The Impact of a 12-week High-Intensity Interval Training Program on Sympathetic Transduction in Healthy Adults: A Pilot Project.

by

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Dalhousie University is located in Mi'kma'ki, the ancestral and unceded territory of the Mi'kmaq.
We are all Treaty people.

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Abstract

Peak pressor responses (i.e., sympathetic transduction) following bursts of muscle sympathetic nerve activity (MSNA) are inversely related to peak oxygen consumption ($\dot{V}O_2$peak) in younger males, but not females. However, it is unknown whether high-intensity interval training (HIIT) decreases sympathetic transduction. I tested the hypothesis that 12-weeks of HIIT (n=9) would improve $\dot{V}O_2$peak and attenuate sympathetic transduction versus a Control group (n=5). At baseline and follow-up, $\dot{V}O_2$peak (via graded cycle ergometry), resting peroneal MSNA (via microneurography) and beat-by-beat diastolic blood pressure (DBP; via finger photoplethysmography) were recorded. Peak DBP increases following MSNA bursts quantified sympathetic transduction. Nadir DBP following non-MSNA heartbeats were also determined. HIIT training improved $\dot{V}O_2$peak (P=0.003), with no changes observed in the Control group (P=0.279). Sympathetic transduction did not change in either group (Interaction P=0.647). However, decreases in DBP during sympathetic quiescence were larger following HIIT (P=0.006), but unchanged in the Control group (P=0.638). These results indicate that HIIT-mediated increases in aerobic fitness did not alter sympathetic regulation of DBP but may have enhanced competing local vasodilatory mechanisms.

List of Abbreviations Used

ATP = Adenosine Triphosphate

CO = Cardiac Output

CVLM = Caudal ventrolateral medulla

DBP = Diastolic blood pressure

HIIT = High-intensity interval training

HR = Heart rate

MAP = Mean arterial pressure

METs = Metabolic equivalents of task

MLC = Myosin light chains

MLCK = Myosin light chain kinase

MSNA = Muscle sympathetic nerve activity

NE = Norepinephrine

NTS = Nucleus of the solitary tract

PAP = Peak aerobic power

RVLM = Rostral ventrolateral medulla

SBP = Systolic blood pressure

TPR = Total peripheral resistance

$\dot{V}O_2$peak = peak volume rate of oxygen consumption

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Chapter 1: INTRODUCTION

The estimated lifetime incidence of developing hypertension in Canada is 90% (1). Having high blood pressure is a major risk factor for the development of cardiovascular disease (2). In addition, low aerobic fitness is an independent risk factor associated with a greater incidence of hypertension (3), cardiovascular disease (4, 5), and all-cause mortality (6, 7). Young adults with low aerobic fitness (<20th percentile) are 3- to 6-times more likely to develop cardiovascular disease risk factors (e.g., hypertension, diabetes, metabolic syndrome, obesity) than those with higher aerobic fitness (>60th percentile) (5). Thus, determining effective strategies to improve aerobic fitness and prevent disease development in young populations is of importance to reduce the future burden of disease on our health care system.

In both healthy and disease states, dynamic information regarding the sympathetic neural regulation of blood pressure can be obtained by calculating a commonly implemented metric known as sympathetic transduction (8–16). Sympathetic transduction is used to assess how sympathetic neural outflow (assessed via muscle sympathetic nerve activity (MSNA)) is transduced into a vascular and/or blood pressure response (17). Two common methods of assessing sympathetic transduction include laboratory stressors to evoke sympatho-excitation and spontaneous resting sympathetic transduction (17). Spontaneous sympathetic transduction can provide insight into the dynamic regulation of beat-by-beat blood pressure under normal resting conditions. Fairfax et al. (8) demonstrated that decreases in spontaneous sympathetic transduction (assessed via decreases in forearm vascular conductance) were robustly decreased when α-adrenoreceptors were inhibited (via phentolamine). As such, sympathetic transduction can provide insight into the end-organ response following MSNA. Using the concept of sympathetic transduction, assessing the transient decreases in blood pressure that occur during

periods of sympathetic quiescence (i.e., periods without MSNA bursts) can provide insight into the non-neural regulation of blood pressure. Both sympathetic transduction and non-burst responses have been used to examine research questions concerning variables such as sex, age, race, disease, and aerobic fitness (8–16).

Younger healthy males (13) and older healthy males and females (18) with higher aerobic fitness have lower spontaneous sympathetic transduction than those with lower aerobic fitness. However, no such differences in sympathetic transduction were observed between young females with higher versus lower aerobic fitness (19). Considering that a high-level of aerobic fitness confers positive cardiovascular health benefits (3–5), as well as attenuated vascular and pressor responses following bursts of MSNA in young males and older males and females (13, 18), reduced sympathetic transduction may be indicative of a healthier vascular environment. Greater sympathetic transduction could be associated with why individuals with lower aerobic fitness are at a greater risk of developing hypertension and cardiovascular disease.

Although relationships between sympathetic transduction and aerobic fitness have been observed in cross-sectional research in some populations (13, 18), no research has investigated the impact of an exercise training intervention on sympathetic transduction in healthy adults. In healthy, young male Fisher rats, 12 weeks of aerobic training decreased α-adrenergic-mediated vasoconstriction (20). Thus, if aerobic training similarly blunts α-adrenergic mediated vasoconstriction in humans, as compared to the male Fisher rats (20), an attenuated sympathetic transduction may also be observed when aerobic fitness is improved. As no differences in sympathetic transduction were observed in more fit versus less fit young females (19), decreases in sympathetic transduction following exercise training may only be apparent in young males, and not young females.

High-intensity interval training (HIIT) is a popular mode of activity that can evoke increases in aerobic fitness, while being more tolerable than high volume endurance type training (21). Our lab has used a 6-week cycle ergometer-based HIIT program in healthy older adults (>54 yrs of age) to assess vascular adaptations to aerobic training (22). This protocol increased aerobic fitness (23 ± 7 to 28 ± 7 ml/kg/min). However, this same protocol conducted in healthy young adults did not increase aerobic fitness (40 ± 9 vs 41 ± 8 ml/kg/min), despite observing a higher peak aerobic power (PAP, 207 ± 45 to 217 ± 42 W) (23). High-intensity exercise improves aerobic fitness in healthy adults, particularly those with higher baseline fitness levels (24). However, it is unclear whether this HIIT protocol conducted over a longer period can improve aerobic fitness in younger adults. Studies using various other HIIT-based protocols have demonstrated improvements in aerobic fitness after 12-weeks of training (21). HIIT may be an effective way to elicit decreases sympathetic transduction via increases in aerobic fitness. Coincident with increases in aerobic fitness, aerobic training has been demonstrated to improve vascular vasodilatory mechanisms (e.g., greater endothelial-derived nitric oxide availability) (25). When sympathetic neural influences are low (i.e., non-burst periods), decreases in blood pressure may provide additional insight into non-neural adaptations to exercise training.

The primary purpose of this study was to investigate whether 12-weeks of HIIT would: 1) improve aerobic fitness, and 2) evoke corresponding decreases in spontaneous sympathetic transduction in healthy adults. Based on previous HIIT training studies that observed improved aerobic fitness after 12-weeks (21), it was hypothesized that the HIIT protocol used in this study would increase aerobic fitness. Based on the inverse relationship between sympathetic transduction and aerobic fitness that our group has observed in younger males (13) and older males and females (18), it was hypothesized that HIIT would decrease sympathetic transduction

when aerobic fitness was improved. Although younger females may exhibit no impact of aerobic fitness on sympathetic transduction (19), this study performed a preliminary, exploratory assessment into whether sex impacted sympathetic transduction responses to HIIT. The results of this study may provide further insight into the potential cardioprotective effects of HIIT.

Chapter 2: LITERATURE REVIEW

2.1 **Aerobic Fitness, Sympathetic Nerve Activity, and Cardiovascular Disease**

The estimated lifetime incidence of developing hypertension in Canada is 90% (1). Having high blood pressure is a major risk factor for the development of cardiovascular disease (2). In addition, poor aerobic fitness is an independent risk factor associated with a greater incidence of hypertension (3), cardiovascular disease (4, 5), and all-cause mortality (6, 7). Young adults with low aerobic fitness (<20th percentile assessed via the modified Balke Treadmill protocol) are 3- to 6-times more likely to develop cardiovascular disease risk factors (e.g., hypertension, diabetes, metabolic syndrome, obesity) than those with higher aerobic fitness (>60th percentile) (5). Thus, even in young adults, maintaining a high level of aerobic fitness is of importance for cardiovascular health.

Excessive sympathetic nervous system activity is associated with the development of hypertension and cardiovascular diseases (26). Specifically, compared to individuals with normal blood pressure, those with hypertension have elevated resting sympathetic nervous system activity (27–30). In patients with cardiovascular disease, the highest levels of sympathetic outflow are associated with the greatest cardiovascular disease mortality (26, 31). Worse cardiovascular outcomes are also commonly observed in individuals with poor aerobic fitness (4). However, the precise mechanisms that corroborate the cardiovascular benefits of maintaining a higher level of aerobic fitness are unknown. Repeated large pressor responses following spontaneous sympathetic outflow may lead to increases in vascular resistance and resistance vessel stiffening overtime (32). A higher fitness level may be associated with blunted pressor responses to sympathetic nerve activity (13, 18). In older and younger male Fisher rats, blunted α-adrenergic receptor vasoconstriction to norepinephrine was observed following

aerobic training (20). Lower α-adrenergic-mediated vasoconstriction in response to sympathetic nerve activity may lead to reduced pressor responses. Thus, the sympathetic regulation of blood pressure may be implicated in the cardiovascular benefits of maintaining higher levels of aerobic fitness.

Aerobic exercise training is an effective non-pharmacological intervention to improve aerobic fitness and reduce cardiovascular disease risk in both healthy and patient populations (33, 34). High-intensity interval training (HIIT) is an increasingly popular form of aerobic exercise, which includes bouts of high-intensity exercise interspersed with periods of rest or active recovery. This form of training is more effective at improving aerobic fitness compared to lower intensity, continuous training of a similar volume (24). As such, HIIT may be an effective form of activity to reduce cardiovascular disease risk via improvements in aerobic fitness.

Investigating how improvements in aerobic fitness influence the sympathetic regulation of blood pressure is of importance due to the links between elevated sympathetic nerve activity, cardiovascular disease development (26–30), and aerobic fitness (3–5). This may provide insight into why being aerobically fit is important for lowering cardiovascular disease risk over the lifetime. It is important to first explore how improvements in aerobic fitness alter the sympathetic nervous system in healthy populations, as it remains unclear what mechanisms contribute to the protective nature of maintaining a greater aerobic fitness over the life course.

2.2 **The Sympathetic Nervous System**

2.2.1 Functional Anatomy of the Sympathetic Nervous System

Mean arterial blood pressure (MAP) is proportional to both cardiac output (CO) and total peripheral resistance (TPR) (i.e., MAP = CO × TPR). Thus, blood pressure can be regulated by changes that occur within the heart and the peripheral vasculature. Sympathetic nervous system

activity to the peripheral vasculature is critical for the effective regulation of arterial pressure. Increased sympathetic activity to resistance vessels (e.g., arterioles) in the peripheral vasculature results in vasoconstriction, which increases the resistance to blood flow and arterial pressure. Changes in diastolic blood pressure (DBP) provide an estimate of sympathetic vasoconstrictor outflow with minimal influence from cardiac-specific factors (i.e., changes in CO). That is, DBP rises in response to sympathetic activity serves as a systemic index of corresponding increases in peripheral vasoconstriction.

Regulation of efferent sympathetic nerve activity is mediated, in part, by the arterial baroreceptors. Arterial baroreceptors are a key mechanism through which short-term blood pressure regulation occurs (35). Mechanosensitive nerve endings located in the carotid sinuses and aortic arch respond to changes in transmural pressure (i.e., internal-external pressure) (Figure 2.1) (35). Baroreceptors operate to maintain blood pressure around a centrally determined 'set point'. Acute increases (or decreases) in arterial/transmural pressure results in greater (or lesser) baroreceptor afferent impulses to the central nervous system (35).

Carotid sinus baroreceptor afferent action potentials are transmitted along the carotid sinus branch of the glossopharyngeal nerve (cranial nerve IX), while those generated in the aortic arch are transmitted along the aortic depressor branch of the vagus nerve (cranial nerve X) (36). These afferent nerve impulses synapse bilaterally in the medullary nucleus of the solitary tract (NTS), providing an excitatory input through the release of glutamate (Figure 2.1) (36). Interneurons project from the NTS to the caudal ventrolateral medulla (CVLM) (36). The CVLM has an inhibitory connection (via the neurotransmitter gamma-aminobutyric acid) onto the rostral ventrolateral medulla (RVLM). The magnitude of baroreceptor-specific NTS-to-CVLM

inhibitory control on the RVLM reduces the strength of efferent sympathetic nervous system activity (36).

Neurons from the RVLM project to the intermediolateral cell column in the thoracolumbar regions of the spinal cord and release acetylcholine when they synapse with pre-ganglionic sympathetic neurons (36). Pre-ganglionic neurons exit the spinal cord and release acetylcholine when they synapse with post-ganglionic neurons just outside of the spinal cord in either the paravertebral (sympathetic chain) or prevertebral ganglia (36). Due to the location of the sympathetic ganglia, the pre-ganglionic neurons are short in comparison to the post-ganglionic neurons, which extend to the effector organ (Figure 2.1) (35). Sympathetic post-ganglionic neurons directly innervate the heart, blood vessels (arteries, arterioles, veins, venules), adrenal glands, and the kidneys (36). However, the largest innervation density is on the arterioles, or resistance vessels, which contributes significantly to arterial blood pressure control via changes in total peripheral resistance (TPR) (37).

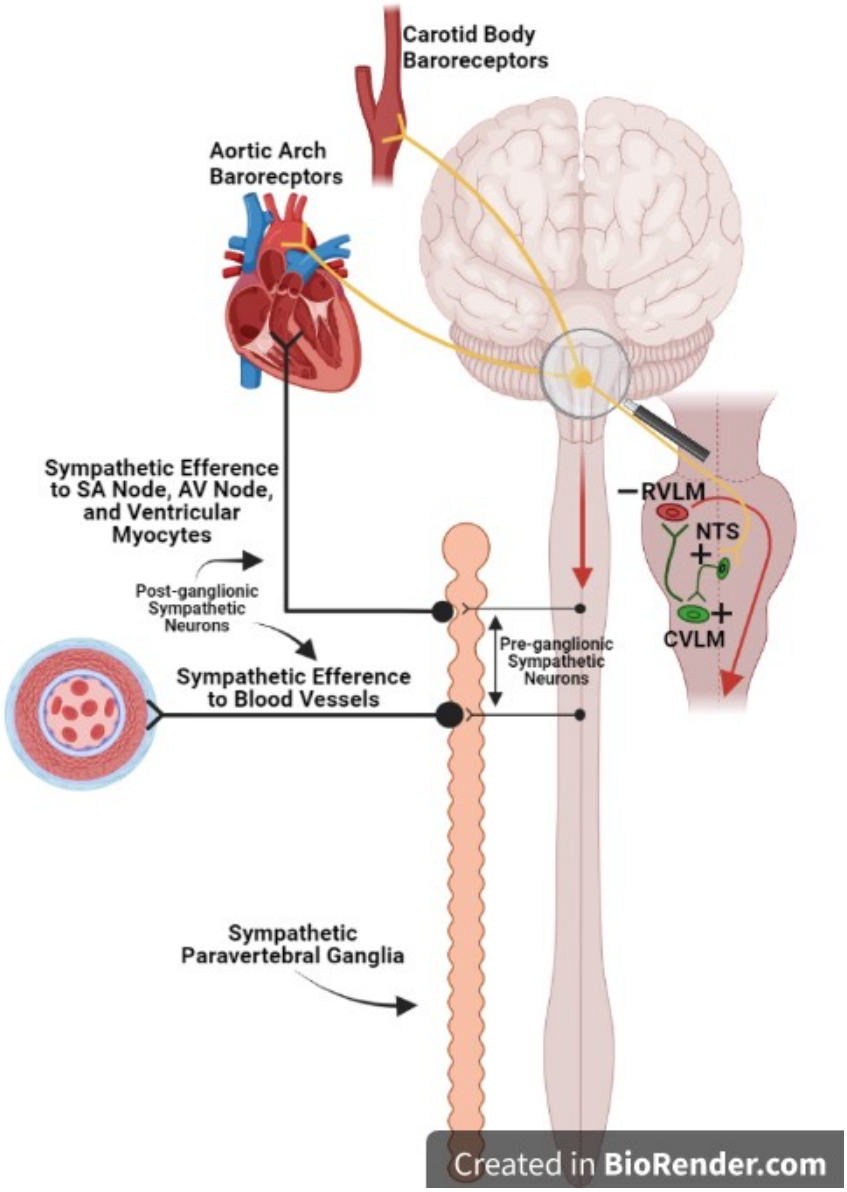

Figure 2.1. Functional organization of the sympathetic nervous system. Higher afferent neural activity from the aortic arch and carotid sinus baroreceptors occurs in response to increases in blood pressure (or vice versa). These excitatory afferent signals are integrated within the nucleus of the solitary tract (NTS) of the medulla oblongata. The caudal ventrolateral medulla (CVLM) receives excitatory inputs from the NTS. Inhibitory interneurons project from the CVLM to the rostral ventrolateral medulla (RVLM). The magnitude of NTS and CVLM inhibitory control on the RVLM alters the strength of efferent sympathetic neural signals (e.g., muscle sympathetic nerve activity). Neurons project from the RVLM through the intermediolateral cell column in the thoracolumbar regions of the spinal cord, where they synapse with pre-ganglionic efferent sympathetic neurons. Sympathetic pre-ganglionic neurons are short and synapse in the paravertebral ganglia adjacent to the spinal cord. Post-ganglionic neurons are long and travel directly to effector organs, which includes the heart and blood vessels. While the sympathetic nervous system does innervate the heart, this project will focus on the influence of the sympathetic nervous system on resistance vessels in skeletal muscle.

Post-ganglionic sympathetic axon terminals are situated in the adventitial-medial border of arteries/arterioles (Figure 2.2) (35). Norepinephrine (NE) is the primary neurotransmitter released from vesicles within the post-ganglionic axon terminals (35). Norepinephrine has the highest affinity for α_1-adrenergic receptors located on the vascular smooth muscle cells, followed by α_2-adrenergic receptors (37). Binding of NE to both receptor subtypes result in contraction of the smooth muscle cells, or vasoconstriction. Vascular smooth muscle cells also express lumen-facing (towards the blood) β_2-adrenergic receptors, which cause vasodilation. However, NE released from post-ganglionic sympathetic fibres binds most readily to α-adrenoreceptors due to the close anatomical proximity between them (37). In addition, β_2-adrenoreceptors have a higher affinity of binding for epinephrine, which has a higher plasma concentration due to a relatively greater release (than NE) from the adrenal medulla into the bloodstream (37). In addition to NE, neuropeptide Y and adenosine triphosphate (ATP) can also be released as sympathetic co-transmitters, both of which result in vasoconstriction through activation of Y_1 or purinergic P_2X receptors, respectively (35).

