=0.821
Massage intervention compared to Control p=0.407
Music and massage intervention compared to Control p=0.163

Each point on the scatter graphs represents a participant’s LF/HF data (after ectopic beats were removed). Where two participants’ data are the same, or similar, the points overlap on the scatter graph. As can be seen by the clustering of points on the scatter graph, the majority of participants’ pre-intervention, intervention, and post-intervention data are all very similar, with no noteworthy visual change. All the scales on all four graphs are identical. Larger versions of these graphs and the raw data are presented in the Appendix (sections 10.3 and 10.4).

Parametric data were also analysed using ANOVA, and as with the non-parametric data p values were greater than 0.05. However there were a number of zero values in the LF/HF parametric data, and as it was uncertain what effect these values might have on the analysis it was decided to primarily analyse non-parametric data. It is believed that in HRV analysis parametric and non-parametric data generally give comparable results (Malik, 1996).

ANOVA testing of parametric data found high p values; for the music intervention p=0.902, for the massage intervention p=0.485, music and massage p=0.821 and the control p=0.767.

4.3 HRV data
4.3.1 LF/HF

A complete set of parametric and non-parametric LF/HF data were collected for all subjects for all interventions (section 10.4 and 10.7). As with the control data, although some change was indicated in mean values of LF/HF between pre-intervention and intervention, intervention and post-intervention for the music, massage and both music and massage combined
interventions, the standard deviations for this data were very large and so no reliable indication of change could be reported from the data.

### 4.3.2 Relating Resting HR to HRV Responses; Some Findings

As it was noted in the literature the possible effect of music on HR, and that music was chosen from a 60-80bpm range to promote entrainment to resting HR, it was considered of interest to look at subjects’ HR and responses relating to their HR. Subjects were grouped into three groups; those with a HR between 60 and 80bpm, those with a HR above 80bpm and those with a HR below 60bpm. Where subjects had a HR from different interventions, their average HR was considered.

The mean HR of all subjects was 66.8bpm (± 13bpm).

The data are presented in section 10.5.

The majority of subjects (nine out of 16 subjects) were included in the 60-80bpm group, with a mean HR of 70bmp (±8bpm). Two subjects had a resting HR above 80bpm (mean 86 ± 8bpm). Five subjects had a resting HR below 60bpm (mean 53 ± 6bpm).

It was found that there was a very strong relationship between HR and response (as measured by percent change in LF/HF from pre-intervention to intervention and intervention to post-intervention – see section 3.7.4 for explanation of responses) shown by the low HR (HR<60bpm) group (p<0.05 using a Chi squared correlation test). However the correlation was less significant for the average (HR=60-80bpm) HR group (P=0.262) and high HR (HR>80bpm) subjects (p=0.299).

Low HR subjects predominantly showed an increase-increase response to massage, a response to massage which no average or high HR subject demonstrated. These low HR subjects however did not show this same response to music and massage combined possibly indicating that it is massage alone which produces this response, or that the effect of the music counteracted this response.
The strong correlation between the low HR group (HR<60bpm) and their LF/HF % change responses can be seen in charts 3 presented below. In every intervention, four out of the five (80%) low resting HR group showed a similar response, being a decrease-increase response to the music, music and massage, and control interventions, and an increase-increase response to massage. In fact, with the exception of one subject in one instance, all the low HR subjects’ responses fitted into these two groups of response. The predominant grouping of the low HR subjects was into the decrease then increase group (13 out of 20 instances), with the remainder (bar one) in the increase-increase group (6 out of 20 instances). Additionally the four low resting HR subjects were the only subjects in the entire experimental group to have an increase-increase response to the massage intervention. From these findings it would appear that subjects with a low resting HR are strongly likely to demonstrate an increase in LF/HF ratio in response to massage, and at all other times will demonstrate an initial decrease followed by an increase in LF/HF ratio.

Average HR subjects showed a decrease-increase response to massage, which although not statistically significant (p=0.262) was still of interest, due to the large grouping of these subjects together in the massage intervention. As with the low HR subjects which made up the entire group of increase-increase responders to massage, so the average HR subjects made up almost the entire decrease-increase responder group to massage. Thus it is indicated that subjects with an average HR are likely to have a decrease-increase response in their LF/HF ratio relating to massage. This response potentially relates to a reduction in sympathetic activity (relaxation) in response to massage, with a return to normal and/or increase in sympathetic activity after the massage intervention is concluded. However further study is needed to test this.

The number of high HR subjects (two) was considered insufficient to analyse for trends.
4.3.3 Explanation of Charts

The four charts represent the four interventions, Music, Massage, Music and Massage, and the Control. The charts are presented in the same order as listed.

Three HRV samples were extracted for each intervention (Pre-Intervention, Intervention, Post-Intervention), and the percent change between them was graphed, then subjects were grouped with other subjects with the same type of response. The responses related to either a percent increase or decrease in LF/HF from between the three samples. Thus four groups were evident, those with a % decrease followed by a % increase, those with a % decrease followed by another % decrease, those with a % increase followed by a % decrease, and those with a % increase followed by another % increase. The four groups are represented together on each of the charts separated by a blank space between groups. The scales on all four charts are equal, and span from -150% to 250%.

The vertical black lines delineate between the four different grouped responses. Black circles indicate the five subjects identified as having low (<60bpm) heart rates (Chart 3). As can be seen these five subjects...
consistently group together, predominantly in the decrease-increase response, with a secondary grouping in the increase-increase response for the massage intervention.

In this series of charts (Chart 4) the black circles indicate the participants with average heart rates (60-80bpm). As can be seen in the massage intervention, these participants predominantly group in the decrease-increase response category.

Larger versions of these graphs are presented in section 10.3.
4.3.4 Individual Inconsistencies

There was no noticeable consistent pattern within individuals’ results. Both parametric and non-parametric data were collected and compared for percent change between pre-intervention and intervention, and intervention and post-intervention. There was no obvious similarity observed between subjects’ parametric and non-parametric results either in value or direction of change. Subjects with a large percent change in their parametric results did not necessarily have an equivalent change in their non-parametric results, and subjects with an increase in one did not necessarily show an increase in the other.

Further, there was no consistent pattern between interventions either. Subjects could show an increase with massage and a decrease with music and massage combined or vice versa. Essentially every permutation of possible outcome was obtained in the data gathered with no obvious pattern.