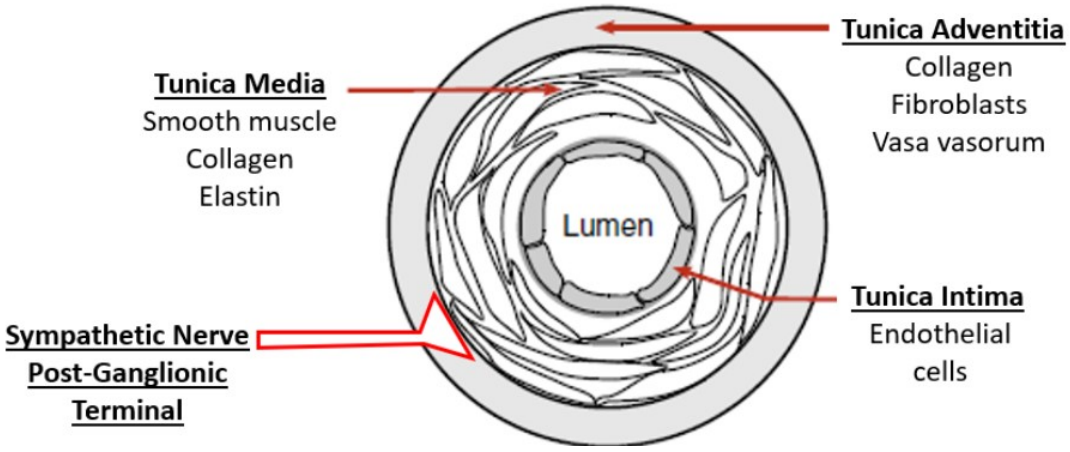

Figure 2.2. Cross-sectional view depicting the 3 main layers of a peripheral artery/arteriole. The inner-most layer (*tunica intima*) is comprised of a single cell layer called the endothelium. The vascular smooth muscle cells predominantly comprise the *tunica media*. The outermost layer is the *tunica adventitia (or externa)*. Post-ganglionic sympathetic nerves terminate at the adventitial-medial border and innervate vascular smooth muscle cells. Image adapted from Klabunde (37).

2.2.2 Sympathetic Control of Total Peripheral Resistance

Total peripheral resistance refers to the magnitude of force affecting the resistance to blood flow imposed by the systemic circulation (i.e., not influenced by the pulmonary vasculature) (37). Factors that contribute to vascular resistance include: 1) the length of the vessel, 2) the viscosity of the blood, and 3) the radius of the blood vessel lumen. However, changes in the lumen radius of resistance vessels (i.e., arterioles) have the largest overall impact on vascular resistance. This is described by the Hagen-Poiseuille equation:

$Resistance\ to\ blood\ flow\ = \left(\frac{\eta \times L}{r^4}\right)$, where η is the blood viscosity, L is the length of the vessel and r is the radius of the vessel lumen (37). While blood viscosity and length influence the resistance to blood flow, changes in the vessel lumen radius provide the most profound impact as represented by this factor raised to the fourth power. Thus, very small changes in blood vessel lumen diameter can result in large changes in vascular resistance. Relative to the other components contained within the various layers (i.e., elastin, fibrous, collagen endothelial cells), arteriole walls contain a relatively larger proportion of vascular smooth muscles. This allows for robust decreases in lumen diameter and strong vasoconstrictive properties (38).

Vascular tone is defined as the degree of vasoconstriction experienced by blood vessels relative to their maximally dilated state (37). A balance between vasodilative and vasoconstrictive substances ensures that blood pressure is maintained while maintaining proper profusion of all organs. Resting sympathetic outflow provides a baseline level of vasoconstriction to resistance vessels (Figure 2.3). Increases in sympathetic outflow can cause additional vasoconstriction, which increases resistance further through greater decreases in lumen diameter. In contrast, reductions in sympathetic outflow lower vascular tone and the resistance to blood flow via increases in lumen diameter.

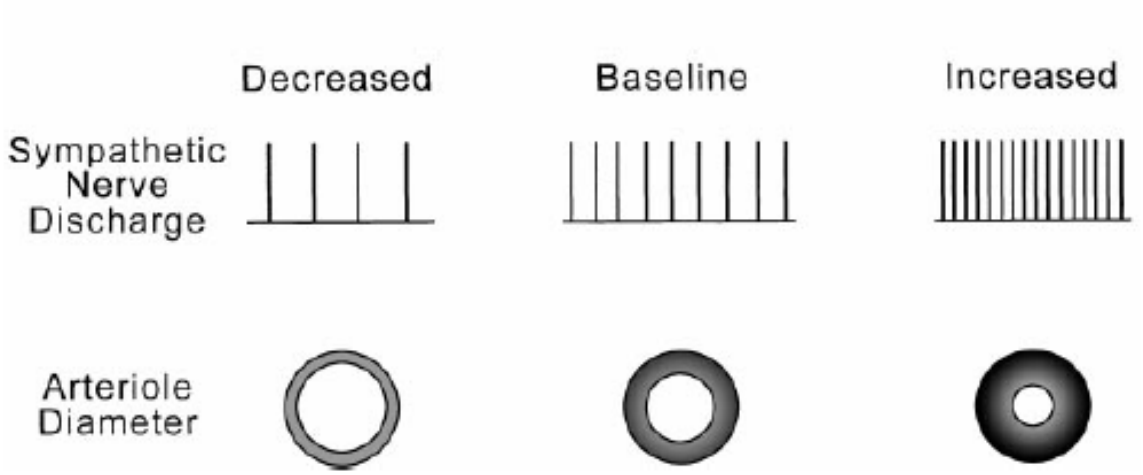

Figure 2.3. The impact of sympathetic nerve discharge frequency on corresponding changes in artery/arteriole lumen diameter from Heesch (35)**. In the center, an example of resting/baseline vascular tone via spontaneous activation of the sympathetic nervous system. Baseline sympathetic activity towards the vasculature provides a background level of vasoconstriction. When sympathetic activity is increased, as shown on the right, greater vascular smooth muscle cell contraction occurs, resulting in more vasoconstriction. On the left, when sympathetic activity is reduced there is less vasoconstriction relative to baseline or resting levels.**

As indicated above in Figure 2.2, arteries and arterioles are composed of three layers: the *tunica adventitia*, the *tunica media*, and the *tunica intima (or externa)* (39). Vascular smooth muscle cells predominantly comprise the *tunica media*, which are the contractile component of blood vessels (39). The contractile proteins within vascular smooth muscle cells are actin and myosin (39). Due to the diagonal orientation of the actin and myosin contractile proteins, vascular smooth muscle contraction pulls the cell together, which encroaches into the lumen to decrease the radius/diameter (39). In contrast, vascular smooth muscle cells flatten out when relaxed, which results in the lumen getting larger (Figure 2.4).

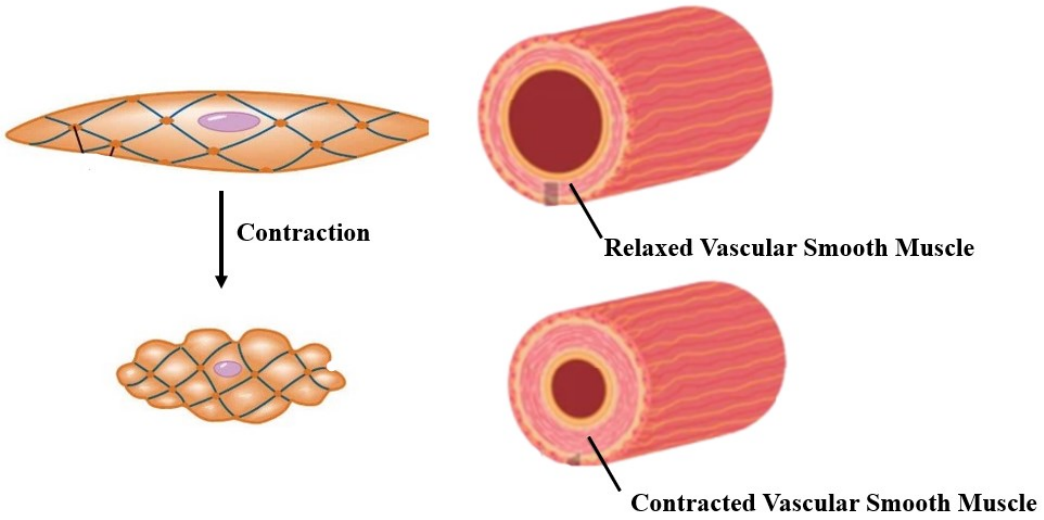

Figure 2.4. Vascular smooth muscle contraction and relaxation. A relaxed smooth muscle cell is shown on the top left. The flattened cell results in a greater blood vessel lumen diameter and vasodilation. In contrast, during vascular smooth muscle cell contraction, the cell "fattens up" and gets larger, which encroaches the lumen and results in vasoconstriction. A cross section of an artery is shown on the right. This displays how the lumen gets larger and smaller with vasodilation and vasoconstriction, respectively.

Contractile activity of vascular smooth muscle cells is initiated by an influx of extracellular calcium, as well as the release of stored intracellular calcium from the sarcoplasmic reticulum (Figure 2.5) (37). Calcium binds to calmodulin to form a calcium-calmodulin complex (37). This complex activates myosin light chain kinase (MLCK), which phosphorylates the myosin light chain (MLC) in the presence of ATP. Phosphorylation of MLC allows the myosin head to bind to actin and form a cross bridge.

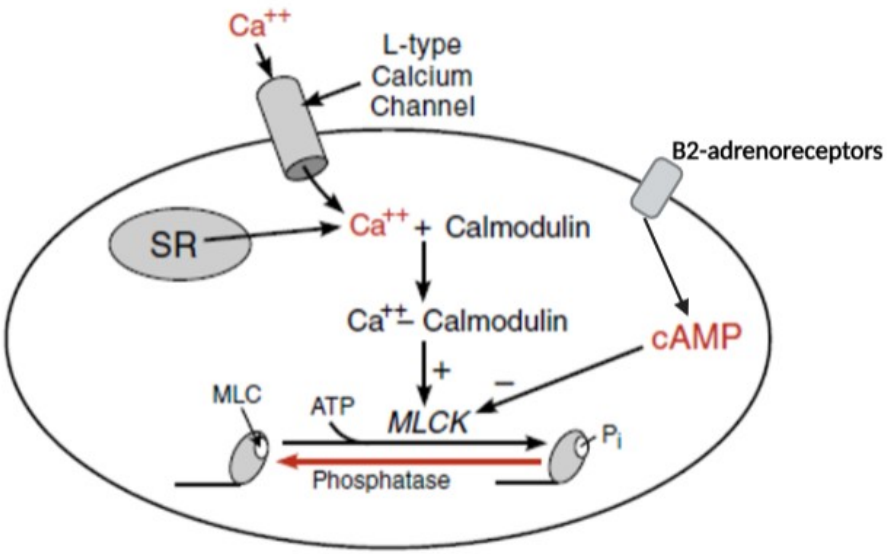

Figure 2.5. Contraction of the vascular smooth muscle cell via activation of myosin light chain kinase (MLCK). Calcium (Ca^{++}) concentrations increase within vascular smooth muscle cells (depolarization) via the entry of extracellular calcium and/or the release from intracellular stores within the sarcoplasmic reticulum (SR). Calcium binds with calmodulin forming a calcium-calmodulin complex. This activates myosin light chain kinase (MLCK). MLCK then phosphorylates myosin light chains (MLC) in the presence of ATP. This allows for cross-bridge formation between myosin and actin and contraction occurs. In the absence of calcium this sequence of events does not happen, allowing relaxation to occur. Cyclic adenosine monophosphate (cAMP) inhibits MLCK, which contributes to relaxation of the cell. Intracellular cAMP increases from binding of epinephrine or NE to β_2-adrenoreceptors.

Post-ganglionic sympathetic nerves innervate the vasculature at the adventitial-medial border and release NE (37). Release of NE at the neurovascular junction activates α_1- or α_2-adrenoreceptors (Figure 2.6) (37). Alpha$_1$-adrenoreceptors are G_q-protein coupled receptors, which activate Phospholipase C. Phospholipase C forms inositol triphosphate and diacylglycerol from membrane bound phosphatidylinositol. Inositol triphosphate opens calcium channels on the sarcoplasmic reticulum to release intracellular stored calcium. Diacylglycerol activates protein kinase C, which contributes to smooth muscle contraction via inhibition (reduced activity) of MLC phosphatase (37). Myosin light chain phosphatase removes phosphate from the MLC, which reduces cross bridge formation and relaxes the vascular smooth muscle cells.

Alpha$_2$-adrenoreceptors are G-inhibitory protein coupled receptors (37) that inhibit adenylyl cyclase. Inhibition of adenylyl cyclase decreases intracellular cAMP, which increases smooth muscle contraction by reducing the inhibitory effect of cAMP on MLCK. Of note, β_2-adrenoreceptors are G_s-coupled receptors, which increase adenylyl cyclase activity and intracellular cAMP concentrations (37). Thus, binding of epinephrine or NE to β_2-adrenoreceptors contributes to vasodilation via a cAMP-mediated inhibition of MLCK (37).

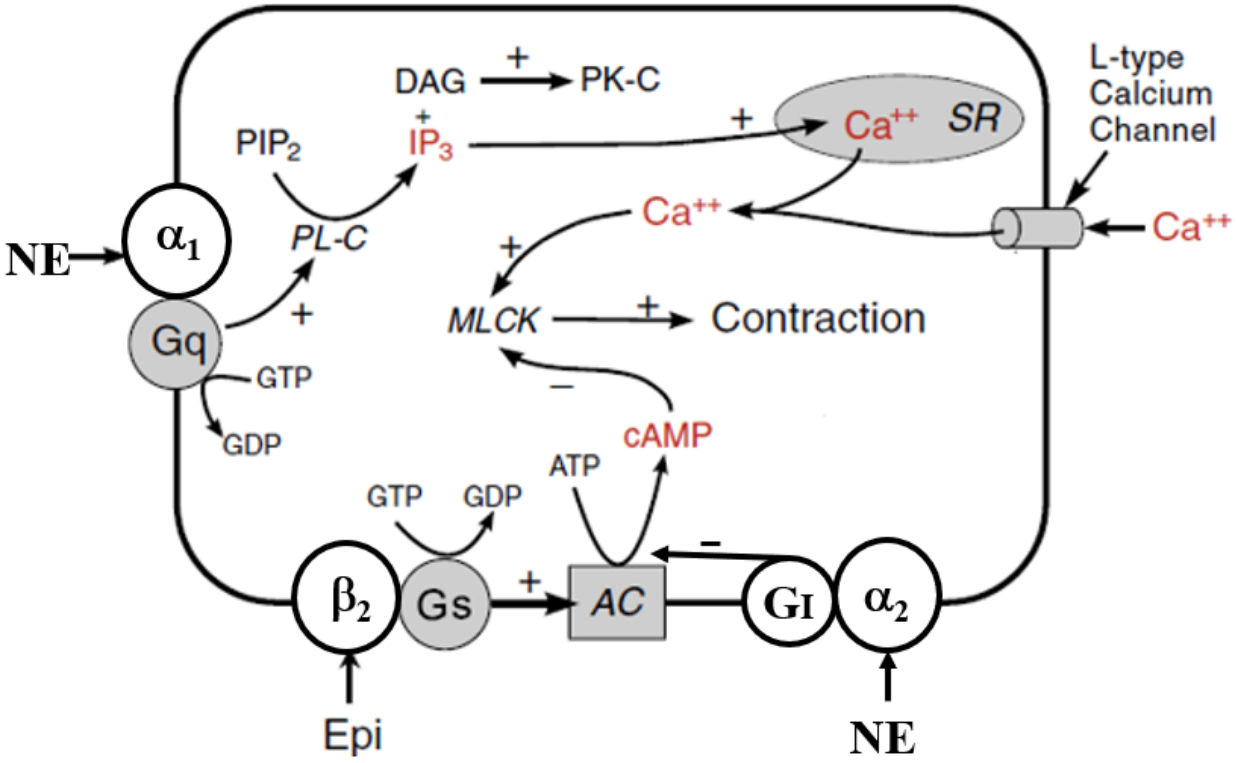

Figure 2.6. Factors influencing vascular smooth muscle contraction and relaxation. *Right side*: Long-lasting calcium channels (L-type) open upon depolarization of the cell and increase intracellular calcium (Ca^{++}). Calcium can also be released from the sarcoplasmic reticulum (SR). *Left side*: Norepinephrine (NE) elicits vasoconstriction via α_1 G_q stimulatory protein-coupled receptors, which activate the phospholipase C (PL-C) pathway. PL-C forms inositol triphosphate (IP_3) and diacylglycerol (DAG) from membrane bound phosphatidylinositol (PIP_2). DAG activates protein kinase C (PK-C). IP_3 increases calcium releases from the SR. Increased intracellular calcium induces smooth muscle contraction via activation of myosin light chain kinase (MLCK) (See Figure 2.5 for more detail). *Bottom*: Epinephrine (Epi) can increase intracellular cyclic adenosine monophosphate (cAMP) through activation of β_2 G_s-coupled stimulatory receptors, which activate adenylyl cyclase (AC). Increased intracellular cAMP opposes vasoconstriction. Norepinephrine can also bind to α_2 G-inhibitory coupled receptors, which inhibits adenylyl cyclase activity and decreases intracellular cAMP (removing inhibition of MLCK activity).

The amount of vasoconstriction elicited by sympathetic neurons depends on the amount of neurotransmitter released, the type and amount of co-transmitters (i.e., ATP, Neuropeptide Y) released, the rate of neurotransmitter removal (via reuptake into post-ganglionic terminals, metabolism, or diffusion into the blood stream), and interactions with other vascular signaling pathways (i.e., other vasoconstricting and vasodilating compounds) (37). The local and systemic hemodynamic effects of sympathetic outflow can be quantified using sympathetic transduction techniques, which provide an index regarding the magnitude of the resultant vasoconstrictor or pressor responses. In humans, the quantification of post-ganglionic sympathetic activity directed towards skeletal muscle vascular beds (i.e., MSNA) can be achieved using microneurographic recording techniques (40).

2.2.3 Measuring Muscle Sympathetic Nerve Activity

First described in the 1960's by Hagbarth and Vallbo (41), microneurography is a technique that allows for direct recordings of efferent post-ganglionic nerve activity in superficial peripheral nerves such as the common peroneal, median and radial nerves. Nerve signals are recorded using a tungsten microelectrode (i.e., active electrode) placed percutaneously and manipulated into a nerve fascicle (40). Prior to manipulation of the active microelectrode, a reference electrode is placed under the skin 2-3cm from the desired recording site (Figure 2.7) (40). The reference electrode is uninsulated, while the active electrode has an epoxy coating, which creates electrical impedance and permits the recording of action potentials from a small surface area at the electrode tip. Active electrodes with an impedance of ~2MΩ are used for multi-unit nerve recordings, as performed in the current project (40).

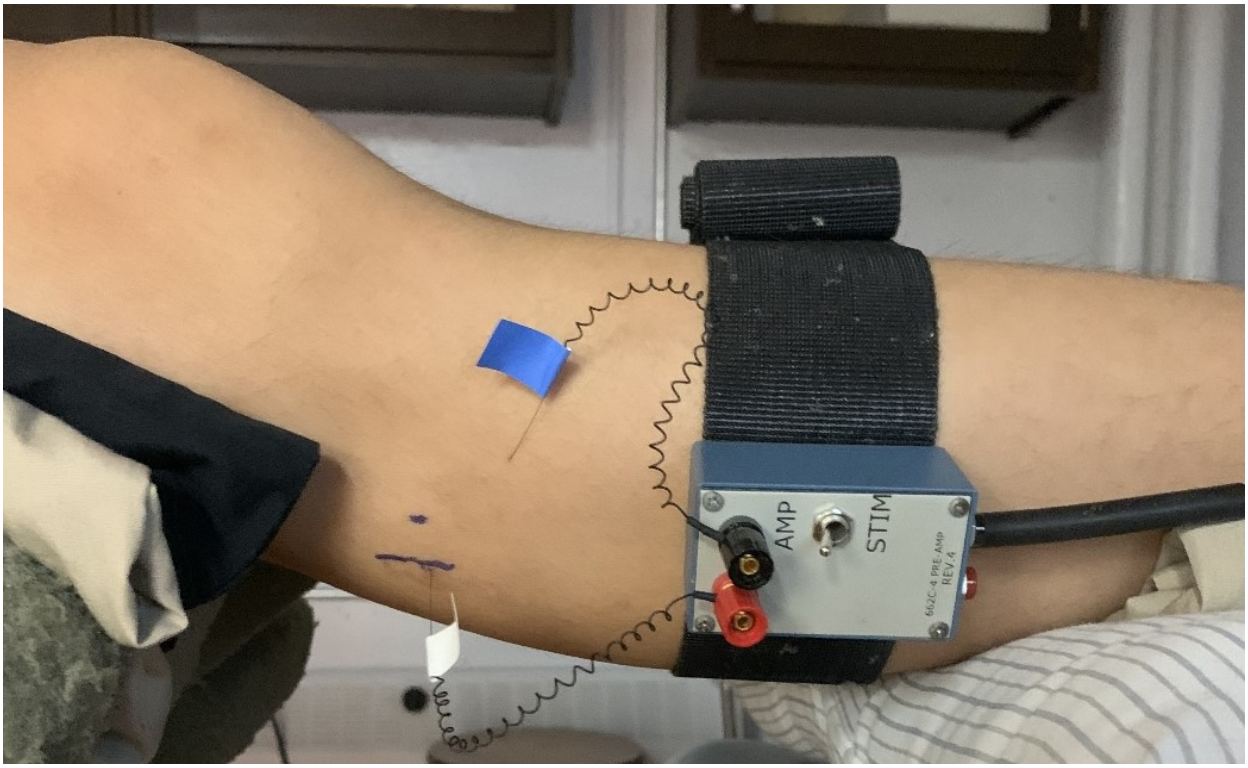

Figure 2.7. Microneurography setup for common peroneal nerve recordings. The differential pre-amplifier is shown attached to the reference (blue flag) and the active (white flag) microelectrodes. The reference microelectrode is uninsulated, while the active microelectrode has an epoxy coating that provides an impedance of ~2mΩ. Above the white electrode, pen marks are visible. These provide visual reference for the fibular head (top mark) and below where peroneal muscle contractions were elicited using mild surface electrical stimulation of the nerve. The latter test is used to help with accurate localization of the active microelectrode.

The reference and active microelectrodes are attached to a differential pre-amplifier (1000×). This differential configuration serves to help minimize noise by subtracting the signal recorded from the reference electrode from the active microelectrode. The output of the differential amplification is connected in series to a variable-gain amplifier, which further amplifies the signal (~75×) for a total signal gain of ~75,000 times. The raw, amplified signal is band-pass filtered between 500-2000 Hz to minimize noise from sources with lower frequency content than neural impulses (e.g., movement artifacts, equipment noise) (40). Subsequently, the signal is full wave rectified and integrated at a time constant of 0.1-seconds to produce a mean voltage neurogram (Figure 2.8).

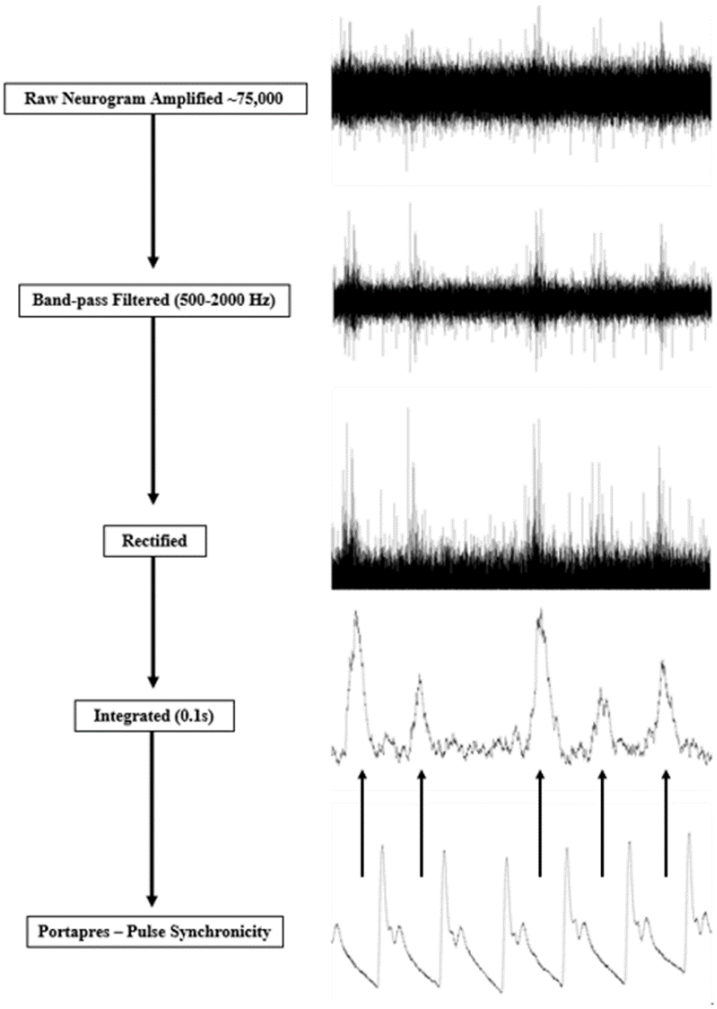

Figure 2.8. Processing steps required to generate a mean voltage neurogram recording using microneurography. First, the raw signal (sampled at 20,000Hz) is amplified with a ~75,000× gain. The amplified signal is band-pass filtered (500-2000 Hz), full wave rectified, then integrated or smoothed (0.1s time constant). The bottom panel of the figure depicts the pulse synchronicity of the final integrated muscle sympathetic nerve activity (MSNA) signal. Each MSNA burst has been time shifted backwards to be aligned with the cardiac cycle where it was generated. This image also shows the morphology of MSNA bursts, which have relatively steep upward and downwards slopes immediately surrounding the peak.

To determine a successful MSNA recording, MSNA signals are inspected for cardiac synchronicity, increased activity in response to a breath hold (i.e., chemoreflex stimuli), and/or Valsalva's maneuver (i.e., baroreflex stimuli) (40). Furthermore, bursts of MSNA have a distinct shape (Figure 2.8). The initiation of an MSNA burst is linked to diastole as baroreceptor afferent innervation to the NTS is lowest during this phase of the cardiac cycle. This lessens the inhibitory effects of the CVLM on the RVLM, which increases sympathetic outflow. The

subsequent systolic pulse increases baroreceptor afferent activity, resulting in a greater stimulation of the NTS-CVLM axis and subsequent inhibition of the RVLM. This corresponds to a decrease in sympathetic outflow and the downward slope of an MSNA burst (Figure 2.8).

After a successful recording (i.e., all the above criteria are met), bursts of MSNA are identified using a 3:1 burst-to-inter-burst noise ratio (i.e., to be considered a burst of MSNA, burst amplitudes must be 3 times larger than the noise of the between burst peaks). Muscle sympathetic nerve activity is traditionally quantified via time-averaged metrics of burst frequency (bursts/minute) and burst incidence (bursts/100 heart beats). Burst frequency provides an index of the amount of sympathetic activity that the vascular smooth muscle cells are exposed to (40). Burst incidence differs in that it represents the percentage of heart beats associated with a burst of MSNA (40). Specifically, a higher burst incidence (i.e., more bursts per 100 heart beats) indicates that more cardiac cycles were associated with a burst and vice versa. The size or strength of MSNA bursts can also be quantified by calculating the mean absolute amplitude (mV), which can be multiplied with burst frequency to derive an index of total MSNA. Additionally, each MSNA burst height can be normalized as a percentage of the largest burst amplitude identified from the recording session. This becomes especially important when comparing MSNA between time points that involve separate recoding sessions (e.g., pre- versus post-intervention) since the population of neurons will differ between recordings, which will alter the absolute burst amplitudes. Some groups may choose to use MSNA burst area instead of MSNA burst height in their analyses of burst strength/size. Burst height and burst area are highly correlated during normal sinus rhythm (42, 43). Thus, this project uses burst height as the measure of burst size, as our analysis software can more accurately calculate this metric.

Traditional measurements of MSNA burst frequency and burst incidence provide relatively static information about sympathetic vasoconstrictor outflow. However, more dynamic information can be obtained about the influence of individual bursts of MSNA on the vasculature by tracking corresponding beat-by-beat changes in DBP following each MSNA burst. This signal-averaging approach is commonly referred to as sympathetic transduction (15).

2.2.4 Measuring Sympathetic Transduction at Rest

In 1982, Wallin and Nerhed (15) first reported the signal-averaging method to assess sympathetic transduction, which tracked changes in mean arterial pressure (MAP) in response to individual bursts of MSNA (Figure 2.9). In their method, each burst of MSNA was identified and tracked for 15 cardiac cycles. The change in MAP for each cardiac cycle following a burst was calculated and the mean peak response determined as the measure of sympathetic transduction. Wallin and Nerhed (15) determined that among their participants, MAP started to rise 1 to 2 seconds following a burst with the peak change occurring ~5-7 cardiac cycles after each burst.

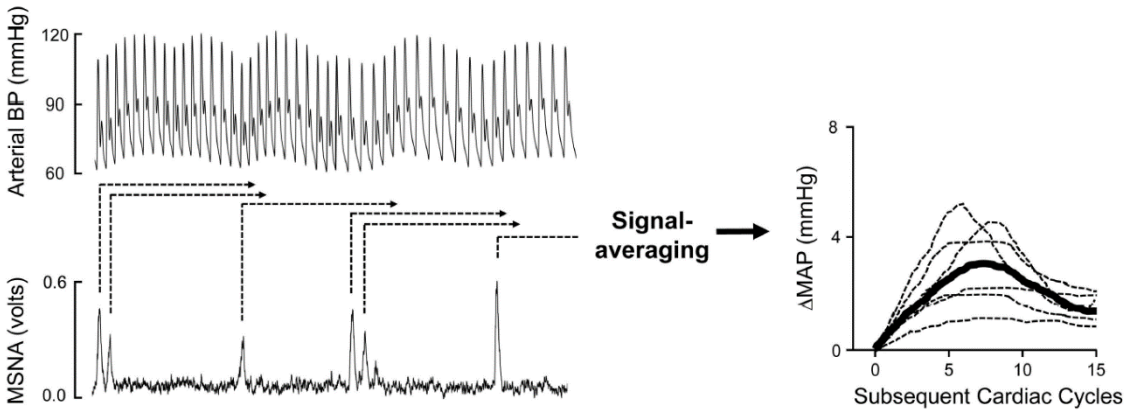

Figure 2.9. Schematic from Young et al. (17) **demonstrating the signal-averaging approach reported by Wallin and Nerhed**(15) **for determining sympathetic transduction. On the left, each burst of muscle sympathetic nerve activity (MSNA) was identified (dotted lines) and mean arterial pressure (MAP) tracked for the next 15 cardiac cycles. On the right, each dotted line tracing represents the increases in MAP following individual bursts. The thick black line shows the mean of the individual burst responses. From the mean peak change in MAP (ΔMAP), a sympathetic transduction value was determined.**

This signal-averaging method has since been adapted by others (8–14) to further investigate sympathetic transduction-focused research questions. With improvements in ultrasound technology, simultaneously recorded arterial blood flow and regional vascular responses (e.g., leg vascular conductance/resistance) have been incorporated as additional peripheral or local sympathetic transduction response variables. Furthermore, this method has been adapted to investigate pressor responses elicited by different MSNA burst patterns (i.e., isolated single bursts versus consecutive multiple burst 'trains'), as well as nadir blood pressure reductions following heartbeats not associated with bursts of MSNA (9, 10). This is of importance as multiple bursts in a row provoke greater responses compared to single, isolated bursts (8, 10). As well, vascular, or systemic responses following cardiac cycles without an associated MSNA burst (i.e., non-burst cardiac cycles) may provide information regarding the control of blood pressure in the absence of sympathetic activity (11).

Briant and colleagues (16) have also developed an analysis technique based on weighted linear regressions between 1% bins of normalized MSNA burst area (or height) (i.e., the area or height of each MSNA burst is determined and normalized as a percentage of the largest burst) versus DBP. Specifically, in their method, each DBP is associated with the burst size within a fixed 2 cardiac cycle window 6-8 cardiac cycles prior (Figure 2.10). If more than 1 burst occurred within the 2 cardiac cycle window, those burst areas or heights were summed. The slope of the relationship between each DBP and the associated normalized MSNA burst area or height provided an index of sympathetic transduction (Figure 2.10).

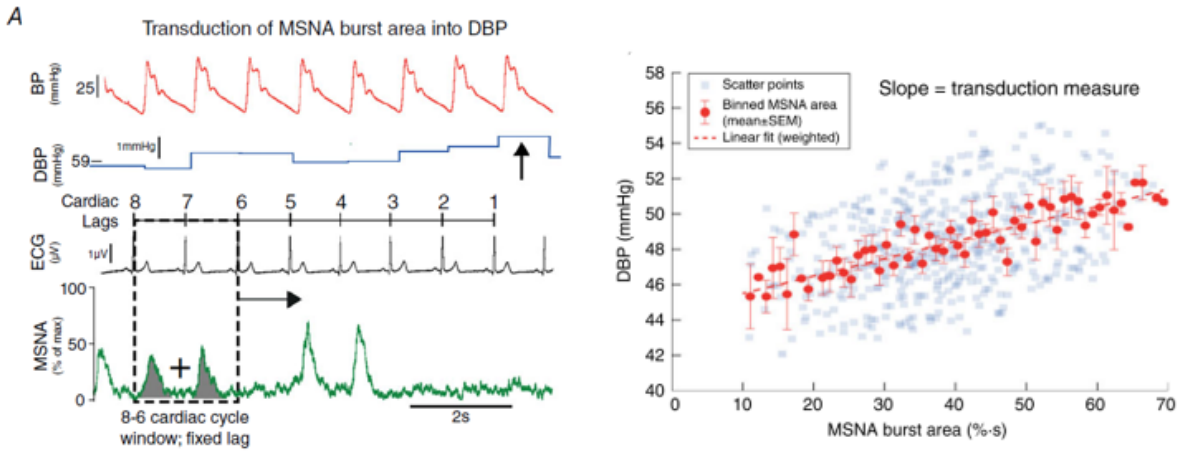

Figure 2.10. Example of the weighted linear regression method used to calculate sympathetic transduction from Briant et al. (16). In this method, each diastolic blood pressure was tracked on a beat-to-beat basis. An example is shown in the second row in blue indicated by the arrow. Each diastolic blood pressure was associated with the burst size within a fixed 2 cardiac cycle window 6-8 cardiac cycles prior (shown by the cardiac lags). The burst area (or burst height) within these two cardiac cycle windows was summed and associated with that diastolic blood pressure. This was repeated for each diastolic blood pressure value over the recording period. Burst areas were then calculated as percentages of the largest burst area for the participant and plotted against diastolic blood pressure in 1% bins. A plot was then generated as shown on the right. The slope of this line was quantified as the sympathetic transduction value for each participant.

For the current study, sympathetic transduction will be quantified using the signal-averaging method developed by Wallin and Nerhed (15). This method was chosen as it allows for more inter-individual variability in the analysis. Specifically, the Briant et al. (16) analysis technique uses a fixed lag of 6-8 cardiac cycles for each DBP value. This does not account for inter-individual differences in the number of cardiac cycles to peak transduction of a burst of MSNA. Finally, the signal-averaging method has been more widely used (8–15), which permits greater comparability to these previous reports.

2.3 **The Impact of Aerobic Fitness on Sympathetic Transduction**

Aerobic fitness is an important contributor to overall cardiovascular health (4, 6, 44). As such, it is important to consider when investigating factors related to sympathetic control of blood pressure. Cross-sectional research has demonstrated that in younger males (Figure 2.11)

(13), and an older mixed group of males and females (18), those with higher aerobic fitness exhibited lower sympathetic transduction. Similarly, Notarius et al. (45) used a sympatho-excitatory stimulus (graded lower body negative pressure protocol) to induce increases in peroneal MSNA and forearm vascular resistance. The relationship between MSNA burst frequency and forearm vascular resistance was compared between more (42.7 ± 2.8 ml/kg/min) and less aerobically fit middle-aged males (25.8 ± 1.2 ml/kg/min). In their less fit middle-aged males, there was a positive relationship between forearm vascular resistance and MSNA burst frequency (Figure 2.12) (45). In contrast, there was no relationship observed (i.e., reduced sympathetic transduction) in the more fit middle-aged males (45). These authors suggested this may have been a protective mechanism wherein bursts of MSNA did not consistently elicit vasoconstriction (measured via increases in forearm vascular resistance). This theory was supported by a study conducted in male Fisher rats that investigated the influence of exercise training on α-adrenergic-mediated vasoconstriction (20). After 12-weeks of treadmill running (1 hour/day, 5 days/week), α-adrenergic-mediated vasoconstriction to NE was attenuated in both old and young rat populations. Although no direct measures of nerve activity were made, it provided evidence that exercise training attenuated the sympathetic regulation of vascular resistance and blood pressure. Alterations in pre-synaptic, inhibitory α_2-adrenergic receptors, luminal facing β_2-adrenergic (vasodilatory) receptors, competing endothelial-derived vasodilators, and the pathways associated with these receptors and vasoactive chemicals may also influence the sympathetic regulation of vascular resistance and blood pressure following exercise training. However, these have yet to be fully explored in relationship to sympathetic transduction.