This inconsistency amongst individual data is why group analysis of data is less useful, as HRV is very individual and variable from individual to individual, and it may not be possible to control variables sufficiently to obtain statistically significant data from individuals due to the significantly large day to day changes in individuals’ HRV data. These issues are addressed in the discussion section.

4.3.5 Statistical Significance

Separate ANOVA tests were performed on all four intervention groups, all resulting in high p values, indicating no statistically significant change within the grouped data. However the relatively lower p value (p=0.063) for the massage intervention suggests further study of massage would be worthwhile.

Although there was no strong statistical basis for doing ANOVA testing on ‘responder’ groups, this was done simply for interest. However even when ‘responders’ were grouped together, ANOVA testing found p values lower than whole group data but still much greater than 0.05. This being the case it was conclusively decided that there was no statistically significant change indicated in any of the LF/HF data collected (both the parametric and non parametric
data as tested for statistical significance using a one way ANOVA and Kruskal-Wallis test respectively) either as a whole group or even as sub-groups with similar responses.
5.0 Significance / Outcomes

The finding of no statistically significant change in the recorded data was considered the primary finding of this study. The significance of this finding hints at the possibility that neither music nor massage have a physiological effect on the autonomic nervous system (ANS) in normal healthy males aged 18-35 years.

The finding of increased low to high frequency (LF/HF) ratio in response to massage alone among the low heart rate (HR) sub-group indicates that massage may in fact cause a sympathetic response, rather than a parasympathetic response. This finding warrants further research.

A low p value (p=0.063) found for the population for the massage intervention may indicate that with selective sampling (i.e. the removal of low HR subjects) a statistically significant decrease in LF/HF may be found. This finding also warrants further research.

Due to these findings the questions which ought to be raised are whether heart rate variability (HRV) is the appropriate method to determine physiological change in response to music and massage interventions, and/or whether music and massage are viable options for producing physiological changes.
6.0 Discussion

6.1 Finding of No Change

The purpose of this study was to determine if music, massage or both combined had an effect on heart rate variability (HRV), and by extension the autonomic nervous system (ANS), and if an effect was present to compare between the interventions. A decrease in low to high frequency (LF/HF) ratio indicating parasympathetic activity was hypothesised. However due to reports of sympathetic responses in the literature review, the possibility of an increase response was not disregarded.

Despite the hypothesis, the findings of this study indicated that no statistically significant change (p values greater than 0.05) in the LF/HF component of HRV was found in response to music, massage or both combined. However the relatively lower p value found in the massage intervention may indicate that with further study a change may be found. Additionally, when the sub-group of low heart rate (HR) (HR<60bpm) responders was examined, it was found that they actually displayed an increase in LF/HF, indicating sympathetic dominance. Several reasons were considered as to why no population change was found, and are discussed.

The reasons no change in HRV was detected may be because:

- there was no effect
- there was no effect in response to this particular type of music and/or massage selection
- the effect was too small to be measured
- the instrumentation was not sensitive enough to detect any effect
- the effect was masked by other variables
- any effect of responders in the population was masked by opposite effects from non-responders.

Although the reviewed literature indicated that similar studies on the effects of music and massage indicated a change in LF/HF, the results of this study indicate the possibility that there is no physiological effect of music or massage on LF/HF. This does not negate however the possibility that an effect to these interventions does exist. For example there may be a
strong psychological or emotional effect of either music or massage, or a physiological effect which is not detected by changes in LF/HF. However as these variables were not studied or specifically reviewed in this paper, no inference as to any effects will be drawn. It is the primary finding of this study that there was no statistically significant change in LF/HF in response to music, massage or both together.

Additionally it was considered that the type of music or massage may relate to the results found. In this study the music used was within a range of 60-80bpm, of tracks chosen from easy listening and classical genres. Massage was performed in an effleurage style using slow gliding strokes. Additional study into different genres and tempos of music as well as different speeds of massage stroke and/or style of massage may help to broaden the extent to which the current findings may be generalised to the population.

It was also necessary to consider the possibility that an effect existed, but that it was too small to be measured. If this were the case, it is possible that an effect would be detected by more sensitive instrumentation. However, if an effect exists which is so small it is indistinguishable from background noise, or too small to be detected by standard equipment, it raises the question as to whether such an effect is significant or relevant.

Similar, an effect may not have been detected by the instrument used due to insensitivity of the instrument to detect HR. In this case any effect could have been large, but in some way undetectable by the Polar HR monitor. Although this possibility could not be disregarded outright, it was considered very unlikely, as there was no indication either in the data collected or in the literature that the Polar HR monitor was in any way insufficient to the task of collecting HR data. HR data appeared normal for all but one subject, who was excluded from the study.

Another possibility considered was that if an effect was present it may have been masked by other variables. Many of the variables known or believed to affect HRV are discussed in sections 6.3 and 6.4 below. However, many variables considered limitations may not be able to be controlled in clinical practice, and so even if an effect is masked by such limitations it must be considered whether such an effect in clinical practice is of a sufficient size to be meaningful. Discussed in section 6.3 is a ranking of activities affecting LF/HF (Bernardi,
Wdowczyk-Szulc, Valenti, Castoldi, Passino, Spadacini & Sleight, 2000; Clifford, 2002). As can be seen by this ranking system, the results found in this study are not smaller or greater than typical clinical activities, such as talking, breathing and mental activities. This supports the belief that if an effect was present but masked by other variables, it was of insufficient size to be considered relevant in clinical practice when compared to the effects of other variables on LF/HF.

Finally the possibility was considered that population data showed no change in LF/HF as any decrease in LF/HF by one subject was potentially nullified by an increase by another subject. This was considered the most probable scenario, with the increase in LF/HF demonstrated by the low HR group counteracting the likely \( p=0.063 \) population decrease in LF/HF. Although selective sampling may be successful in demonstrating a statistically significant response to either music or massage interventions, such sampling would reduce the extent to which results could be generalised back to the population, reducing the usefulness of any results found.

### 6.2 Finding of Increased LF/HF in Low HR Participants

Several possibilities were considered to be discussed relating to the reason for the finding of an increased LF/HF in low HR participants. These were:

- that this may be the normal response to massage
- that the type of massage used in the study may have caused an SNS response
- that low HR people may be stimulated by massage, possibly due to their already reduced HR
- that this finding may have been purely co-incidental and non-repeatable.