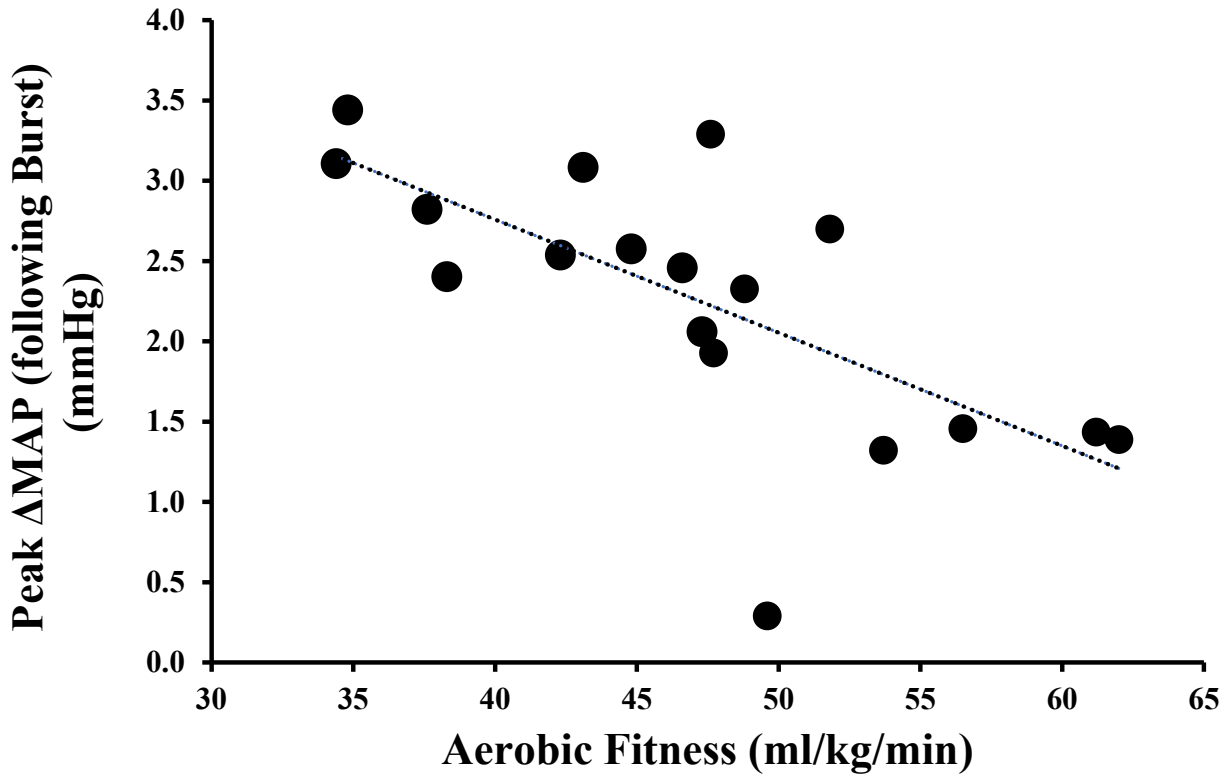

Figure 2.11. Relationship between peak increases in mean arterial pressure (ΔMAP) and relative aerobic fitness ($\dot{V}O_2$: relative peak volume of oxygen consumption) reported in younger males by O'Brien et al. (13). This figure demonstrates that younger males with higher aerobic fitness exhibited smaller increases in MAP following bursts of muscle sympathetic nerve activity.

In contrast, a recent pilot study investigated differences in sympathetic transduction between young female habitual exercisers (n=5, 44±3 ml/kg/min $\dot{V}O_2$peak) and non-habitual exercisers (n=6, 30±8 ml/kg/min $\dot{V}O_2$peak) (19). This study reported no differences in peak MAP responses following spontaneous bursts of MSNA between their exerciser (1.87±0.62 mmHg) and non-exerciser groups (1.53±0.66 mmHg). More research is needed in this population to determine whether these disparate between-study outcomes (13, 19) are due to sex differences or the small sample reported in the pilot study.

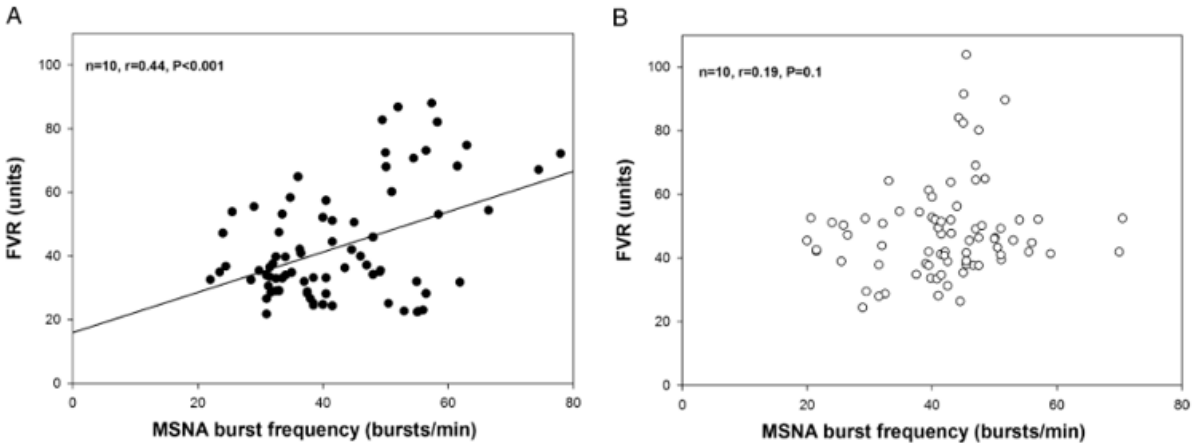

Figure 2.12. Data from Notarius et al. (45) **demonstrating the relationship between forearm vascular resistance (FVR) and peroneal muscle sympathetic nerve activity (MSNA) burst frequency in less fit (left) versus more fit (right) middle-aged males. A graded lower body negative pressure protocol elicited increases in MSNA. The lack of relationship on the right in the more fit males was suggested to be a protective effect wherein bursts of MSNA did not consistently elicit vasoconstriction. The relationship in the less fit group suggested that in middle-aged males with lower aerobic fitness, bursts of MSNA were more closely related to increases in vascular resistance.**

Most of the cross-sectional research that has been conducted suggests that, at least in males, those with higher aerobic fitness exhibit lower sympathetic transduction compared to those with lower aerobic fitness. However, it is unknown whether decreases in sympathetic transduction are achieved when healthy adults improve their aerobic fitness via exercise training. If decreases in sympathetic transduction are observed following improvements in aerobic fitness, this would provide support that improving aerobic fitness can modulate the sympathetic control of blood pressure. This may be an important cardiovascular benefit of improving aerobic fitness.

2.4 **High-Intensity Interval Training**

High-intensity interval training is a tolerable mode of exercise that permits individuals to work at greater percentages of their maximum aerobic capacity when compared to moderate-intensity continuous aerobic activities (21). This is of importance when the primary aim of

exercise training is to elicit improvements in aerobic fitness, particularly in healthy populations. Previous research has demonstrated that in young healthy individuals who are already relatively fit, a greater training stimulus is required to observe improvements in $\dot{V}O_2$peak in relatively fit younger adults (24). This is most effectively achieved via high-intensity training (24).

High-intensity interval training will be used in the current study to elicit improvements in aerobic fitness. The specific HIIT protocol has been used previously by our lab (22) and others (23). While the focus of the current study is on younger adults, future research will focus on the impact of HIIT on sympathetic transduction in older adults. As such, HIIT was selected over moderate-intensity continuous training as previous research from our lab (22) has demonstrated greater improvements in vascular endothelial-dependent function in older adults following HIIT compared to isoenergetic moderate-intensity continuous aerobic training and whole-body resistance training. This suggests that in addition to being important for improving aerobic fitness, HIIT may provide additional vascular adaptations. The HIIT protocol chosen for this study involves intervals of 15-s of cycling at 100+% of peak aerobic power (PAP) interspersed with 15-s of passive recovery. The number of intervals and the percentage of PAP progressively increases throughout the training period to maintain a high training stimulus due to anticipated improvements in fitness over the course of the program. This protocol was chosen as it was previously compared to four other HIIT protocols in coronary heart disease patients and was determined to be the most tolerable, elicited the largest quantity of time spent cycling above 80% of $\dot{V}O_2$peak, and the longest time to exhaustion (46). Like pervious studies, this protocol will be completed on an electromagnetically braked cycle ergometer. This modality was determined to be safer and more feasible than conducting intervals on a treadmill. In addition, the cycle

ergometer provides a simple and accurate way to control the desired training intensity and allows participants to cycle at a cadence that is comfortable for them.

Previously, 6-weeks of this HIIT protocol was determined to be sufficient to elicit improvements in aerobic fitness in older adults (23 ± 7 versus 28 ± 7 ml/kg/min) (22). However, when this 6-week protocol was used in younger adults, no improvements in relative $\dot{V}O_2$peak were observed (40 ± 9 versus 41 ± 8 ml/kg/min) (23). However, this cohort did improve their PAP (207 ± 45 versus 317 ± 42 W), which suggests that training adaptations were apparent (23). Other studies using different HIIT protocols have demonstrated improvements in aerobic fitness after 12-weeks of training (21). Based on the improvements in PAP observed by Mekari et al. (23) (indicative of training adaptations), and other studies observing training-induced improvements in aerobic fitness after 12-weeks (21), 12-weeks of HIIT will be used in the current study to determine whether younger adults can improve their aerobic fitness using the aforementioned HIIT protocol.

2.5 **Purpose and Hypotheses**

The primary purpose of this study was to investigate whether 12-weeks of HIIT would: 1) improve aerobic fitness, and 2) evoke corresponding decreases in spontaneous sympathetic transduction in healthy adults. Based on previous HIIT studies that observed improved aerobic fitness after 12-weeks (21), it was hypothesized that the HIIT protocol used in this study would increase aerobic fitness. Based on the inverse relationship between sympathetic transduction and aerobic fitness that our group has observed in younger males (13) and older males and females (18), it was hypothesized that HIIT would decrease sympathetic transduction when aerobic fitness was improved. However, as younger females may exhibit no impact of aerobic fitness on sympathetic transduction (19), this study performed a preliminary, exploratory assessment into

whether sex impacted these responses to HIIT. The results of this study may provide further insight into the potential cardioprotective effects of HIIT.

Chapter 3: METHODS

3.1 **Participants**

Seventeen healthy adults (10 females, age 20-46) with no physical limitations to exercise (as determined by the Canadian Society for Exercise Physiology Get Active Questionnaire, Appendix A) were recruited to participate in the 12-week HIIT intervention or control condition. Smoking (47), obesity (48), and hypertension (27–30) are all known to increase MSNA and thus may influence the primary outcomes. As such, all participants were non-smokers, had a resting seated blood pressure <135mmHg/<85 mmHg (49), and a body mass index <30 kg/m^2 (i.e., obese classification). Furthermore, certain cardiovascular medications (e.g., non-selective or $beta_2$-blockers, calcium channel blockers, renin-angiotensin acting agents, lipid-lowering drugs) may impact sympathetic neurovascular control. Therefore, participants prescribed these medications were also excluded. Smoking status and medication history were determined by self report from a health history questionnaire (Appendix B).

Participants were informed of the methods and procedures verbally and in writing before providing written informed consent. All protocols and procedures conformed to the Declaration of Helsinki and were approved by the Dalhousie Health Sciences Research Ethics Board (REB# 2021-5555; Appendix C).

3.2 **Experimental Procedures and Analyses**

3.2.1 Anthropometrics and Aerobic Fitness

Height and body mass were measured using a calibrated stadiometer (Health-O-Meter, McCook IL, USA) to the nearest 0.5 cm and 0.1 kg, respectively. Participants completed an incremental, maximal exercise test on an electromagnetically braked cycle ergometer (Lode Excalibur Sport, Groningen, The Netherlands) to determine $\dot{V}O_2peak$ via a commercial, mixing chamber-based metabolic system (TrueOne 2400, Parvo Medics Inc., Sandy, UT, USA).

Participants were equipped with a chest strap heart rate monitor (Polar H9, Kempele, FI) and chose either a full-face mask or mouthpiece and nose clip combination. The ergometer seat was adjusted to a comfortable position for the participant while ensuring that a slight knee bend of 10-20 degrees was present at the lowest pedal position. Following a 5-minute warm-up period of light-intensity cycling (50 W), the workload was set at 70 watts and increased by 20 watts/min until voluntary exhaustion. Strong verbal encouragement was provided throughout the test. Upon completion, the workload was immediately reduced to the warm-up level for a 5-minute cool-down period. Ratings of perceived exertion were determined approximately every 2-minutes using the Borg 6-20 scale (50).

For the test to be considered a maximal effort (i.e., $\dot{V}O_2max$), participants had to achieve a plateau in oxygen consumption (<1.5ml/kg/min) despite an increase in workload. In summary, 10/14 participants at baseline, 5/9 at mid-training, and 9/14 participants at follow-up achieved a plateau in oxygen consumption. Since not all participants achieved a plateau in $\dot{V}O_2$, $\dot{V}O_2peak$ will be used to quantify aerobic fitness. For those that did not achieve a $\dot{V}O_2max$, a $\dot{V}O_2peak$ was qualified if ≥2 of the following criteria were achieved: 1) maximal heart rate ≥ 90% of age-predicted maximal heart rate (i.e., 208 - 0.7 × age) (all participants achieved at baseline, mid, and follow-up) (51), 2) a respiratory exchange ratio ($\dot{V}CO_2/\dot{V}O_2$) ≥ 1.10 (all participants at baseline, mid, and follow-up), or 3) a maximal Borg perceived exertion rating >17 (12/14 at baseline and all participants at mid and follow-up). In conclusion, every participant met ≥2 of these criteria at all time points.

Heart rate (HR), absolute $\dot{V}O_2$ (L/min), relative $\dot{V}O_2$ (ml/kg/min), respiratory exchange ratio, and minute ventilation were averaged every 15-seconds during the graded exercise test. Ventilatory threshold was calculated via the Parvo Medics software as a percentage of maximum

$\dot{V}O_2$ peak. The heart rate at the ventilatory threshold was also determined. This was automatically calculated by the software and confirmed for accuracy by looking at the relationship between minute ventilation (y-axis) versus $\dot{V}O_2$ and determining the inflection point where minute ventilation started to increase at a greater rate than $\dot{V}O_2$. $\dot{V}O_2$ peak, and peak respiratory exchange ratio were determined from the greatest consecutive 30-second average. Peak metabolic equivalents of task (METs) were determined from the Parvo Medics output by dividing relative $\dot{V}O_2$ peak by a standardized resting relative $\dot{V}O_2$ value of 3.5 ml/kg/min. Resting METS are ~3.5ml/kg/min in young adults and thus, this standardized value was used (52). Using the American College of Sports Medicine sex- and age-based normative relative $\dot{V}O_2$ data, the normative aerobic fitness percentile based on participants relative $\dot{V}O_2$ peak was determined (53). The final workload achieved during the maximal exercise test was considered the peak aerobic power (PAP). For participants in the intervention group, this PAP was used to determine cycling intensity for the subsequent HIIT protocol.

3.2.2 Activity Monitoring

All participants wore an activPAL triaxial inclinometer (Pal Technologies Ltd., Glasgow, UK) 24-h per day for 7-d at baseline and follow-up. The activPAL is a valid measure of time spent stepping, standing, and sitting/lying down (54). The activPAL was waterproofed in a nitrile finger cot and secured using transparent medical dressing (Tegaderm™, 3 M, London, ON, Canada) to the midline of their right thigh, one-third of the way between the hip and knee. Participants self-reported their waking hours (for sedentary time determination) to accommodate activPAL analysis. This monitoring was conducted to provide insight into participants' habitual physical activity and sedentary time patterns to address whether these physical behaviors (e.g.,

time spent sedentary and engaged in stride-based light, moderate, or vigorous activity) changed over the course of the intervention (or control period).

The activPAL data were analyzed using a customized LabVIEW program (LabVIEW 2020, National Instruments, Austin, TX, USA) that confirmed waking hours and summarized daily averages of time spent standing, stepping, and sitting/lying down. Time spent in different stride-based physical activity intensities (light, moderate, vigorous) were determined using height-adjusted step rate thresholds (55).

3.2.3 Resting Systemic Hemodynamics

During microneurography sessions, HR was calculated from the time between successive cardiac intervals (i.e., R-R intervals) obtained from lead II of a standard bipolar electrocardiogram configuration. Beat-by-beat systolic (SBP) and diastolic blood pressure (DBP) were measured using finger photoplethysmography (Portapres; Finapres Medical Systems, Amsterdam, The Netherlands). The finger used for recording was maintained at heart level to prevent hydrostatic pressure differences between the heart and finger and ensure a stable blood pressure recording. Three upper arm SBP and DBP measurements were recorded using an automated vital signs monitor (Carescape v100, General Electric Healthcare, Mississauga, ON, Canada). The mean of these values was used to perform a 'physiological correction' of the Portapres waveform in LabChart (Version 1.1, ADInstruments, Sydney, Australia) to better represent 'true' brachial arterial pressure. The electrocardiogram (1000 Hz) and Portapres (200 Hz) recordings were sampled continuously using a PowerLab (PL3508 PowerLab 8/53, ADInstruments, Sydney, Australia) data acquisition system and analyzed offline using LabChart software (Version 8, ADInstruments, Sydney, Australia).

Both SBP and DBP were determined from the Portapres waveform as the maximum and minimum within beat values, respectively. MAP was calculated as ⅓ SBP + ⅔ DBP. Baseline beat-by-beat hemodynamic data were averaged over a 10-minute recording period.

3.2.4 Muscle Sympathetic Nerve Activity (MSNA) Recordings

Multi-unit post-ganglionic MSNA was recorded from the lower leg in the common peroneal nerve using standardized microneurography techniques (56). Simultaneous MSNA recordings are synchronous between the left and right peroneal nerves (57). As such, MSNA was recorded from either leg in this study. In brief, the nerve was first mapped using external isolated stimulation to determine the optimal location for insertion of the active microelectrode. A non-insulated reference electrode was then placed under the skin approximately 2-3 cm from the desired recording location. A 2-mΩ unipolar tungsten microelectrode (FHC, Bowdoin, MA), attached to an isolated differential pre-amplifier was inserted percutaneously into the nerve fascicle. Nerve signals were further amplified (~75,000 times), bandpass filtered (500-2000 Hz), full-wave rectified and integrated (0.1 s time constant) (662C-4 Nerve Traffic Analysis System, University of Iowa Bioengineering, Iowa City, IA). This resulted in a mean neurogram recording from which 'bursts' of integrated MSNA were quantified.

Acceptable MSNA recordings were considered via the presence of cardiac synchronicity, a 3:1 signal-to-inter burst noise ratio, and overall burst morphology. In addition, MSNA responsiveness to an end-expiratory breath hold, and Valsalva's maneuver, as well as lack of responsiveness to skin stimulation or mental arousal were confirmed. Finally, only MSNA recordings absent of shifts in baseline activity were used for analysis. A shift in the baseline is indicative of microelectrode movement and may indicate a different population of neurons

recorded from and/or a lost signal. Once a stable recording was obtained, participants remained in the supine position in a darkened room for at least 10-min of data collection.

Time-averaged MSNA was quantified as burst frequency (bursts/minute) and burst incidence (bursts/100 heart beats). The absolute amplitude (mV) was determined for each MSNA burst and normalized as a percentage of the tallest burst measured during the recording period. Normalization of burst height was especially important for comparing between time points over separate recording sessions (e.g., pre- versus post-intervention) since the population of neurons differs between recordings, which impacts the absolute burst amplitudes (i.e., more single units recorded from produces larger burst amplitudes). Total normalized MSNA activity was calculated as the product of the mean relative burst amplitude and burst frequency (13, 58).