The finding of an increased LF/HF to the massage intervention may be a normal physiological response. Whereas the normal response for this sub-group between pre-intervention and intervention appeared to be a decrease in LF/HF for the other three interventions, for the massage intervention a strong increase response was shown (see Chart 3). The possibility was considered that this response may be a normal and typical response to massage. This finding is of particular significance, as massage is generally considered to cause relaxation, a belief which may need to be revisited.
Alternatively the type of massage used may have been interpreted by the low HR sub-group as being stimulating, or may have otherwise had a physiologically stimulating effect which resulted in increased SNS activity, leading to the finding of increased LF/HF (a finding which may not be a normal or typical response). This response in turn may relate to the participants’ HR, or be purely coincidental.

With regards to participants’ HR, low HR may be an indication of parasympathetic dominance (Berntson et al., 1997), and as noted in the literature, PNS effects can saturate the HRV response (Goldberger et al., 2001). This in turn would mean that only a sympathetic effect would be demonstrable, as any PNS effects would not alter the saturated HRV response. Thus, if low HR is evidence of PNS saturation, only an increase in LF/HF may be possible. Although this consideration is possible, it was also believed to be unlikely based on the findings of this study. The same participants demonstrated a decrease in LF/HF in the other three interventions leading to the belief that they were not showing PNS saturation.

Finally it was considered possible that this finding was purely coincidental. The number (five) of participants in this sub-group was considered possibly too small to draw any significant conclusions. Additional evidence that this finding may have been coincidence was that this finding was not present in the music and massage combined intervention amongst this sub-group of participants. Unless the use of music completely counteracted the effect of massage, it would be expected that the same effect of massage should be present in both interventions which involved massage. As this was not the case the very real possibility has to be considered that the finding of increased LF/HF among low HR participants is an example of type 1 error – a finding of change where no change exists. A repetition of the study using the same methods would provide a better indication as to whether the finding of increased LF/HF is repeatable.

6.3 Limitations of HRV Measurement

A comprehensive list of factors known and/or suspected to affect HRV is explored below. In providing adequate controls for these variables in future studies any presence or absence of effect can be attributed to the intervention with more certainty.
HRV is affected by a vast variety of activities, both mental and physical. These activities can be ranked by their average LF/HF ratio, from the lowest being controlled breathing at 15 breaths per minute with a LF/HF score of 0.69 (±0.37), reading silently/aloud having a score of 1.52/1.59 (±0.26/±0.21) respectively, performing mentally stressful tasks such as arithmetic with a LF/HF score of 3.05 (± 0.39), and free talking which rates highest at 3.58 (±0.45) (Bernardi et al., 2000; Clifford, 2002). Considering a subject’s physical and mental activity has such a vast effect on HRV, inter- and intra-subject comparisons of HRV must take into consideration all these factors as possible influences on any results found (Clifford, 2002; Clifford & Tarassenko, 2005), and even strictly controlled studies can be confounded by changes in a participant’s mental activity over the duration of the study, as mental activity is perhaps the single greatest factor affecting HRV (Clifford, 2002). Controlling mental activity however is virtually impossible, with the possible exception of collecting data while subjects are asleep (Clifford, 2002), a method which was not feasible in this current study.

6.4 Effect of Controlled (and Uncontrollable) Variables

Variables believed to be of primary importance relating to the music and massage interventions were the volume of the music (and the volume and/or amount of additional noise), the type of massage (such as technique, and intensity of force used), and the environment in which the research was conducted (such as temperature and massage bed comfort). A significant number of secondary variables are also discussed below. In general, any factor known or suspected of affecting HR or HRV was considered important enough to be controlled in as much as was possible, and accounted for when not possible to control.

Variables identified by Iñesta et al. (2008) known to affect HR were environmental factors such as temperature, moisture (humidity (Bernardi et al., 2006)), atmospheric pressure, time of day, height, adaptation level and noise, as well as physiological factors such as age, gender, digestion and health state. A meta-analysis on the effects of massage on relaxation identified a number of variables which were considered relevant to the current study for consideration. These were the length of the massage, the gender of the person administering the massage, the environmental variables (such as room selection, lighting, temperature, use of lotion and lotion temperature), position used during massage, length and use of pre- and
post-massage rest periods, sample size, gender, age and health status (Labyak & Metzger, 1997).

Subjects were asked not to eat, drink or exercise for two hours before each session, as these were considered likely to have an effect on HR. Exercise causes HR to increase through vagal withdrawal thus affecting HRV. Additionally metabaroreceptors in the muscle become stimulated and further increase SNS activity (Clifford, 2002). The effect of physical activity on HRV is considered significant enough to require exercise to be controlled (Clifford & Tarassenko, 2005). The effects of food and drink on HRV are discussed later in this section.

6.4.1 Environmental Variables

**TEMPERATURE**

Temperature was considered a vital variable in this research project. Some participants indicated difficulty in relaxing if the temperature was too low, thus it was endeavoured to make room temperature as comfortable as possible for each participant (Bernardi et al., 2006) so that cool temperatures would not distract the participant. In addition to the room temperature, the temperature of the chest strap was considered, after a participant commented it was particularly cold when first attached to the participant.

As data collection was done during winter months, temperature was required to be raised for almost all sessions. This was done with fan, bar or gas heaters and wood fires. Some participants were covered with blankets to keep warm and some chose to remain wearing clothing where possible (it was not possible to wear clothing for the massage sessions). For a few sessions in the afternoon the temperature was warm enough to exclude the use of a heater.

Participants were asked if the temperature was comfortable or warm enough, and the temperature adjusted accordingly if necessary. Most participants were comfortable at the same level of heat, which on the fan heater was the second of three settings, while a few preferred it warmer. Temperature was not measured precisely in degrees. Warm water was used to moisten the chest strap on some occasions.
Thermoregulation is controlled by a number of factors, both internal and external, and can have affects on peripheral resistance to blood flow thus affecting HR, BP and HRV (Clifford, 2002). Increased ambient temperature may cause increased LF, LF/HF and HR and decreased HF, indicating increased sympathetic activity (Bruce-Low, Cotterrell & Jones, 2006; Fox, O'Regan & Matthews, 1991).