3.2.5 Sympathetic Transduction

A signal-averaging approach to quantify sympathetic transduction was determined using an open-source Microsoft Excel-based program developed by our lab (59). In LabChart (AD Instruments, Sydney, Australia), separate semi-automatic macros were used to place a 'Beat' comment in the middle of each cardiac cycle in the electrocardiogram channel (i.e., between two R-waves) and a 'Burst' comment at the peak of each MSNA burst (Figure 3.1).

Depending on the size of the burst and the height of the individual, the latency between the time each burst was generated in the RVLM and recorded by the active electrode will vary. This latency can be calculated by determining the time between the peak of the R-wave associated with the cardiac cycle from which each burst was generated and the peak of the burst itself. Larger bursts tend to have a shorter latency because they are associated with the recruitment of faster conducting post-ganglionic sympathetic neurons (60). Taller individuals will have a longer latency due to the greater distance that the nerve signal must travel from the

RVLM to the recoding microelectrode (61). Previous research has suggested that on average, bursts are recorded ~1.2-1.5 seconds following the cardiac cycle in which they were generated (61). To time-align beat-by-beat cardiac intervals and integrated MSNA signals for sympathetic transduction analysis, the preceding R-wave associated with a range of smaller and taller 'clean' MSNA bursts (i.e., non-noisy bursts with >3:1 signal-to-inter burst noise ratio) were identified. The average time latency for these bursts was used to shift the MSNA channel in LabChart. The MSNA channel was shifted so that the peak of each 'clean' burst was time aligned with the 'Beat' comment for the cardiac cycle encompassing the R-wave from which the MSNA burst was generated. Because of the slight delay in transmission between the electrocardiogram recording and the Portapres waveform, the Portapres was also shifted backwards so the systolic pulse was in line with the peak of the R-wave. This ensured that the DBP associated with each cardiac cycle was used instead of an average DBP of two cardiac cycles. Step-by-step analysis instructions are included in Appendix D.

Time-aligned, beat-by-beat systemic hemodynamic and MSNA data were extracted from LabChart by extracting data between successive 'Beat' comments and exported to an Excel spreadsheet (Microsoft, Washington, USA) such that each row of data corresponded to one cardiac cycle. The primary hemodynamic outcome variable used to quantify sympathetic transduction was DBP, as it provides the best estimate of the sympathetic vasoconstrictor-mediated response on the peripheral vasculature with minimal influence from cardiac-related factors (e.g., changes in cardiac output). Built-in functions within Excel were used to track absolute beat-by-beat changes in DBP for 12 cardiac cycles following the heartbeat in which the burst originated (i.e., cardiac cycle 0). Other groups have previously tracked bursts for 10 (15, 62), 12 (19, 63), or 15 (8, 10, 12) cardiac cycles after the burst originated. However, all groups

determined an average peak increase in blood pressure within 5-8 cardiac cycles (8, 10, 12, 15, 62). Therefore, 12 cardiac cycles would ensure that the peak pressor response was extracted assuming some variability within or between participants. The absolute changes in DBP across cardiac cycles 1 through 12 for all bursts were then averaged. The average peak increase in DBP was extracted to represent sympathetic transduction. The number of cardiac cycles to the peak DBP response was determined for each participant.

Successive cardiac cycles containing bursts provokes a greater pressor response than bursts that occur in isolation (8, 10). Therefore, bursts that were part of a sequence (i.e., multiple bursts immediately adjacent to one another) versus those separated by at least one cardiac cycle from another burst were identified. Burst sequences were analyzed separately for singlets (one burst without any adjacent bursts), doublets (two adjacent bursts) and triplets + (≥3 adjacent bursts). For each participant, the percentage of total bursts classified into each sequence was calculated as: (# of bursts per sequence ÷ total # of bursts) × 100%.

In addition, the absolute beat-by-beat changes in DBP were tracked for 12 cardiac cycles following all heartbeats not associated with an MSNA burst. The average nadir DBP responses during cardiac cycles absent of bursts of MSNA were also calculated. The number of cardiac cycles to the nadir DBP response were determined for each participant. These non-burst nadir DBP responses provide information regarding cardiovascular regulation during sympathetic quiescence. Furthermore, assessing the non-burst nadir responses serves as an internal control to ensure that peak DBP responses are truly MSNA mediated. A summary figure displaying average group data tracked for 12 cardiac cycles for all transduction outcomes (peak increases, nadir non-burst decreases, and burst sequences) is presented in Supplemental Figure 2 (Appendix E) to visually display the time course of changes.

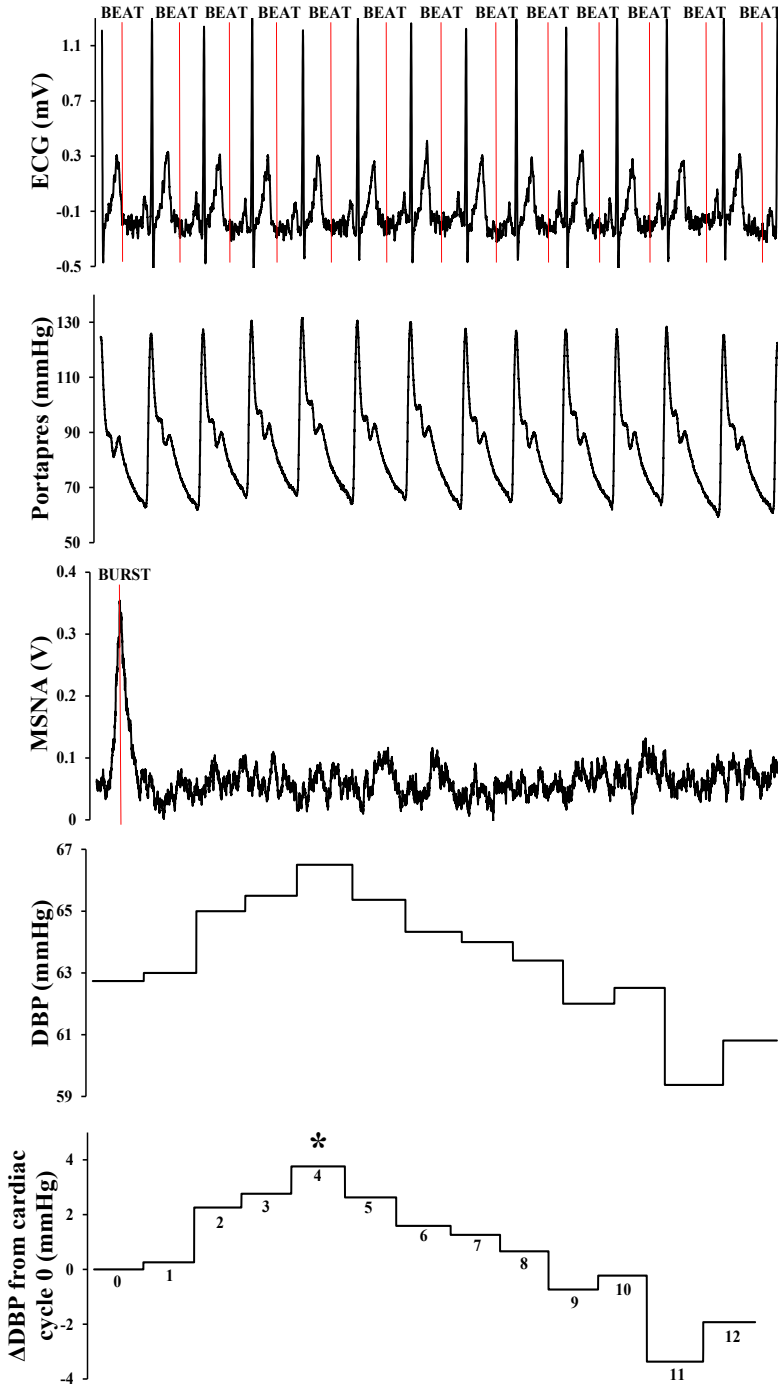

Figure 13.1. Example of time-aligned electrocardiogram, Portapres, integrated muscle sympathetic nerve activity (MSNA), beat-by-beat calculated diastolic blood pressure (DBP), and absolute ΔDBP recordings in LabChart. Each cardiac cycle is identified by a comment labelled 'Beat'. The integrated MSNA channel is shifted backwards to align with the 'Beat' comment from the cardiac cycle during which it was generated. This allows for extraction of the data based on each cardiac cycle. As shown in the ΔDBP channel (bottom), the numbers (0-12) represent the cardiac cycle from the first burst in the MSNA channel and the asterisk corresponds to the peak increase in DBP that occurred 4 cardiac cycles following the burst. The ΔDBP is based on the calculated DBP channel as the difference from cardiac cycle zero (e.g., ~4.5 mmHg from cardiac cycle 0 to 4).

3.3 **Experimental Design**

Figure 3.2 displays a schematic of the study design involved with the 12-week HIIT intervention. Participants in the HIIT group visited the lab 5 times (excluding training days) and participants in the Control group visited the lab 4 times. Anthropometrics and aerobic fitness were assessed at 3 time points (baseline, week 7, follow-up/post-training) for the HIIT group and 2 time points (baseline and follow-up after 12-weeks) for the Control group. Microneurography and hemodynamic measurements were completed during a separate session at baseline and follow-up for each group. At least 48-hour separated each pair of assessments. Physical activity monitoring was conducted at baseline and follow-up for both the HIIT and Control groups. Participants in the Control group were asked to maintain their usual habitual physical and sedentary activity patterns over the 12-week period.

All microneurography sessions were performed >6-hours post-prandial in a thermoneutral environment (~21°C) (40). Participants voided their bladder before testing to minimize the impact of bladder distension on increases in blood pressure and MSNA (64). Furthermore, participants avoided strenuous physical activity 24-hours prior, and the consumption of products known to acutely influence vascular function (e.g., caffeine, alcohol, chocolate, citrus fruits, saturated fats, folic acid, antioxidants, and multivitamin supplements) for 12-hours before these visits (65). Participants were reminded of the testing requirements ~36 hours before the testing session. Pre-study testing requirements were confirmed verbally upon arrival for testing.

The supervised, cycle ergometry-based (via Lode Excalibur Sport or Lode Corival, Groningen, The Netherlands) HIIT sessions occurred in groups of 2-3 for 3-days/week (Mondays, Wednesdays, Fridays). To ensure an adequate training stimulus, participants needed

to complete >80% of the 36 HIIT training sessions (i.e., 29 sessions minimum) and could not miss 3 sessions in a row. All training sessions consisted of a 5-minute warm-up and cool-down period of cycling at 25% PAP. This protocol differs from other HIIT protocols as it is a higher volume protocol (>45-minutes) with shorter intervals (e.g., 15-second versus ≥1-minute on/off periods). During the first 3-weeks of HIIT training, participants completed a set of forty, 15-second intervals at 100% PAP, interspersed with 15-seconds passive recovery periods (i.e., sitting on the ergometer without pedalling). This set was repeated with a 5-minute passive recovery in between (i.e., 40-min of exercise in total). To account for anticipated increases in aerobic fitness, PAP, and exercise tolerance, the set durations were increased to 46-minutes (2 × 23-minutes; 2 sets of 46 intervals) for weeks 4 and 5 (i.e., 15% increase in duration). However, the duration of training was decreased back to 40-minutes (2 × 20-minutes; 2 sets of 40 intervals) for week 6 to provide a taper before the mid-training (i.e., week 7) assessment of aerobic fitness. This taper week ensured that participants were recovered for optimal performance on the mid-training assessment of aerobic fitness, and ensured participants were not over-training, to avoid injury and undue fatigue (53).

Mid-training and baseline PAP were compared, and the higher PAP used to set the intensity for the second half of training. During weeks 8-9, the intensity was set to 100% of the highest PAP and the set durations were increased to 52-minutes (2 × 26-minutes; 2 sets of 52 intervals) (i.e., 30% total increase in duration from week 1). The intensity was increased to 110% of PAP for weeks 10-11 and 115% PAP for week 12. The set durations were decreased to 40-minutes with the intensity reduced to of 100% PAP for the final week of training (i.e., week 13). This training regimen was based on the protocol reported by Mekari et al. (23). This protocol follows the typically recommended progressions for an exercise program wherein the volume of

training is initially increased via longer durations, followed by a higher exercise intensity (53).

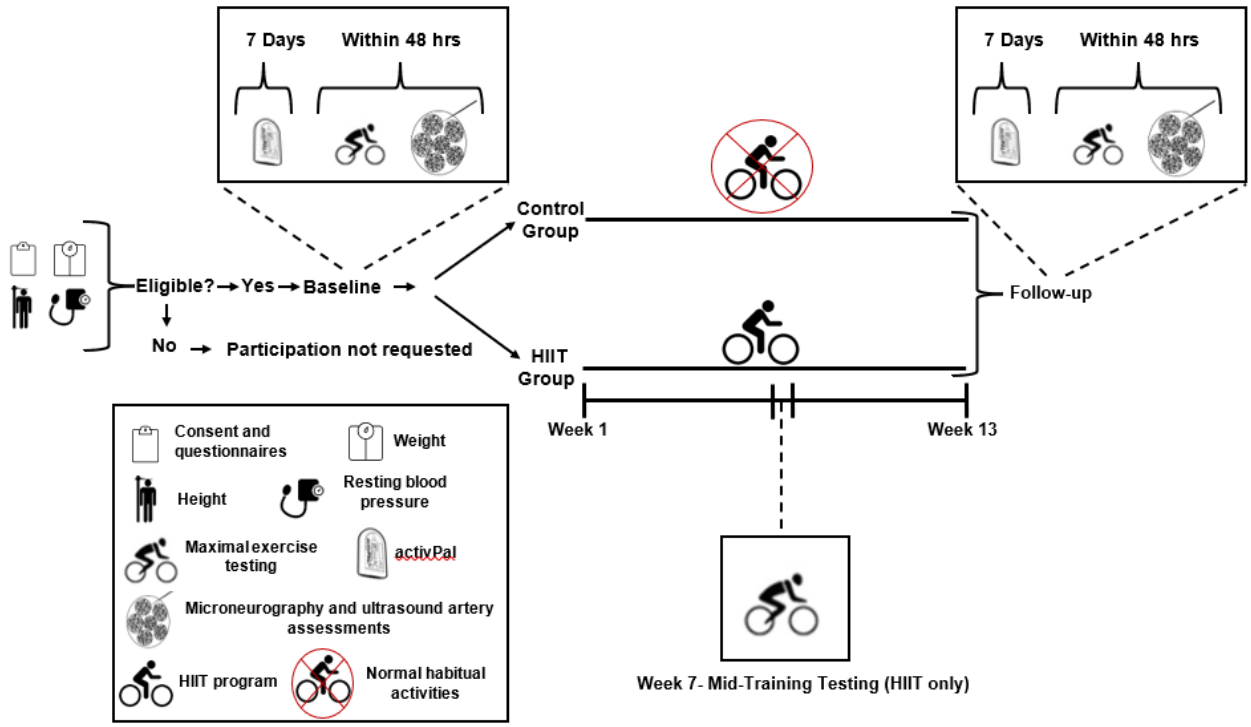

Figure 14. 12-week high intensity interval training (HIIT) research protocol schematic. Questionnaires include the health history questionnaire and Canadian Society for Exercise Physiology Get Active Questionnaire.

3.4 **Statistical Analyses**

All dependent variables were assessed for normality using a Shapiro Wilk test. All variables were confirmed to be normally distributed apart from ratings of perceived exertion, vigorous physical activity, and cardiac cycles to peak and nadir DBP responses. Non-normal data were analyzed for between subject effects using a Mann-Whitney U test and within subject effects using a Wilcoxon signed-rank test. Normally distributed participant characteristics, systemic hemodynamic data, MSNA burst characteristics, as well as peak and nadir DBP responses were compared between groups (i.e., Control versus HIIT) using 2-way repeated measures (Pre- versus Post-intervention) analysis of variance (ANOVA). The variance of

differences (assumption of sphericity) was assessed using Mauchly's test and when violated, the Greenhouse-Geisser correction to the degrees of freedom applied. Bonferroni *post hoc* testing was conducted for statistically significant ANOVAs. Similarly, baseline, 7-week, and 13-week aerobic fitness measurements were compared within the HIIT group using a one-way repeated measures ANOVA. ANOVA effect sizes were calculated for main effects and *post hoc* analyses as Partial Eta Squared (η_p^2). Strength of effect sizes were determined as small (0.01- 0.06), medium (0.06-0.14) or large ($\geq$0.14). All statistical analyses were conducted in SPSS Version 26 (IBM, NY). Data are presented as means ± SD. Statistical significance was accepted as $P < 0.05$.

Chapter 4: RESULTS

4.1 **Participant Characteristics and Aerobic Fitness**

Seventeen participants were recruited for this study. Fourteen participants completed all measurements. Two Control participants were excluded due to the inability to get a successful MSNA signal. One additional participant was excluded from the Control group at follow-up due to COVID-19 related illness, resulting in an inability to complete aerobic fitness testing. This resulted in a final Control group sample of 5 individuals (23 ± 1 years, 3 females). Nine individuals (25 ± 8 years, 5 females) completed the 12-weeks of HIIT training. Adherence to the 36 HIIT sessions was 95 ± 5.5%.

All data obtained from the $\dot{V}O_2$ peak tests are presented in Table 4.1. No Group × Time interaction effect was observed for changes in absolute $\dot{V}O_2$ peak (P=0.156). No changes in relative $\dot{V}O_2$ peak were observed in the Control group after 12-weeks (P=0.279; Figure 4.1A). There were also no differences in PAP, peak METs, or ventilatory threshold (% of $\dot{V}O_2$ peak) observed in the Control group (all, P>0.201; Table 4.1).

In contrast, the HIIT group improved relative $\dot{V}O_2$ peak after both 6- (P=0.007) and 12-weeks of training compared to baseline (P=0.003; Figures 4.1A and B). No differences in relative $\dot{V}O_2$ peak were observed between the HIIT and Control groups at baseline (P=0.457) or follow-up (P=0.115). Corresponding with these HIIT-mediated increases in aerobic fitness, PAP and peak METs were also greater after 6- and 12-weeks of training (all, P<0.007, Table 4.1). Furthermore, relative $\dot{V}O_2$ peak, peak METs, and PAP were all greater after 12-weeks versus 6-weeks of HIIT (all, P<0.042). A Group × Time interaction effect was observed for increases in the ventilatory threshold (P=0.043). Although no differences were observed within the HIIT group (P=0.061), the HIIT group had a greater ventilatory threshold than the Control group at

follow-up (P=0.001). No Group × Time interaction effect was observed for HR at the ventilatory threshold (P=0.130).

Habitual activity data are also presented in Table 4.1. Both groups completed a similar number of steps and time spent engaged in light, moderate, and vigorous physical activity at baseline and follow-up (all, P>0.516). Additionally, both groups had similar amounts of sedentary and standing time at these time points (both, P>0.630).

4.2 **Resting Hemodynamics and Muscle Sympathetic Nerve Activity**

In the HIIT group, the MSNA success rate was 100% (9/9) for both baseline and follow-up. In the Control group, the MSNA success rate was 86% (6/7) at baseline and 83% (5/6) at follow-up. Data are only presented for participants with complete data sets (i.e., 9 HIIT and 5 Control). Systemic hemodynamics, MSNA burst characteristics, and sympathetic transduction outcomes are presented in Table 4.2. The HIIT and Control groups had similar resting SBP, DBP, MAP, and HR that were unchanged at follow-up (all, P>0.070). No interaction effects were observed for MSNA burst frequency, burst incidence, normalized burst size, or total activity (all, P>0.523).

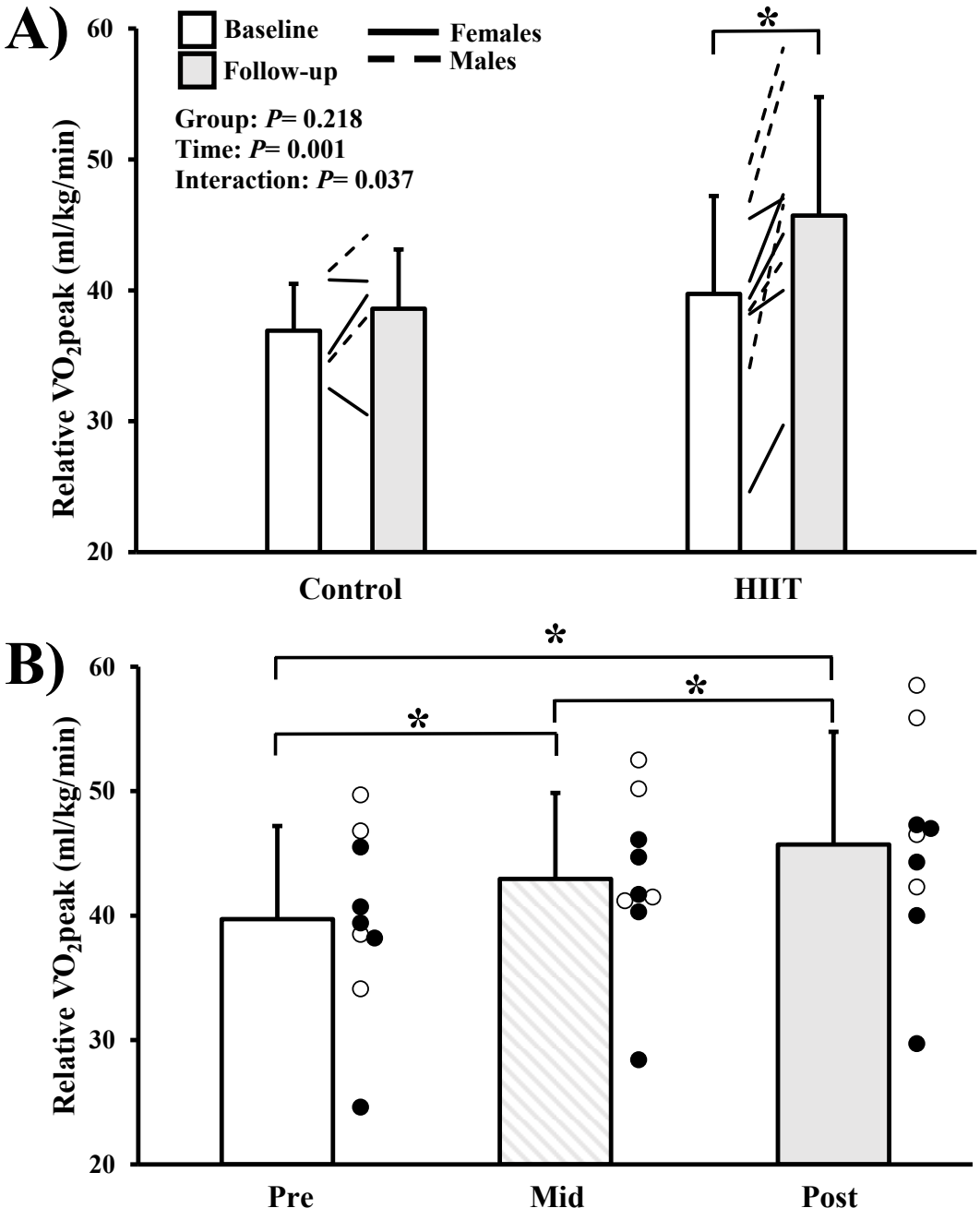

Figure 15. A) Group relative $\dot{V}O_2$ peak for both Control and HIIT between baseline and follow-up. Baseline data are presented in white and follow-up data are presented in grey. Individual data for Control (n=5) and HIIT (n=9) are presented between the group mean bars. Females are presented as solid lines and males as dotted lines. Group × Time interactions were assessed using a 2-way repeated measures analysis of variance with Bonferroni *post hoc* pairwise comparisons. *, P<0.05. B) HIIT group increases in relative $\dot{V}O_2$ peak between baseline (Pre), 6-week (Mid), and follow-up (Post) aerobic fitness testing. Individual data (n=9) are presented to the right of the group means. Females are presented as filled circles. Differences between time points were assessed using a one-way repeated measures analysis of variance with Bonferroni *post hoc* pairwise comparisons. *, P<0.05.

Table 4.1. Participant characteristics, aerobic fitness, and habitual activity

	HIIT (n=9, 5♀)		Control (n=5, 3♀)		Interaction *P* Value	Effect Size (η_p^2)
	Pre	Post	Pre	Post		
Participant Characteristics						
Weight (kg)	70 ± 10	68 ± 10	73 ± 12	74 ± 13	0.126	0.184
BMI (kg/m^2)	23.7 ± 2.7	23 ± 3	23.5 ± 2.9	24 ± 3	0.183	0.143
Aerobic Fitness						
Peak Aerobic Power (watts)	241 ± 60	274 ± 61*	258 ± 41	254 ± 29	**0.002**	0.560
Absolute $\dot{V}O_2$peak (L/min)	2.8 ± 0.7	3.1 ± 0.8	2.7 ± 0.5	2.9 ± 0.6	0.156	0.161
Relative $\dot{V}O_2$peak (ml/kg/min)	39.7 ± 7.5	45.7 ± 9.0*	36.9 ± 3.6	38.6 ± 4.5	**0.037**	0.313
Peak METs	11 ± 2	13 ± 3*	11 ± 1	11 ± 1	**0.028**	0.341
Aerobic Fitness Percentile Rank	50 ± 29	72 ± 29	34 ± 19	42 ± 21	0.127	0.183
Maximum Heart Rate (beats/min)	188 ± 9	190 ± 10	192 ± 5	190 ± 5	0.150	0.165
Peak RER ($\dot{V}CO_2$/ $\dot{V}O_2$)	1.21 ± 0.06	1.29 ± 0.08	1.23 ± 0.08	1.23 ± 0.08	0.322	0.082
Peak RPE	20 ± 1	20 ± 0	19 ± 1	19 ± 0	-	-
VT (%$\dot{V}O_2$peak)	69 ± 9	75 ± 6†	64 ± 4	58 ± 7	**0.043**	0.300
Heart Rate at VT (beats/min)	161 ± 11	169 ± 12	152 ± 7	150 ± 11	0.130	0.181
Habitual Activity						
Step Count (steps/day)	12445 ± 3121	11893 ± 4367	11989 ± 2347	10854 ± 2960	0.517	0.036
Sedentary Time (mins/day)	434 ± 109	463 ± 107	462 ± 56	494 ± 61	0.630	0.020
Standing Time (mins/day)	390 ± 62	363 ± 89	370 ± 80	340 ± 47	0.772	0.007
Light Physical Activity (mins/day)	95 ± 22	81 ± 30	88 ± 24	81 ± 18	0.675	0.015
Moderate Physical Activity (mins/day)	57 ± 24	50 ± 27	37 ± 17	41 ± 15	0.516	0.036
Vigorous Physical Activity (mins/day)	5 ± 4	6 ± 7	6 ± 7	5 ± 6	-	-
MVPA (mins/week)	413 ± 176	395 ± 226	280 ± 125	298 ± 113	0.794	0.006

Data are presented as means ± standard deviations. $\dot{V}O_2$peak, peak volume rate of oxygen consumption; METs, metabolic equivalents of task; RER, respiratory exchange ratio; RPE, rating of perceived exertion; VT, Ventilatory Threshold; MVPA, moderate-to-vigorous physical activity. Group × Time interaction effects were assessed using a repeated measures analysis of variance with Bonferroni *post hoc* pairwise comparisons to determine within and between group differences. Between subjects (Mann-Whitney U) and within subjects (Wilcoxon signed-rank) non-parametric tests were used to assess Peak RPE and Vigorous Physical Activity. No differences were observed between or within groups for Peak RPE (all, *P*>0.655). †, P<0.05 vs Control group at same time point. *, P<0.05 within group.

4.3 **Sympathetic Transduction**

No Group × Time interaction effect was observed for sympathetic transduction between baseline and follow-up in the Control (1.43±0.90 vs. 1.47±0.64 mmHg) and HIIT groups (1.23±0.50 vs. 1.37±0.52 mmHg, P=0.647, η_p^2=0.018; Figure 4.2A).

In contrast, there was a Group × Time interaction effect for the nadir non-burst decreases in DBP (P=0.049, Figure 4.2B). Specifically, nadir non-burst decreases in DBP were lower within the HIIT group after training (-0.49±0.16 vs. -0.66±0.16 mmHg, P=0.006), but unchanged in the Control group (-0.38±0.20 vs. -0.41±0.19 mmHg, P=0.638). As such, the HIIT group exhibited larger decreases in DBP following periods of sympathetic quiescence at follow-up compared to the Control group (P=0.022).

In both groups and at all time points, there was a graded sympathetic transduction response to burst sequences, with triple+ bursts producing greater increases in DBP than doublets and singlets (all, P<0.023). Furthermore, doublets produced greater increases in DBP than singlets (P<0.001). However, there were no changes in sympathetic transduction following singlets, doublets, or triplet+ between baseline and follow-up or between groups (all, P>0.243). As these data do not address the primary purpose of the study, they are displayed in Supplemental Figure 1 in Appendix E. Time course changes across cardiac cycles for all bursts, singlets, doublets, triplets+, and nadir non-burst responses are presented in Supplemental Figure 2 in Appendix E.

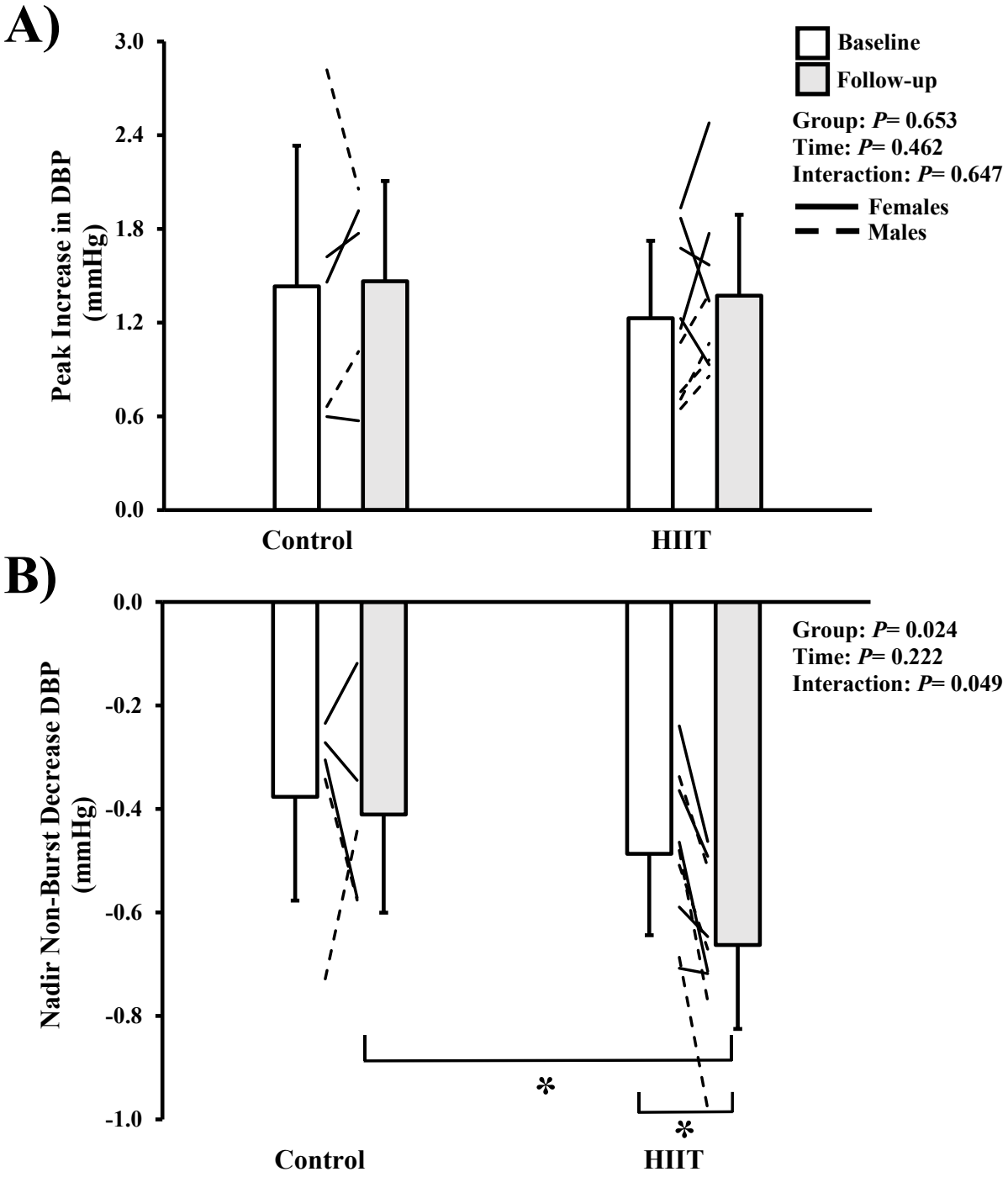

Figure 16. A) Peak increases in diastolic blood pressure (DBP) following spontaneous bursts of muscle sympathetic nerve activity. B) Nadir decreases in DBP during periods of sympathetic quiescence (i.e., cardiac cycles without muscle sympathetic nerve activity bursts). Baseline data are presented in white and follow-up data are presented in grey. Individual data for Control (n=5) and HIIT (n=9) are presented between the group mean bars. Females are presented as solid lines and males as dotted lines. Group × Time interaction effects were assessed using a repeated measures analysis of variance with Bonferroni *post hoc* pairwise comparisons to determine within and between group differences. *, P<0.05.

Table 4.2. Resting hemodynamics, muscle sympathetic nerve activity burst characteristics, and sympathetic transduction characteristics

	HIIT (n=9, 5♀)		Control (n=5, 3♀)		Interaction *P* Value	Effect Size (η_p^2)
	Pre	**Post**	**Pre**	**Post**		
Resting Hemodynamics						
Systolic Blood Pressure (mmHg)	116 ± 8	111 ± 10	112 ± 4	110 ± 2	0.938	0.001
Diastolic Blood Pressure (mmHg)	64 ± 7	65 ± 7	64 ± 5	64 ± 3	0.733	0.010
Mean Arterial Blood Pressure (mmHg)	81 ± 5	80 ± 7	80 ± 4	79 ± 3	0.809	0.005
Heart Rate (beats/min)	61 ± 8	59 ± 7	59 ± 6	59 ± 6	0.070	0.248
Muscle Sympathetic Nerve Activity						
Total Number of Bursts	184 ± 86	164 ± 104	123 ± 23	128 ± 38	0.312	0.085
Burst Frequency (bursts/min)	18 ± 8	17 ± 9	12 ± 3	12 ± 4	0.659	0.017
Burst Incidence (bursts/100 heartbeats)	30 ± 12	23 ± 3	21 ± 7	21 ± 8	0.864	0.003
Normalized Burst Size (% maximum)	45 ± 6	43 ± 8	47 ± 7	45 ± 10	0.939	0.001
Total MSNA (%maximum · bursts/min)	788 ± 319	708 ± 282	578 ± 162	584 ± 308	0.523	0.035
Sympathetic Transduction						
Cardiac Cycles to Peak DBP	5 ± 1	6 ± 3	7 ± 4	5 ± 2	-	-
Cardiac Cycles to Nadir DBP	6 ± 2	6 ± 1	8 ± 4	6 ± 3	-	-
Singlets (% Total Bursts)	41 ± 15	40 ± 17	59 ± 13	50 ± 11	0.230	0.118
Doublets (% Total Bursts)	32 ± 8	31 ± 6	22 ± 8	25 ± 9	0.164	0.155
Triplets+ (% Total Bursts)	27 ± 19	29 ± 18	19 ± 10	25 ± 7	0.355	0.078

Data are presented as means ± standard deviations. 8 participants are included in the percentage triplets+ for the HIIT group. Group × Time interaction effects were assessed using a repeated measures analysis of variance with Bonferroni *post hoc* pairwise comparisons to determine within and between group differences. Between subjects (Mann-Whitney U) and within subjects (Wilcoxon signed-rank) non-parametric tests were used to assess cardiac cycles to peak and nadir DBP. No differences were observed between or within groups (all, *P*>0.147). †, P<0.05 vs Control group at same time point. *, P<0.05 within group.

Chapter 5: DISCUSSION

The primary purpose of this study was to investigate whether 12-weeks of HIIT in younger adults would: 1) improve aerobic fitness and 2) evoke corresponding decreases in sympathetic transduction. Based on previous HIIT studies that observed increases in aerobic fitness after 12-weeks (21), it was hypothesized that the HIIT protocol used in this study would increase relative $\dot{V}O_2$peak. Based on the inverse relationship between sympathetic transduction and aerobic fitness that our group observed in younger males (13), as well as older males and females (18), it was hypothesized that HIIT would decrease sympathetic transduction when aerobic fitness was improved. However, as younger females may not exhibit the same attenuation of sympathetic transduction with higher aerobic fitness levels (19), this study attempted to identify whether there were any discrepancies between sexes in the response to HIIT. Consistent with the first hypothesis, aerobic fitness improved in the HIIT group and remained unchanged in the Control group (Figure 4.1A). However, no corresponding decreases in sympathetic transduction were observed in either group (Figure 4.2A). The results of this study provide the first longitudinal investigation into the impact of aerobic training on sympathetic transduction.

A greater training stimulus is required to observe improvements in $\dot{V}O_2$peak in relatively fit younger adults (24), as represented by our HIIT group (39.7 ± 7.5 ml/kg/min, 50th percentile at baseline, Table 4.1). Specifically, a meta-analysis by Swain and Franklin (66) demonstrated that across differing baseline fitness levels, high-intensity training (>60% $\dot{V}O_2$reserve) improved aerobic fitness to a greater extent than low-to-moderate-intensity training. However, in those with higher baseline fitness (>39 ml/kg/min) a larger relative increase was observed following high-intensity training (66). In the current study, although no increases in absolute $\dot{V}O_2$ were

observed following HIIT (2.8 ± 0.7 vs. 3.1 ± 0.8 L/min), there were also no decreases in body mass (70 ± 10 vs. 68 ± 10). As such, the increases in relative $\dot{V}O_2$peak that were observed cannot be attributed to decreases in body mass. Furthermore, PAP and ventilatory threshold increased in the HIIT group (Table 4.1). Both these findings support that the increases in relative $\dot{V}O_2$peak observed following HIIT are indicative of improved aerobic fitness. Thus, our findings support that 12-weeks of HIIT was effective at improving aerobic fitness in a young, healthy population. Such improvements were observed after only 6-weeks of HIIT (Figure 4.2B). This was in contrast with the results of Mekari et al. (23), who utilised a similar 6-week HIIT protocol in younger adults and reported increases in PAP, but not relative $\dot{V}O_2$peak. Despite starting at a similar level of baseline fitness (39.7 ± 8.7 vs. 39.7 ± 7.5 ml/kg/min), they only observed an ~3% increase in relative $\dot{V}O_2$peak and ~5% increase in PAP (23). In comparison, our HIIT group experienced an ~8% increase in both variables after 6-weeks. Both relative $\dot{V}O_2$peak (~15%) and PAP (~14%) increased further following an additional 6-weeks of training in our participants (Table 4.1). This provides evidence that the continued duration of training in our study elicited further increases in aerobic fitness.