Other studies similar to the current one ensured that temperature was controlled, for example the Urakawa and Yokoyama study maintained room temperature at 22ºC (Urakawa & Yokoyama, 2005) while Hatayama et al. (2008) controlled temperature at an average of 26.8ºC.

As temperature can affect HRV it is a variable which needs to be strictly controlled, ideally using a thermostat to ensure consistency of temperature within the session. Temperature should also be recorded for each session to ensure consistency between participants and between sessions, with temperature set at a comfortable level for the majority of subjects.

**LIGHTING**

It was considered that the effect of lighting on participants in this research project to be of minor importance as participants were lying face down for the entire duration of data collection. As participants were lying face down, facing away from the light, and generally shut their eyes to help relax for all sessions (Bernardi et al., 2006), the type and intensity of light made very minimal difference to most participants.

Sessions were lit with a combination of artificial and natural light. Artificial light came from ceiling lighting, and depending on location was either from fluorescent lights or ordinary light-bulbs. Afternoon sessions naturally tended to have a higher proportion of natural lighting compared to morning and evening sessions. A couple of sessions were done at night without any lighting. Some night sessions were lit with wall lights rather than ceiling lights.

Light is connected with an alert or wakeful state in humans, and affects biological functions and behaviour (Cajochen, 2007) causing a possible sympato-excitatory effect (Schäfer & Kratky, 2006). Bright natural light may improve mood (Kaida, Takahashi & Otsuka, 2007; Knez
& Niedenthal, 2008) and is believed to be more beneficial to health than artificial light (McColl & Veitch, 2001; Veitch & McColl, 2001). Ultraviolet light may reduce pain (Taylor, Kaur, LoSicco, Willard, Camacho, O'Rourke & Feldman, 2009) and fluorescent light may cause stress (Küller & Laike, 1998). A “dimly lit” room is considered beneficial to subject comfort (Urakawa & Yokoyama, 2005).

Proper controls of lighting would dictate data be collected in a blackened room lit with the same light amount/intensity and colour for all participants and for all sessions, in order to maximally control any effects of lighting on HRV.

**TIME OF DAY**

It was intended that participants have all four of their sessions at similar times of the day on successive days; however this was not possible for all participants. Although generally most participants had all their sessions at similar times of the day, some had sessions at different times of the day, and between all participants data was collected at all times of the day. Generally sessions fell into three main times of day, morning, afternoon and evening, and only a few pre-breakfast and late night sessions.

In one study of performing musicians HR recorded at the same time of day was found to be almost identical (Iñesta et al., 2008), indicating the importance of the timing of data collection. It is important to collect data at the same time of day to avoid any diurnal/circadian variation to the data (Urakawa & Yokoyama, 2005).

Ideal controls would dictate a fixed start time for all sessions; however this would mean only one piece of data collected per day, making for very lengthy data collection times, and is not a practical means of data collection. The next most ideal scenario would be for data to be collected at as close as possible to the same time of day for individual subjects, i.e. if a subject’s first data are collected in the afternoon, all further data should also be collected in the afternoon.
LOCATION / ROOM SELECTION

It was endeavoured to gather all data in the same location, however for some subjects this was not practical or possible. As much data as possible was collected in the student clinic at Unitec during normal working hours. Data was collected in one of the treatment rooms which was isolated from the rest of the clinic to reduce distractions and interruptions from other students and patients in the clinic. Sessions outside of the clinic were done in participant’s homes, generally in a quiet bedroom in the house. It was endeavoured to ensure that the house would be vacant except for the participant and the researcher to minimise distractions from others.

Ideal controls would involve data being collected in a private room (Hatayama et al., 2008) with all data collected in the same room.

BACKGROUND NOISE (WHITE NOISE) AND VOLUME

During all sessions there was some level of background white noise throughout the session. White noise was not deliberately or intentionally produced for the experiment, but instead was an artefact of general day-to-day activities, for example traffic. Typically white noise was produced by the same sources for each session, the main ones being the researcher’s laptop, the heater in the room and traffic outside. Most white noise was considered unavoidable, as it was not able to be changed or removed from the experimental setting without significant expense and difficulty.

For a few sessions there was additional or unexpected white noise. Some examples of these were lawn-mower noise in the background for one session, and a heavy rainfall during one session. Some other background noise examples were other people in the room or an adjacent room, or entering the room during the experiment.

It was considered that distractions such as loud noises, coughs and sneezes could potentially affect the experiment (Greene & Sutor, 1971). Further, one study indicates that ambient noise may also affect HRV as indicated by an elevated LF in employees exposed to high job strain and/or high noise levels (van Amelsvoort, Schouten, Maan, Swenne & Kok, 2000).
Noise (as a source of stress) with an intensity of 95dB was shown to result in significant increases in LF/HF ratio as well as LF, suggesting a sympathoexcitatory effect on the heart and HRV (Tzaneva, Danev & Nikolova, 2001). This indicates the need for appropriate volume controls. Music volume in this study was adjusted to the subject’s preference before the study began, and was adjusted for comfort and preference during the study as needed. Possibly a better form of control would be to have allowed the subject to control the volume of music themselves as was done in one study, by having a volume control by the subject (Urakawa & Yokoyama, 2005).

Ideal controls of white noise would involve a soundproofed room (Greene & Sutor, 1971; Hatayama et al., 2008; Urakawa & Yokoyama, 2005) to prevent outside noise from affecting the experiment, or alternatively the use of headphones (Bernardi et al., 2006). Music would be played on a machine in another room to speakers in the experiment room so as to avoid white noise from the electronic device delivering music. Heat would be produced from a relatively non-noise producing machine such as a bar heater as opposed to a fan heater. The experiment should be conducted in silence.

**Distractions**

A number of additional factors were identified which may have had an effect on participants’ data. Some of the more major of these factors were the potential effect of the participant engaging in conversation with the researcher and the potential effect of other occupants in the room or entering the room during the session.

It was endeavoured as much as was possible to conduct all sessions in silence, however a few participants had questions which they only thought to ask after the data collection had begun, generally relating to the research regarding topics which were not directly covered in the information sheet which they received. Some common questions asked of the researcher included an interest in the ongoing results, (with participants asking whether general results to date showed much of a change, or in the case of follow up sessions whether the participant’s previous data showed any change) and general queries relating to HRV, how it works and relates to HR in general. With regards to enquiries about outcome measures it was explained that results could not be disclosed until after the conclusion of all data collection.
In most such cases participant questions were answered by the researcher to the satisfaction of the participant. The researcher made it a particular point to not initiate or prolong conversation in order to promote silence for all sessions. In cases where participant conversation appeared to be persistent, the participant was asked to remain silent for the intervention part of the session.

Although time was allowed at the beginning of each session for participants to ask questions, and though a few did make use of this time, it appeared more common for participants to think of questions after lying down and beginning the ten minute pre-intervention relaxation period. Due to strict data collection times it was considered impractical to allow additional extra time before sessions began to allow participants to sit in silence to attempt to think of all the questions they might have.

As mentioned in section 6.3 with regards to the limitations of HRV, free talking was indicated as having a high LF/HF (3.58) compared to other activities such as metronome paced breathing (Bernardi et al., 2006; Clifford & Tarassenko, 2005). Strict controls for future studies would be recommended, providing additional time before beginning data collection to allow for questions and conversation, as well as specific requests made to participants to remain in silence for the duration of the experiment.

Another distraction noted was one instance of a clinic tutor knocking on the door and entering the research room briefly. This interruption happened during the pre intervention relaxation period of one participant. Additionally a few of the houses used for data collection had other occupants in the house in other rooms. As previously discussed these additional distractions may potentially affect HRV and should be properly controlled, by ensuring data are collected solely in a private room free from distractions.

A further form of distraction was the use of cell phones by participants, a form of activity which potentially falls under the category of mental activity (Bernardi et al., 2000; Clifford, 2002; Clifford & Tarassenko, 2005) which could affect HRV. Participants who used cell phones were asked to put their phones away during the study.
Another distraction which was noted was participants observing their HR on the Polar wrist watch. There were two incidences where participants picked up the Polar monitor because of an interest in seeing what their HR was and/or how the device works. These incidences were brief; the Polar monitor was recovered by the researcher and participants asked to put their head back down and relax.

The use of strict instructions and an isolated room would control the effects of most forms of distractions.

6.4.2 Physiological Variables

EATING / DRINKING

Participants were asked to abstain from eating or drinking for at least two hours before each session, in order to control or eliminate any ANS digestion effects on HRV. In addition to the effects of digestion it is known that caffeine-containing drinks such as coffee are known to affect blood pressure (BP), HR, HRV and the SNS (Bonnet, Tancer, Uhde & Yeragani, 2005; Mort & Kruse, 2008; Sondermeijer, van Marle, Kamen & Krum, 2002) and thus were needed to be controlled or accounted for in the study. Caffeine affects BP through inhibition of adenosine receptors and by increasing the release of certain neurotransmitters (Mort & Kruse, 2008). The effect is reduced with caffeine tolerance (Mort & Kruse, 2008; Sudano, Binggeli, Spieker, Lüscher, Ruschitzka, Noll & Corti, 2005), with decreased LF demonstrated in habitual coffee drinkers (Rauh, Burkert, Siepmann & Mueck-Weymann, 2006) and increased BP and SNS activation in non-habitual coffee drinkers (Sudano et al., 2005). However a study of normal young adult subjects who were administered caffeine prior to sleep found that LF/HF ratios were significantly higher during rapid eye movement sleep after caffeine administration compared to placebo indicating caffeine increased sympathetic activity regardless of caffeine tolerance (Bonnet et al., 2005). Also the ingestion of caffeine tablets may lower HF and increase LF/HF ratio (Sondermeijer et al., 2002). In contrast to this however an increase in LF in subjects who drink three or more cups of coffee per day was indicated (Tsuji et al., 1996), meaning that the presence and type of response may relate as much to conditioning as to the pharmacological effects of caffeine (Flaten & Blumenthal, 1999; Quinlan, Lane & Aspinall, 1997; Quinlan, Lane, Moore, Aspen, Rycroft & O'Brien, 2000).
For most participants compliance to abstinence of food and drink was satisfactory, with sessions conducted at least two hours after eating. However the occasional participant forgot they were asked to not eat or drink before the study. Some participants ate light snacks or drank before the study. No participants reported having eaten an entire meal within two hours of the study, though one or two had eaten just over two hours before the study.

Further review raised the question as to whether setting the limits for no drinking at two hours was sufficient, however, as caffeine has a half-life of 3-6 hours (Mort & Kruse, 2008), meaning that the effects of drinking coffee or caffeine containing drinks are likely to last at least this long. Caffeine levels peak at 30-120 minutes after intake, while BP changes occur within 30 minutes of intake, peak at 1-2 hours, and persist for more than 4 hours (Mort & Kruse, 2008). In retrospect it may have been more appropriate to set limits for caffeine consumption to none for at least 12 hours before the study.

**Respiratory rate**

Respiration has a large influence on HR and HRV (Berntson et al., 1997; Malik & Camm, 1995). As briefly mentioned in section 2.3.1, HRV displays rhythmic changes relative to respiration, a phenomena known as respiratory sinus arrhythmia (RSA). RSA results in gradual lengthening on expiration and shortening on inspiration of R-R intervals (Guyton & Hall, 1996; Yasuma & Hayano, 2004) due to the change in thoracic pressure caused by respiration. On inspiration, intra-thoracic pressure decreases allowing greater venous return. This results in an increased stretch to the right atrium causing a local reflex increase in HR (cardio-acceleration) (MacArthur & MacArthur, 2000; Yasuma & Hayano, 2004). The reverse (cardio-deceleration) happens on expiration (Clifford, 2006).