Based on the Fick equation ($\dot{V}O_2$max = cardiac output$_{max}$ × arterial-venous O_2 difference), factors associated with improvements in $\dot{V}O_2$peak may be attributable to improvements in oxygen delivery (i.e., increases in maximal cardiac output) or oxygen extraction (i.e., arterial-venous O_2 differences) (67). Thus, it is likely that the HIIT-mediated increases in aerobic fitness are associated with changes in central or peripheral factors impacting oxygen delivery and/or extraction. Increases in maximal heart rate are not commonly observed following training (68). This is consistent with our results (Table 4.1). Thus, it is plausible that increases oxygen delivery were mediated primarily via increases in stroke volume leading to increases in cardiac output.

Increases in stoke volume may have been achieved through enhanced diastolic filling (69), greater plasma and erythrocyte volume (70, 71), and improved left ventricular function (69). Furthermore, peripheral adaptations may have occurred which could increase arterial-venous O_2 differences. These adaptations may have included an increased mitochondrial volume (72) and mitochondrial enzyme activity following training (73) which permit greater extraction of oxygen from the blood. Although, these factors were not able to be confirmed in the current study, it is likely that some combination of these physiological adaptations contributed to the HIIT-mediated increases in aerobic fitness.

Despite observing increases in aerobic fitness, no corresponding reductions in sympathetic transduction were observed (Figure 4.2A). This is inconsistent with the previous cross-sectional observations from our lab that reported an inverse relationship between relative $\dot{V}O_2$peak and sympathetic transduction in younger males (13) and older adults (18). However, these findings support the results of Stickford et al. (19) who observed no differences in resting sympathetic transduction between young females with higher or lower aerobic fitness levels. As such, sex may be an important factor to consider when investigating the impact of aerobic exercise interventions on sympathetic transduction. Although this study was underpowered to determine sex differences, 5 out of 9 HIIT participants were female. Previous research has demonstrated that females have lower α-adrenergic receptor-mediated vasoconstrictor responses to norepinephrine (74, 75) and greater β_2-adrenergic receptor-mediated vasodilation (75–77) compared to age-matched males. Considering that sympathetic transduction is α-adrenergic receptor-mediated (8), the null HIIT-mediated sympathetic transduction result may have been impacted by having females included in the sample. However, exploring the responses separately between males (Pre: 0.80 ± 0.56 vs. Post: 1.07 ± 0.23mmHg) and females (Pre: 1.57 ± 0.36 vs.

Post: 1.62 ± 0.57mmHg) (Figure 4.2), it does not appear that there were any consistent differences between the sexes (2 females increased, 2 decreased, 1 similar and all males increased). This is interesting given that there was an apparently larger increase in relative $\dot{V}O_2$peak in the male (~20% from baseline) compared to the female participants (~10% from baseline). Nonetheless, considering the discrepant cross-sectional findings regarding the relationship between aerobic fitness and sympathetic transduction in males versus females highlighted above, it will be important to determine in a larger sample whether sex differences exist in sympathetic transduction following HIIT.

Compared to baseline, there were greater decreases in DBP during periods of sympathetic quiescence in the HIIT group at follow-up (Figure 4.2B). This decrease was apparent in both male and female participants. Although this study was not designed to investigate mechanisms associated with training-induced decreases in DBP following heartbeats without bursts of MSNA, this finding may be indicative of enhanced local vascular vasodilatory mechanisms (e.g., greater endothelial-derived nitric oxide availability) following HIIT (78, 79). Furthermore, evidence of enhanced limb blood flow capacity following aerobic training has been suggested to reflect a greater total cross-sectional area of resistance vessels as well as improved resistance vessel responsiveness (25, 80–82). This may be of importance as resistance vessels have a larger overall impact on blood pressure compared to larger arteries. If resistance vessel remodelling occurs after HIIT, this could contribute to a decreased vascular resistance and lower DBP during periods without MSNA (83). These potential adaptations suggest that although HIIT did not influence the pressor response to MSNA bursts, other vascular adaptations may have occurred to lead to larger decreases in blood pressure following heartbeats without bursts of MSNA. Vascular adaptations to exercise training may occur in a shorter period than neural adaptations.

Perhaps, over a longer training period, decreases in sympathetic transduction would be apparent coincident with the decreases in blood pressure during heartbeats without MSNA.

5.1 **Strengths and Limitations**

This study was the first longitudinal investigation into aerobic training-induced changes in sympathetic transduction. As such, in addition to the HIIT aspect of the study, our Control group provided the first repeated measurements of sympathetic transduction. To this extent, while we attempted to control for between-day variation in diet, exercise, hydration, and sleep, it was apparent that day-to-day reproducibility of sympathetic transduction may be low (Figure 4.2A). There are many potential confounding variables that can influence blood pressure variability on a day-to-day basis. Factors such as mental stress (e.g., participant was uncomfortable, anxious, or impatient) (84), breathing rate (e.g., deep slow breaths vs. shallow faster breaths) (12) and hydration level (e.g., hormone levels of the renin-angiotensin-aldosterone system) (85) all have an influence on MSNA and blood pressure and therefore, may influence sympathetic transduction. Further research is needed to explore this variability and establish the day-to-day reliability of sympathetic transduction. Future consideration of specific factors that may influence sympathetic transduction is needed to ensure consistency between studies. In our study, lifestyle factors such as aerobic fitness and habitual activity remained similar within the Control group at baseline and follow-up and thus, are unlikely to have influenced the variability of the sympathetic transduction measures.

In the HIIT group, peak aerobic fitness increased by 2 METs. Previous longitudinal follow-up studies have demonstrated that a 1 MET increase in aerobic fitness is associated with a 10-25% greater survival rate at follow-up in healthy asymptomatic adults (6). Thus, a 2 MET increase in our population is indicative of a positive cardiovascular effect which is beneficial for

our participants. It remains unknown whether the increases in aerobic fitness are attributable to improvements in oxygen delivery, oxygen extraction, or a combination of both. Although 12-weeks of HIIT was sufficient to evoke increases in aerobic fitness, it is unknown whether this is a long enough period to observe changes in sympathetic transduction. Perhaps, in healthy humans, a longer period is needed to truly alter α-adrenergic receptor sensitivity and/or density to reduce sympathetic transduction.

An assumption of this research is that following an MSNA burst, there was a corresponding increase in peripheral vascular resistance, which led to a transient rise in DBP. However, we did not determine the amount of norepinephrine and/or co-transmitter released and the subsequent local effects on the vasculature. Previous work by Fairfax and colleagues (8) demonstrated a robust attenuation of sympathetic transduction of forearm vascular conductance following α-adrenergic blockade. However, this was unable to be confirmed in the current study. Therefore, it is not known with certainty how HIIT specifically impacted local sympathetic neurovascular control mechanisms. Future research should investigate how systemic measures of sympathetic transduction, such as DBP, are influenced following α-adrenergic receptor inhibition and the impacts of HIIT on local neurovascular sympathetic transduction.

5.2 **Perspectives and Future Directions**

In the current study, it remains possible that despite the HIIT-induced improvements in aerobic fitness, there may have been little room for further reductions in sympathetic transduction to be gained (i.e., flooring effect) since the cohort was healthy, young, and physically active,. Moving forward, it will be important to address this question in populations with increased cardiovascular disease risk or existing disease. To this point, sympathetic transduction is known to decrease with aging (9, 16, 86). However, the inverse relationship

between aerobic fitness and sympathetic transduction persists with age (18), despite the overall reduced sympathetic transduction. Thus, whether a flooring effect would also been observed in an older population who already have lower sympathetic transduction is unknown. In those with resistant hypertension, reduced sympathetic transduction has also been observed (27). This brings about a paradox regarding what having 'low' sympathetic transduction means, as it relates to being a healthy versus unhealthy phenotype. In older populations and cardiovascular disease, patient's lower sympathetic transduction may be related to divergent mechanisms (e.g., increased MSNA, changes in arterial structure) and as such, how increases in aerobic fitness modify sympathetic transduction may differ from a healthy, young population and should be explored further.

5.3 **Conclusion**

This study demonstrated that 12-weeks of HIIT training improved relative $\dot{V}O_2$ peak in fit young adults. Although the peak DBP response to bursts of MSNA were unchanged after HIIT, larger nadir non-burst decreases in DBP were observed after training. This may be indicative of improvements in local vascular vasodilatory mechanisms. These positive adaptations following training may reduce vascular resistance and thus, lower blood pressure during sympathetic quiescence. These adaptations corresponded with the well-established positive cardiovascular health effect of improving aerobic fitness. This study provides the first longitudinal investigation into changes in sympathetic transduction and provides a starting point for determining the repeatability of the transduction outcomes, as well as the influence that improved aerobic fitness has on the sympathetic control of blood pressure.

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Appendix A: Health History Questionnaire

Age: _____ years

PARTICIPANT I.D. (Completed by Research Team): _____
Determination of body mass index (BMI):

i. What is your approximate weight in kilograms? _____
To convert from pounds to kilograms, multiply by 0.454

ii. What is your approximate height in meters? _____
To convert from inches to meters, multiple by 0.0254

iii. To calculate BMI, please click this link:
http://www.nhlbi.nih.gov/health/educational/lose_wt/BMI/bmicalc.htm

Calculated BMI: _____kg/m^2

The following questions will determine your eligibility for the study. If you answer 'Yes to any question you will <u>not</u> be able to participate in the study.

1. Was your calculated body mass index above <u>greater than</u> 30 kg/m^2? ☐YES ☐ NO
2. Are you younger than 18 years old? ☐YES ☐ NO
3. Have you smoked or consumed any nicotine/marijuana-containing products daily within the past 6 months? ☐YES ☐ NO
4. Are you allergic to Tegaderm™ (3M) medical adhesive dressing? ☐YES ☐ NO
5. Have you been prescribed medications for high blood pressure? ☐YES ☐ NO
6. Do you have a cardiovascular, neural (e.g., Raynaud's disease), respiratory or metabolic disorder (e.g., diabetes)? ☐YES ☐ NO
7. Are you afraid of needles? ☐YES ☐ NO

<u>For females only:</u>

7. Are you pregnant, breastfeeding or intending to become pregnant in the next 3 months? ☐YES ☐ NO
1. Are you currently on, or planning on starting, hormone replacement therapy? ☐YES ☐ NO
2. If you are <u>55 years or older</u>: Have you *had* a menstrual period in the last 12 months? ☐ not applicable ☐YES ☐ NO
10. If you are <u>younger than</u> 55 years: Have you been *without* a menstrual period for the last 12 months? ☐ not applicable ☐YES ☐ NO

Appendix B: Get Active Questionnaire

CSEP | SCPE
THE GOLD STANDARD IN EXERCISE SCIENCE AND PERSONAL TRAINING

Get Active Questionnaire

CANADIAN SOCIETY FOR EXERCISE PHYSIOLOGY –
PHYSICAL ACTIVITY TRAINING FOR HEALTH (CSEP-PATH®)

Physical activity improves your physical and mental health. Even small amounts of physical activity are good, and more is better.

For almost everyone, the benefits of physical activity far outweigh any risks. For some individuals, specific advice from a Qualified Exercise Professional (QEP – has post-secondary education in exercise sciences and an advanced certification in the area – see csep.ca/certifications) or health care provider is advisable. **This questionnaire is intended for all ages – to help move you along the path to becoming more physically active.**

- ☐ I am completing this questionnaire for myself.
- ☐ I am completing this questionnaire for my child/dependent as parent/guardian.

PREPARE TO BECOME MORE ACTIVE

The following questions will help to ensure that you have a safe physical activity experience. Please answer **YES** or **NO** to each question before you become more physically active. If you are unsure about any question, answer **YES**.

	YES	NO
1 Have you experienced **ANY** of the following (A to F) **within the past six months**?		
A A diagnosis of/treatment for heart disease or stroke, or pain/discomfort/pressure in your chest during activities of daily living or during physical activity?	○	○
B A diagnosis of/treatment for high blood pressure (BP), or a resting BP of 160/90 mmHg or higher?	○	○
C Dizziness or lightheadedness during physical activity?	○	○
D Shortness of breath at rest?	○	○
E Loss of consciousness/fainting for any reason?	○	○
F Concussion?	○	○
2 Do you currently have pain or swelling in any part of your body (such as from an injury, acute flare-up of arthritis, or back pain) that affects your ability to be physically active?	○	○
3 Has a health care provider told you that you should avoid or modify certain types of physical activity?	○	○
4 Do you have any other medical or physical condition (such as diabetes, cancer, osteoporosis, asthma, spinal cord injury) that may affect your ability to be physically active?	○	○

➤ **NO** to all questions: go to Page 2 – ASSESS YOUR CURRENT PHYSICAL ACTIVITY ➤

YES to any question: go to Reference Document – ADVICE ON WHAT TO DO IF YOU HAVE A YES RESPONSE ➤➤

Get Active Questionnaire

ASSESS YOUR CURRENT PHYSICAL ACTIVITY

Answer the following questions to assess how active you are now.

1 During a typical week, on how many days do you do moderate- to vigorous-intensity aerobic physical activity (such as brisk walking, cycling or jogging)? ____ DAYS/WEEK

2 On days that you do at least moderate-intensity aerobic physical activity (e.g., brisk walking), for how many minutes do you do this activity? ____ MINUTES/DAY

For **adults**, please multiply your average number of days/week by the average number of minutes/day: ____ MINUTES/WEEK

Canadian Physical Activity Guidelines recommend that adults accumulate at least 150 minutes of moderate- to vigorous-intensity physical activity per week. For children and youth, at least 60 minutes daily is recommended. Strengthening muscles and bones at least two times per week for adults, and three times per week for children and youth, is also recommended (see csep.ca/guidelines).

GENERAL ADVICE FOR BECOMING MORE ACTIVE

Increase your physical activity gradually so that you have a positive experience. Build physical activities that you enjoy into your day (e.g., take a walk with a friend, ride your bike to school or work) and reduce your sedentary behaviour (e.g., prolonged sitting).

If you want to do **vigorous-intensity physical activity** (i.e., physical activity at an intensity that makes it hard to carry on a conversation), and you do not meet minimum physical activity recommendations noted above, consult a Qualified Exercise Professional (QEP) beforehand. This can help ensure that your physical activity is safe and suitable for your circumstances.

Physical activity is also an important part of a healthy pregnancy.

Delay becoming more active if you are not feeling well because of a temporary illness.

DECLARATION

To the best of my knowledge, all of the information I have supplied on this questionnaire is correct.
If my health changes, I will complete this questionnaire again.

I answered NO to all questions on Page 1

Sign and date the Declaration below

I answered YES to any question on Page 1

Check the box below that applies to you:

- ☐ I have consulted a health care provider or Qualified Exercise Professional (QEP) who has recommended that I become more physically active.
- ☐ I am comfortable with becoming more physically active on my own without consulting a health care provider or QEP.

Name (+ Name of Parent/Guardian if applicable) [Please print] | Signature (or Signature of Parent/Guardian if applicable) | Date of Birth

Date | Email (optional) | Telephone (optional)

With planning and support you can enjoy the benefits of becoming more physically active. A QEP can help.

☐ Check this box if you would like to consult a QEP about becoming more physically active.
(This completed questionnaire will help the QEP get to know you and understand your needs.)

Appendix C: Letter of REB Approval

DALHOUSIE UNIVERSITY
Research Services

Health Sciences Research Ethics Board
Letter of Approval

May 25, 2021

Derek Kimmerly
Health\School of Health and Human Performance

Dear Derek,

REB #: 2021-5555
Project Title: The impact of a 12-week high-intensity interval training program on arterial health

Effective Date: May 25, 2021
Expiry Date: May 25, 2022

The Health Sciences Research Ethics Board has reviewed your application for research involving humans and found the proposed research to be in accordance with the Tri-Council Policy Statement on *Ethical Conduct for Research Involving Humans*. This approval will be in effect for 12 months as indicated above. This approval is subject to the conditions listed below which constitute your on-going responsibilities with respect to the ethical conduct of this research.

Effective March 16, 2020: Notwithstanding this approval, any research conducted during the COVID-19 public health emergency must comply with federal and provincial public health advice as well as directives from Dalhousie University (and/or other facilities or jurisdictions where the research will occur) regarding preventing the spread of COVID-19.