RSA is mainly mediated through efferent vagal activity (Akselrod, Gordon, Madwed, Snidman, Shannon & Cohen, 1985; Berntson et al., 1997; Clifford, 2002; Malik et al., 1996; Malik & Camm, 1995), and as such is believed to indicate the level of vagal activity in the heart. Thus the HF component of HRV which relates to vagal (parasympathetic) activity is strongly connected to RSA (Clifford, 2002). However it appears that a RR lower than 10 breaths per minute tends to affect the LF band instead (Grossman, Wilhelm & Spoerle, 2004; Holmqvist, Stridh, Waktare, Brandt, Sornmo, Roijer & al., 2005), and so metronome controlled breathing
at a rate of 0.25 Hz (15 breaths a minute) is recommended in order to control the effects of breathing and RSA and keep RSA within the HF band, as well as improve the internal validity of the study (Berntson et al., 1997; Brown, Beightol, Koh & Eckberg, 1993; Igarashii & Budgell, 2000). In spite of this, the beat to beat changes caused by RSA are generally ignored (Clifford, 2006). Further, as metronome paced breathing (0.2 Hz) can increase HF (Jáuregui-Renaud, Hermosillo, Márquez, Ramos-Aguilar, Hernández-Goribar & Cárdenas, 2001), the control of respiration may bias HRV results.

**POSTURE**

HRV changes between upright and supine positions (Malliani, Pagani, Furlan, Guzzetti, Lucini, Montano, Cerutti & Mela, 1997). Active change of posture caused significant increases in HR and LF/HF ratio (Jáuregui-Renaud et al., 2001).

Upright tilt is associated with increased sympathetic tone (probably through increased β adrenergic neural activity), withdrawal of vagal activity and increased HR (Ingemansson et al., 1998). SNS modulation was found to be most reduced in the right lateral reclining position, a self-protecting position in patients with heart conditions (Chen & Kuo, 1997; Fujita, Miyamoto, Sekiguchi, Eiho & Sasayama, 2000; Kalisnik, Avbelj, Trobec & Gersak, 2007; Kuo & Chen, 1998).

There is believed to be a connection between the SNS and baroreflex function (Formes, Wray, O-Yurvati, Weiss & Shi, 2005). Baroreceptor reflex works through humoral and neuronal pathways in order to maintain circulatory homeostasis when perfusion pressure is altered. For example as BP increases stretch sensitive baroreceptors in the carotid sinus and aortic arch become activated, affecting the ANS by causing an inhibition to the sympathetic supply of the heart as well as increasing vagal output, resulting in decreased HR and vasodilatation (Stauss, 2002). Thus both BP and HRV are affected by orthostatic (standing upright) autonomic responses (Fagard, Pardaens & Staessen, 2001; Guyton & Hall, 1996).

Additionally, medications which affect BP will also affect baroreflexes and HRV (Chern, Hsu, Hu, Chen, Hsu & Chao, 2006; Formes et al., 2005; Pagani, Montano, Porta, Malliani, Abboud, Brikett & Somers, 1997)
Essentially SNS predominates in the upright position, while PNS predominates in the supine position. It was believed that in this study the pre-intervention supine relaxation period was sufficient to account for this change.

**Music Training**

On data analysis some unusual HR data were found with one subject (subject 19). On discussion with this subject it was learnt that one of the pieces of music which the subject chose to listen to had also been a piece of music which they had learned and performed previously. Although the selection of music was generally of reasonably well known tracks, no other participants in the study reported any strong familiarity with the pieces of music which they chose to listen to in the same way as this subject.

Upon a thorough search into the possibility of musical training affects on HR and HRV it was found that there was a very small amount of research into the effects on musicians of playing and performing. Indicated in the research was that musicians display a higher HR when performing music, even if the piece being performed is known to have a relaxing affect on HR (Iñesta et al., 2008; Mulcahy et al., 1990), and that musicians have a lower RR than non-musicians at baseline and during listening to music (Bernardi et al., 2006).

Additionally structural and functional changes in musically relevant brain regions (for example sensorimotor brain areas) have been found in adult musicians (Hyde, Lerch, Norton, Forgeard, Winner, Evans & Schlaug, 2009) indicating music and the learning of an instrument affects brain development. Non-musicians listen to music using the non-dominant hemisphere of the brain while musicians use the dominant hemisphere (Bernardi et al., 2006) a fundamental difference which may be significant in relation to HRV.

With regards to these findings it can be speculated that the effects of music training on HR will also affect HRV, and that subjects with music training should be studied separately from subjects without music training.
**EXERCISE TRAINING**

As with music training, HRV displays changes with regular exercise. Over time regular exercise is believed to modify autonomic balance, causing a higher maximum and lower resting HR and faster sympathovagal recovery (Clifford, 2002).

As with music training, subjects with exercise training should be studied separately or excluded from the study due to the potential differing baseline HRV levels compared to non-exercising subjects.

**GENDER**

As the researcher performed all massage sessions in this study, this variable identified by Labyak and Metzger (1997) was considered sufficiently controlled. However some other related issues were identified by the researcher such as the effect of the researcher-subject relationship, and the effect of researcher posture, comfort and mood on the massage intervention. These are discussed below.

**THE RESEARCHER-SUBJECT RELATIONSHIP**

Through the process of selection and recruitment it was endeavoured to find participants who were not in any way related to or associated with the researcher. However as participants were largely recruited from among the Unitec population it was inevitable that some participants would be known by the researcher. As a result, the potential for how well or poorly the researcher and participant knew each other to affect participant comfort and relaxation was considered.

It is possible there may have been some kind of experimenter effect (Hoyt et al., 1979) relative to the relationship between the researcher and participant (ranging from complete stranger to fellow student). However little research could be found to quantify this variable. Ideally all participants should be previously unknown to the researcher in order to ensure the researcher-subject relationship is equal for all participants.
RESEARCHER POSITION, COMFORT AND MOOD

A number of sessions were conducted in participant’s homes on participant’s beds which were considerably lower than an Osteopathic treatment table. As a result it was observed by the researcher while conducting the interventions which required the application of massage that the additional stooping required to accommodate the lower bed heights caused some discomfort to the researcher in some situations. As a result it was considered that the increased discomfort and tension felt by the researcher may have affected the participant receiving the massage, though none was reported. Intentionality (the way the therapist presents themselves through their presence, the manner of communication used, and the therapist’s intent to produce a therapeutic response in the patient) (Kutner et al., 2008) may have been affected, reducing the active intention to affect the ANS due to poor posture and discomfort. Intentionality may also be affected by researcher mood with potential repercussions on data collection.

Ideally controls would have all sessions conducted on a proper Osteopathic treatment table to reduce any chance of discomfort to the person administering the soft tissue massage.

PRE-SESSION ACTIVITIES

Due to time constraints on some subjects, it was necessary for the second session to be done without a great deal of washout time in between. Data were collected from subjects when they were available, which was not necessarily a fixed time period in the day, nor necessarily a fixed day of the week. While for one participant it was possible for sessions to be held Tuesday and Thursday evenings on consecutive weeks at approximately the same time of night each night, for another participant there was several weeks between sessions due to unavailability.

One participant attended a session beginning by mentioning they’d just had a nap and were already feeling quite relaxed, causing it to be considered that some form of control for pre-relaxation activity if possible should be implemented. Controls for pre-relaxation activities such as massage, listening to music, napping or a previous session should be implemented to ensure participants begin study sessions at the same baseline levels of autonomic activity.
One possible solution to ensure comparable baseline levels would be the introduction of a stress test, such as an arithmetic stress test (Ruediger, Seibt, Scheuch, Krause & Alam, 2004), prior to each session. Ideally sessions should be conducted at the same time of day on successive days for all participants.

**USE OF LOTION**

It was decided not to use any massage lotion or wax for any of the sessions. The reasons for this included the potential for a bias towards the sessions with massage due to the aroma of the wax or oils (Chang, 2008). Aromatherapy has been shown to reduce pain, depression and anxiety scores and improve sleep and quality of life scores in a variety of studies (Chang, 2008; Soden, Vincent, Craske, Lucas & Ashley, 2004; Wilkinson, Aldridge, Salmon, Cain & Wilson, 1999; Yip & Tam, 2008) and inhaling essential oils in conjunction with soft music was found to increase PNS activity in healthy young individuals (Peng, Koo & Yu, 2009). Thus the use of scented waxes or oils would potentially complicate or bias the results. Additionally, the use of massage wax/oil is not as common amongst Osteopaths as it is with massage therapists, and it was not considered necessary for this study which was using Osteopathic soft tissue massage as the intervention.

Using no lotion for any sessions provided better controls than using lotion for some sessions and not others, or some participants and not others.

**THE INTERVENTION DURATION**

As HRV data needs to be collected over a window of at least five minutes (MacArthur & MacArthur, 2000) the intervention time was required to be at least this long. A duration of ten minutes was chosen to allow a sufficiently long enough window of data collection to gain enough usable data, as well as to have enough data to exclude the first and last data of each segment collected. Ten minutes was also considered the minimum time to be long enough for participants HR to reach resting relaxation baseline before the intervention started. Generally participants showed a trend to baseline within the first three minutes of pre-intervention relaxation.
Although it is possible the duration of massage may affect HRV (Labyak & Metzger, 1997) it was considered that as all participants received massage for the same period of time that this variable was adequately controlled and that a ten minute massage period was sufficiently long enough.

6.5 Summary

By providing ideal controls using the above described methods and eliminating all possible limitations, any effect if present could be attributed with more certainty to the intervention being studied. However as inferred earlier, if an effect is not of sufficient size to be found in spite of what are for the most part normal clinical variables, it has to be questioned as to whether such an effect is in fact clinically relevant.
7.0 Conclusion

The findings of this research are that there are no statistically significant changes to heart rate variability (HRV) as measured by the low to high frequency (LF/HF) ratio to either music or massage interventions of ten minutes duration in healthy (young) adult male subjects.

Due to the findings of this research paper two possible conclusions have been drawn. Firstly, that there may not be a physiological effect of either music or massage as measured by HRV. And secondly, that HRV may not be the appropriate measure of physiological response to music and massage interventions, due to the large number of factors which affect HRV. A larger scale repeat of the study with strict controls may strengthen these hypotheses.

An additional side finding to the study’s aim was that soft tissue massage appears to cause a specific response relative to a person’s heart rate (HR) during the study, being a strong trend towards sympathetic activation (increased LF/HF ratio) in subjects with a low [less than 60 beats per minute (bpm)] HR, and in normal HR subjects (60-80bpm) a LF/HF decrease followed by a post intervention LF/HF increase, indicating a slight parasympathetic dominance during massage followed by a post massage increase in sympathetic activity.

Only if all possible variables are fully controlled can changes in HRV begin to be attributed to an individual intervention, and multiple testing of individual subjects to the same intervention stimulus, as well as large scale studies would be required in order to begin to counteract the amount of inter- and intra-subject variability found in this study.
8.0 Recommendations

Due to the finding of no change in this study it is recommended that future studies implement very strict control protocols in order to eliminate as much as possible any influence of extraneous factors on heart rate variability (HRV). Most especially the effect of mental activity, which has the greatest influence on HRV (Clifford, 2002), ought to be controlled if possible. Potentially one method to do this is through the use of metronome paced breathing and strict instructions to the participant to concentrate only on the music and/or massage, and breathing during the non-intervention periods during the study. The use of a sound-proofed room is also highly recommended.
9.0 References


Larsen, P. D., & Galletly, D. C. (2005). The sound of silence is music to the heart. Heart (December 9).


A comparative study of music and soft tissue massage on heart rate variability.

I have had the research project explained to me and I have read and understand the information sheet given to me. I have had an opportunity to ask questions and have had them answered.

I understand that I don't have to be part of this if I don't want to and I may withdraw at any time prior to the completion of the research project.

I understand that everything I say is confidential and none of the information I give will identify me and that the only persons who will know what I have said will be the researchers and their supervisor. I also understand that all the information that I give will be stored securely on a computer at Unitec for a period of 5 years.

I understand that I can see the finished research document.

I have had time to consider everything and I give my consent to be a part of this project.

Participant Signature: ………………………….. Date: ……………………… ……

Project Researcher: ……………………………. Date: ……………………………

UREC REGISTRATION NUMBER:2009-936
This study has been approved by the Unitec Research Ethics Committee from 30-4-2009 to 30-4-2010. If you have any complaints or reservations about the ethical conduct of this research, you may contact the Committee through the UREC Secretary (ph: 09 815-4321 ext 6162). Any issues you raise will be treated in confidence and investigated fully, and you will be informed of the outcome.
10.2 Participant Information Sheet

Information for participants

A comparative study of music and soft tissue massage on heart rate variability.

My name is James Morgan and I am a Master of Osteopathy student at Unitec. Part of my degree programme involves a research paper on a subject of my choice. My research topic looks at the effects of soft tissue massage and music on heart rate variability. I am doing the research at (your) school and have the approval of the school to carry out the research.