Sincerely,

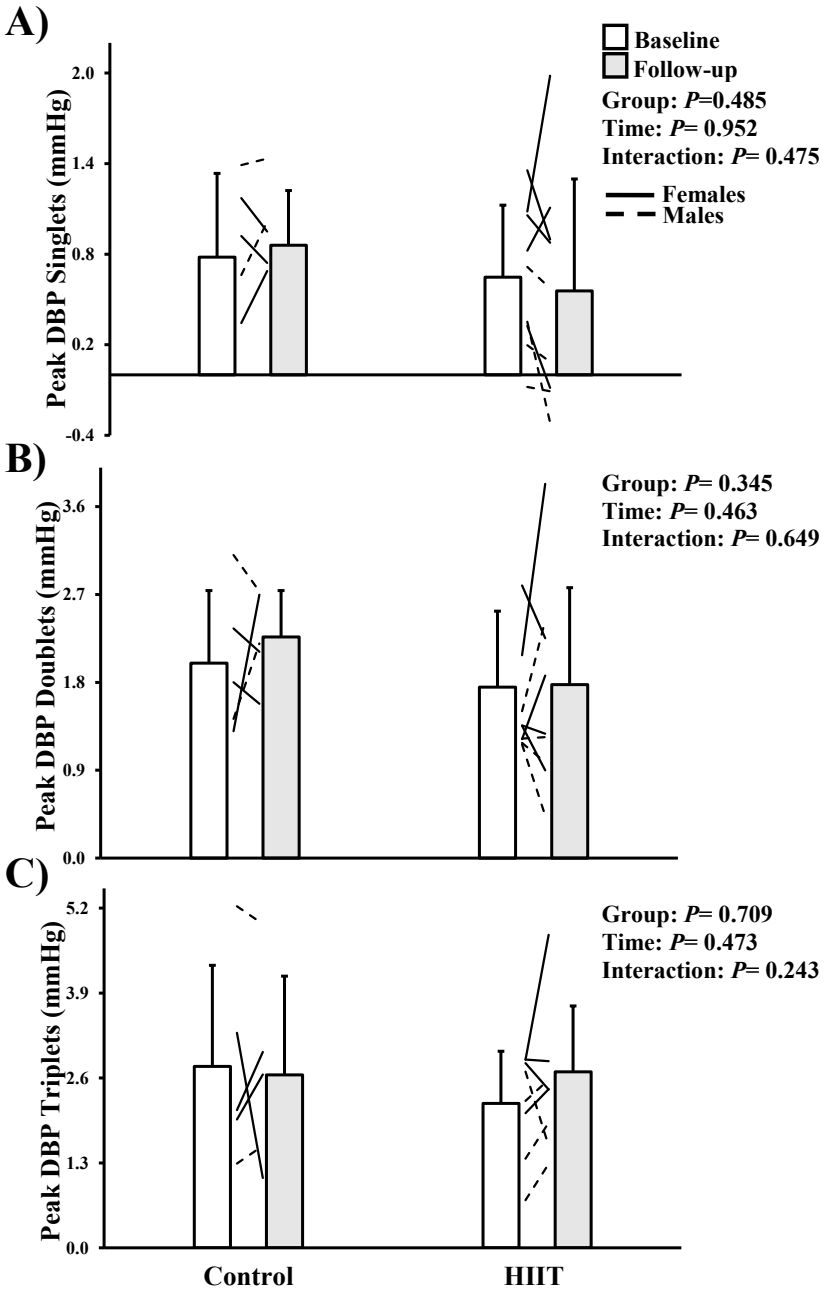
Dr. Lori Weeks, Chair

Appendix D: LabChart Analysis Instructions

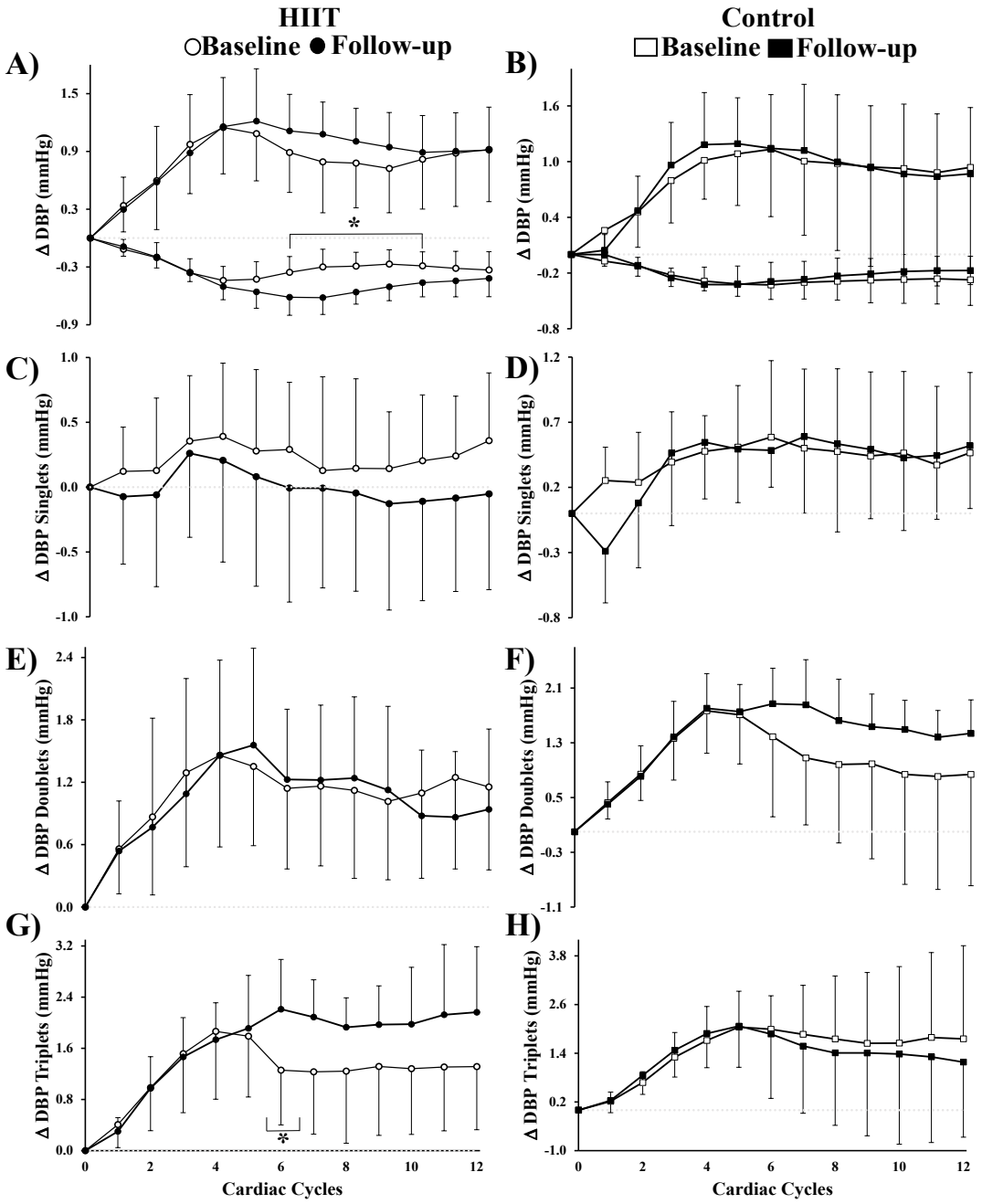
1. Calibrate the Portapres signal using the static automated blood pressure measurements using unit conversion.
2. Derive heart rate, stroke volume, systolic blood pressure, and diastolic blood pressure, into their own channels using cyclic measurements or the non-invasive cardiac output add-on (stroke volume only).
    a. Setup → Channel Settings → Use number of channels along bottom of pop-up box to add or remove channels → Create new channels for HR, SBP, DBP, SV
    b. Heart rate → Calculation → Cyclic Measurements → Source → ECG → Measurement → Rate → Change preset detection setting to "ECG - Human" → adjust detection adjustment if it isn't picking up the peak r-waves
    c. Systolic blood pressure → Calculation → Cyclic Measurements → Source → Portapres → Measurement → Maximum → Preset detection to "Cardiovascular - Arterial pressure"
    d. Diastolic blood pressure → Calculation → Cyclic Measurements → Source → Portapres → Measurement → Minimum → Preset detection to "Cardiovascular - Arterial pressure"
    e. Stroke Volume → Calculation → Non-invasive cardiac output → Pressure signal → Portapres (must already be calibrated to mmHg) → Method→ Windkessel Model → Output → Stroke Volume → Adjust subject age and sex
3. Time align the Portapres signal to the ECG signal using the marker ('M' in bottom left-hand corner).
    a. Zoom in on a small section of data → Drag "M" to the peak of an r-wave in the ECG channel → place cursor at the closest peak in the Portapres channel → a time difference between the "M" and where the cursor is will show in the top right hand corner of the screen above where the channels are listed → this time difference is the shift in the data → Click on the Portapres menu → Shift → insert time difference (negative numbers will shift backwards, positive shifts forwards)
    b. The MSNA signal will need to be shifted to the middle of the Portapres and ECG signal so that each burst occurs in the middle of the ECGs R-R interval to ensure each MSNA burst corresponds to a cardiac cycle. This can be done using the same method as above, but should be done after completing the "beat macro" in section 2 (i.e. shift bursts to align with "beat comment").
4. Set up data pad. In data pad, each channel will have its own column automatically. Create the following new channels (in any order):
    a. Int MSNA Maximum – Minimum (Statistics → Source Int MSNA)
    b. Int MSNA Comment Number (Comment → Source Int MSNA)
    c. Int MSNA Integral Relative to Baseline (V*s) (Integral → Source Int MSNA)
    d. Time (Selection & Active Point)
    e. Int MSNA Comment Time (Comment → Source Int MSNA)
    f. ECG Full Comment Text (Comment → Source ECG)
    g. ECG Comment Number (Comment → Source ECG)
    h. Mean Heart Rate (Statistics → Source Heart Rate)
    i. Mean SBP (Statistics → Source SBP)
    j. Mean DBP (Statistics → Source DBP)
    k. Mean SV (Statistics → Source SV)

5. If you have the macros already on your computer (they can be imported), play them or create them following instructions below.
    a. Two macros are needed for this. One to identify each MSNA burst and one to identify each cardiac cycle. Click anywhere in the corresponding channel to begin (e.g., integrated MSNA recording in row 3).
        i. MSNA Burst Macro: Start Recording Macro → Recording Commands → Repeat while in Block → Commands → Find → Search in MSNA Channel → Find data → Local Maxima → Threshold =1% → Single point selection → Hit 'Enter' → Add Comment 'Burst' in MSNA Channel
            1. Repeat using 'Commands' → 'find next'. After adding a second comment to the next burst, press 'Macro' → Stop Recording (Do not close any other windows that have popped up before stopping recording).
        ii. ECG Beat Macro: Identical to above except for searching in ECG channel, naming the comment 'Beat' and using 'select to previous point' under the find data heading.
    b. Go to the beginning of your resting period with stable MSNA data and run both macros.
6. Go through the integrated MSNA channel and ensure all valid bursts are selected. Add or delete 'Burst' comments when applicable.
7. Delete 'BEAT' comments that are inaccurately detected (could change macro threshold if necessary).
8. Highlight area of interest. Click 'Multiple add to Data Pad'. Find using comment from ECG channel containing 'Beat'. Select 0.9s around comment. Current selection.
    a. 0.9s may not be applicable for everyone, go through the file and use the marker to get a sense of how wide most of your bursts are (record this value). Adjust if bursts are larger than 0.45s to the left and 0.45s to the right of the "burst" comment.
    b. Copy and paste entire Data pad into the first sheet on the excel template provided
    c. (Control-A, control-C, and Control-V to select all data, copy the data and paste the data).

Appendix E: Supplemental Figures



Supplemental Figure 17. A) Peak increases in diastolic blood pressure (DBP) following single spontaneous bursts of muscle sympathetic nerve activity (i.e., a single heartbeat with a burst without bursts associated with the preceding or subsequent heart beats). B) Peak increases in DBP following doublets (i.e., two consecutive heart beats with bursts). C) Peak increases in DBP following triplets+ (i.e., three or more consecutive heart beats with bursts). Baseline data are presented in white and follow-up data are presented in grey. Individual data for Control (n=5) and HIIT (n=9) are presented between the group mean bars. Females are presented as solid lines and males as dotted lines. Group × Time interaction effects were assessed using a repeated measures analysis of variance with Bonferroni *post-hoc* pairwise comparisons to determine within and between group differences. *, $P<0.05$.



Supplemental Figure 18. Averaged 12 cardiac cycle time courses of changes in diastolic blood pressure (DBP) for A) all bursts and non-bursts, C) singlets, E) doublets, and G) triplets+ within the HIIT group for Baseline (open symbols) and Follow-up (filled in symbols). Averaged 12 cardiac cycle time course of changes in diastolic blood pressure (DBP) for B) all bursts and non-bursts, D) singlets, F) doublets, and H) triplets+ within the Control group for Baseline (open symbols) and Follow-up (filled in symbols). Repeated measures analysis of variance compared the responses across cardiac cycles between groups. Significant interaction effects were followed with Bonferroni *post-hoc* pairwise comparisons to determine where differences occurred between groups. *, $P<0.05$ vs. follow-up.

Appendix F: Curriculum Vitae

ACADEMIC BACKGROUND

MSc Kinesiology Candidate, Dalhousie University, Canada

Supervisor: Dr. Derek Kimmerly

Defense Date: 06/2022

BSc Honors Kinesiology, Dalhousie University, Canada

Supervisor: Dr. Derek Kimmerly

Thesis Title: "The importance of endothelial-derived hyperpolarizing factor (EDHF) on low-flow-mediated constriction (L-FMC) in young adults: The influence of aerobic fitness"

EXPERIENCE

Research Assistant, Faculty of Health, Dalhousie University, Halifax, NS **09/2019-present**

- Collect and analyze data for activity monitor validation studies and arterial health studies
- Conduct article screening and data extraction using Covidence for two systematic reviews

Database and Inventory Assistant, Dalhousie University, Halifax, NS **07/2021-present**

- Organize/catalog research and teaching equipment within the Dalhousie Kinesiology department
- Prepare consumable supplies list and ensure all products are purchased and available

Crisis Responder, Kids Help Phone Text Service **02/2022-present**

- Active listening and collaborative problem-solving with texters in moments of distress

Teaching Assistant, Dalhousie University, Halifax, NS **01/2020-05/2022**

- Instruct exercise physiology labs and grade lab assignments

Qualified Exercise Professional, ACCESS, Halifax, NS **01/2019-04/2020**

- Implement exercise training sessions for clinical research study

Tutor, Dalhousie University, Halifax, NS **09/2019-12/2019**

- 2nd year exercise physiology tutor

Athletic Trainer, Dalhousie University, Halifax, NS **08/2018 – 05/2019**

- 300 hours of sideline care, massage, performance taping, first aid for varsity athletes

DISTINCTIONS/ AWARDS

Frederick Banting and Charles Best Canada Graduate Scholarship-Master's, CIHR

Monetary allotment: $17,500 over 1 year

Top Graduate Student Presentation, APES+ Conference

Conference distinction and monetary prize ($75)

Dalhousie University Graduate Entrance Scholarship

Monetary allotment: $7,250 over 1 year

Dalhousie University Undergraduate Academic Renewable Entrance Scholarship

Monetary allotment: $16,000 over 4 years

CERTIFICATIONS

TCPS 2 CORE-2022 Certification, Government of Canada Panel on Research Ethics

Health Canada Division 5 – Drugs for Clinical Trials with Human Subjects, CITI

Transportation of Dangerous Good TDG/IATA, CITI

Good Clinical Practice, CITI

Cancer and Exercise Training for Health and Fitness Professionals Certification, THRIVE

PRESENTATIONS:

- American College of Sports Medicine (June 2022). Title: Lower Neurohemodynamic Transduction in Young Females Versus Males with Similar Aerobic Fitness and Sympathetic Outflow.
- Atlantic Province Exercise Scientists and Socio-Culturalists (March 2021). Title: The Impact of Aerobic Fitness on Sympathetic Neurohemodynamic Transduction in Older Males.
- Canadian Society for Exercise Physiology (October 2020). Title: Influence of prostaglandins and endothelial-derived hyperpolarizing factors on brachial and popliteal endothelial-dependent function in young adults.

PUBLICATIONS

Abstracts:

- **Petterson, J.L.**, O'Brien, M.W., Ramsay, D.J., Johnston, W., O'Neill, C.D., Dogra, S., Mekari, S., Flora, J.S., Kimmerly, D.S. (2021). Sympathetic Neurohemodynamic Transduction is Attenuated in Older Males Independent of Age-Related Declines in Aerobic Fitness. *Applied Physiology, Nutrition, and Metabolism,* 46(10), Suppl.2, S70
- **Petterson, J.L.**, O'Brien, M.W., Johns, J.A., Chiasson, J., Kimmerly, D.S. (2020). Influence of Prostaglandins and Endothelial-Derived Hyperpolarizing Factors on Brachial and Popliteal Endothelial-Dependent Function in Young Adults. *Applied Physiology, Nutrition, and Metabolism*, 45(11), Suppl.3, S315.

Peer Reviewed Publications:

- **Petterson, J.L.**, O'Brien, M.W., Ramsay, D.J., Johnston, W., O'Neill, C.D., Dogra, S., Mekari, S., Flora, J.S., Kimmerly, D.S. (2021). Sympathetic Neurohemodynamic Transduction is Attenuated in Older Males Independent of Aerobic Fitness, *Clinical Autonomic Research*, https://doi.org/10.1007/s10286-021-00845-y
- **Petterson, J.L.**, McPhee, B.N., Wu, Y., O'Brien, M.W. (2021). Does COVID-19 Influence the Sympathetic Regulation of Blood Pressure? *Journal of Physiology*, https://doi.org/10.1113/JP282362
- **Petterson, J.L.**, O'Brien, M.W., Johns, J.A., Chiasson, J., Kimmerly, D.S. (2021). Influence of Prostaglandins and Endothelial-Derived Hyperpolarizing Factors on Brachial and Popliteal Endothelial-Dependent Function in Young Adults, *Journal of Applied Physiology*, https://doi.org/10.1152/japplphysiol.00698.2020
- O'Brien, M.W., **Petterson, J.L.**, Kimmerly, D.S. (2022). Impact of Sampling Duration on Spontaneous Sympathetic Transduction, *Clinical Autonomic Research*, https://doi.org/10.1007/s10286-022-00861-6
- O'Brien, M.W., **Petterson, J.L.**, Johns, J.A., Mekary, S., Kimmerly, D.S. (2022). The Impact of Different Step Rate Threshold Methods on Physical Activity Intensity in Older Adults, *Gait & Posture*, https://doi.org/10.1016/j.gaitpost.2022.02.030

- O'Brien, M.W., **Petterson, J.L.**, Kimmerly, D.S. (2021). An Open-Source Program to Analyze Spontaneous Sympathetic Neurohemodynamic Transduction. *Journal of Neurophysiology*. https://doi.org/10.1152/jn.00002.2021
- O'Brien, M. W., **Petterson, J. L**., Johns, J.A., Mekary, S., Kimmerly, D.S. (2021). A Larger Low-Flow Mediated Constrictor Response is Associated with Augmented Flow-Mediated Dilation in the Popliteal Artery. *Clinical Physiology and Functional Imaging.* https://doi.org/10.1111/cpf.12725
- O'Brien, M.W., Johns, J.A., **Petterson, J.L.**, Mekary, S., Kimmerly, D.S. (2021). The Impact of Age and Sex on Brachial and Popliteal Artery Endothelial-Dependent Vasodilator and Vasoconstrictor Function, *Experimental Gerontology*. https://doi.org/10.1016/j.exger.2020.111221
- O'Brien, M.W., **Petterson, J.L.,** Wu, Y., Bray, N.W., Kimmerly, D.S. (2022). What is the Impact of Aerobic Fitness and Movement Interventions on Low-Flow-Mediated Vasoconstriction? A Systematic Review of Observational and Intervention Studies, *Vascular Medicine.* https://doi.org/10.1177/1358863x211073480
- O'Brien, M.W., Wu, Y., **Petterson, J.L.,** Bray, N.W., Kimmerly, D.S. (2022). Validity of the ActivPAL Monitor to Distinguish Postures: A Systematic Review. *Gait & Posture.* https://doi.org/10.1016/j.gaitpost.2022.03.002
- O'Brien, M.W., Wu, Y., **Petterson, J.L**., Frayne, R.J., Kimmerly, D.S. (2022). Ecological Validity of Prolonged Sitting Studies – How Well Do They Represent Real Life Sedentary Patterns? *Translational Journal of ACSM.* https://doi.org/10.1249/tjx.0000000000000182
- O'Brien, M.W., Al-Hinnawi, A., Wu, Y., **Petterson, J. L.**, Shivgulam, M. E., Johns, J. A., Frayne, R. J., Kimmerly, D. S. (2021). The Influence of Habitual Breaks in Sedentary Time on Cardiovagal Baroreflex Function. *Applied Physiology, Nutrition and Metabolism.* https://doi.org/10.1139/apnm-2021-0246
- O'Brien, M.W., Ramsay, D. J., O'Neill, C., **Petterson, J.L.,** Dogra, S., Mekary, S., Kimmerly, D.S. (2021). Aerobic Fitness is Inversely Related with Neurohemodynamic Transduction and Blood Pressure Variability in Older Adults. *GeroScience.* https:// doi.org/10.1007/s11357-021-00389-z.