1. Introduction to the Research and invitation to take part.

Relaxation is an important aspect of our lives, and there are many different things which can be done to help promote relaxation. Some of these things include listening to music or receiving a relaxing soft tissue treatment. Heart rate variability (HRV) is commonly used to give an indication of the autonomic nervous system (ANS) which in turn gives an indication of physiological relaxation through the way in which its two main parts – the sympathetic and parasympathetic nervous system – cause excitation and relaxation of various parts of the body, including the heart. You are invited to be part of a study that will help give us a better indication of the effects of music and soft tissue technique on HRV.

2. What is the research study about?

The aim of this experiment is to investigate whether the effects of a soft tissue massage intervention causes a greater effect on HRV compared to music, and whether the effects of both combined are greater than each individually.

3. What will I have to do?

As a volunteer you will be required to attend the laboratory on four separate occasions. You will be asked to choose approximately 10 minutes worth of music to listen to out of a pre-screened selection. This music is selected from a range of classical and easy listening tracks with a 60-80 beat per minute rhythm, which is shown to have a relaxing effect. You will need to remove your shirt and be fitted with a chest strap which will record your heart rate during the session. Each session will involve lying down for 30 minutes. There will be a 10 minute pre-intervention period, followed by a 10 minute intervention, and a final 10 minute post-intervention period. One intervention will involve listening to relaxing music, one intervention
will involve soft tissue massage to your back and shoulders, one intervention will involve both music and soft tissue massage together, and one will be a control with no intervention. The soft tissue massage will be delivered by the researcher. The order in which you receive the interventions will be randomised. The information on your heart rate is collected by the electronic computer on the chest strap and transmitted to the Polar wrist watch monitor.

4. What are the benefits?

By participating in this study you will make a valuable contribution to the body of evidence based research relating to Osteopathic practice, and the effects of different interventions on HRV, and by extension the autonomic nervous system. It is hypothesised that you will also feel the benefits of 20 minutes rest and relaxation each session.

5. What are the risks?

There are no risks involved in this study. There are no invasive or painful techniques to be used. If you feel mistreated or feel your interests are otherwise being ignored, neglected or denied at any time during the participation in this research study, you should inform the project supervisor.

6. What if I don’t want to take part?

Your participation in this study is 100% voluntary. You will be under no pressure to participate if you do not want to. If you withdraw from this study it will not have undesirable consequences. You will also have time to consider whether or not you wish to participate in this study.

7. What happens to the information?

The information gathered during the course of this study is totally confidential. The information will be used purely for research purposes and at no point will any outside the research team be made aware of any of the results. We may, at some point, wish to publish the results but at no point will your personal data be attributed to you and confidentiality will be practised at all times.

8. Who else is taking part?

The volunteers in this study are from the student body of Unitec. Those on medications causing sedation or with medical conditions which may affect relaxation are excluded from this study.

9. What if something goes wrong?

If anything goes wrong during the study there will be a qualified first aid trained individual present at all times.
10. What happens at the end of the research study?

At the end of the study the data will be analysed to see what, if any, effect music and soft tissue massage have on heart rate variability.

11. What if I have any more questions or do not understand something?

If at any point before deciding to participate, or during the research study, you have any queries about any part of the experiment, you are free to ask the primary investigator or the project supervisor who shall endeavour to answer all questions to the extent of their knowledge to satisfactory standard.

12. What happens now if I decide to take part?

If you want to volunteer, all you have to do is contact the research team on the numbers given below and set up times to attend for the three data collection periods.

13. What happens if I change my mind during the research study?

If at any point while you are still involved in the study you decide that you no longer wish to participate you are free to withdraw your services without any ill feeling. You are under no obligation to continue in the research study if you decide that you don’t want to. You will have two weeks after your final data is collected if you wish your data removed from the study for any reason.

14. Contact name and number.

Name: James Morgan

Tel: 021 268 9675

E-mail: james.is.jimincricket@gmail.com

UREC REGISTRATION NUMBER: 2009-936

This study has been approved by the Unitec Research Ethics Committee from 30-4-2009 to 30-4-2010. If you have any complaints or reservations about the ethical conduct of this research, you may contact the Committee through the UREC Secretary (ph: 09 815-4321 ext 6162. Any issues you raise will be treated in confidence and investigated fully, and you will be informed of the outcome.
10.3 Data Graphs (LF/HF % change and Raw LF/HF scatter graphs)

Grouped (NonParametric) Data - Music

Grouped (NonParametric) Data - Massage

Participant

Percent Change

% Change from PreIntervention to Intervention
% Change from Intervention to PostIntervention
# 10.4 Raw (Non Parametric) LF/HF Data

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10.6 Graphs of Average % Change

**Average % Change (Parametric)**

- Average % Change from PreIntervention to Intervention
- Average % Change from Intervention to PostIntervention

**Average % Change (Non-Parametric)**

- Average % Change from PreIntervention to Intervention
- Average % Change from Intervention to PostIntervention

Error bars = 1 SD
### 10.7 Raw (Parametric) LF/HF Data

#### Music Intervention

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You are invited to participate in my research project for the Master of Osteopathy degree program here at Unitec. I am studying the effects of relaxation music and soft tissue massage on heart rate variability, a measure of the autonomic nervous system and an indicator of levels of relaxation in the body. If you are able to attend four sessions of approximately 40min please consider being part of my research.

For more information contact the researcher:
Name: James Morgan   Number: 021 268 9675
E-mail: james.is.jiminycricket@gmail.com

You may be able to participate if:
- You are a male between 18 and 30 years of age
- You are healthy!

Sorry you can not participate if:
- You do not find music relaxing or are hearing impaired
- You have a known heart or lung or skin condition which may affect the study
- You are on sedative medications (or taking mood altering drugs)
- You cannot lie down comfortably for the duration of the study

UREC REGISTRATION NUMBER: 2009-936
This study has been approved by the UNITEC Research Ethics Committee from 30-4-2009 to 30-4-2010. If you have any complaints or reservations about the ethical conduct of this research, you may contact the Committee through the UREC Secretary (ph: 09 815-4321 ext 6162). Any issues you raise will be treated in confidence and investigated fully, and you will be informed of the outcome.