METHODOLOGY IN PSYCHOPHYSIOLOGICAL STUDIES: APPLICATIONS IN PHYSICAL ACTIVITY

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ABSTRACT
Herein we describe the psychophysiological methods of electrodermal recording, electromyography, electroencephalography, impedance cardiography, and micro-neurography as each has been employed in physical activity related research. In our coverage of each method, we first establish a brief historical context, then review some essential guidelines for research, provide examples of the types of physical activity related research questions that can be addressed, and describe selected studies that have employed each method. Psychophysiological measures are valuable and offer advantages over other methods of assessing behavioral responses. Physiological responses are objective and can be measured continuously without distraction during an experimental task, can be obtained more quickly and reliably than detailed analyses of overt behavior, and analogous behaviors may be differentiated based on underlying patterns of neuromuscular, electrocortical, cardiographic, or muscle sympathetic nervous activity. Much progress in the fields of exercise and sport psychology is yet to be realized through the complementary employment of sound psychophysiological methods, self-evaluative judgments, and the observation of behavior.

Keywords: Physical Activity, Exercise, Psychophysiology, EMG, EEG, MSNA, Skin Conductance, Impedance Cardiography.

INTRODUCTION
In his quest to understand the impact of physical activity on psychological phenomena, Dr. William P. Morgan maintained that research studies should include the examination of self-evaluative judgments, overt behavior, and physiological responses (the three-systems approach; Lang, 1993). In this paper we will highlight the impact of Dr. Morgan’s approach to research in the fields of sport and exercise psychology, and will focus on selected psychophysiological methods that have been employed in our efforts to un-
understand central nervous system responses to physical activity. We will provide a brief introduction to the psychophysiological methods of skin conductance recording, electromyography (EMG), electroencephalography (EEG), impedance cardiography, and muscle sympathetic nerve activity (MSNA).

It is beyond the scope of this paper to provide a detailed account of every psychophysiological method. As such, we refer the interested reader to excellent reviews of each method. We will briefly discuss what each method measures, some essential guidelines for research, the types of physical activity-related questions that can be addressed, and examples of studies that have utilized each method.

**Advantages of Psychophysiological Methods**

There are several benefits of using psychophysiological methods over other methods of assessing behavioral responses, such as reaction time or subjective report. First, physiological responses can be measured continuously without distraction during an experimental task, and do not require the participant’s attention (Tassinary & Cacioppo, 2000). Second, physiological indices provide more sensitive detection and quantification of muscle tension, and can be obtained more quickly and reliably than detailed analyses of observable behavior or self-report (Cohn, Zlochower, Lien, and Kanade, 1999). Third, similar overt behaviors may be differentiated based on underlying patterns of neuromuscular, cardiographic, or muscle sympathetic nervous activity (Ghez, 1991). Fourth, some psychological processes are subtle and only occur with very fast or very small changes in physiological activity, without any visible movements (Tassinary & Cacioppo, 2000). Finally, physiological indices of psychological constructs are less affected by behavioral artifacts related to experimenter expectancies, such as the halo effect (Thorndike, 1920), the Rosenthal effect (Rosenthal, 1966), and demand characteristics (Orne, 1962), compared to subjective indices based on self-report, but are not immune to such artifacts. Psychophysiological studies, though they employ more objective measures, should strive to employ a rigorous experimental design, and include protections against experimenter expectancy effects, such as blinding the experimenter to the experimental condition of the participant (Morgan, 1997).

**Electrodermal Activity**

Electrodermal activity is one of the most widely studied response systems in the history of psychophysiology. The measurement of electrodermal activity was first demonstrated by Fere (1888), and was shown to be sensitive to sensory and emotional stimuli. The exosomatic method of recording electrodermal activity (also called the psychogalvanic reflex and the galvanic skin response) involves passing a small electric current between two electrodes attached to the skin. Skin conductance (the reciprocal of skin resistance) is typically measured on the palmer surface of the hand or fingers and is a measure of the ease with which an electric current passes through the skin. The principle of Ohm’s law (R = V/I), which states that resistance (R) is equal to the voltage (V) applied between two electrodes divided by the current (I) being passed through the skin, is applied in
the measurement of electrodermal activity. When the voltage is held constant, as is the case when a voltage is passed between the two sensors on the skin, then current flow can be measured, which will vary directly with the reciprocal of skin resistance (i.e., skin conductance in units of micro-Siemens). Among the most essential guidelines for research are to be certain that the voltage applied is calibrated and held constant, and that the surface area of recording is the always the same within and across participants (for review and research guidelines, see Dawson, Schell, & Filion, 2000).

The primary physiological mechanism for increased skin conductance is activation of the eccrine sweat glands, which are found all over the body and most densely on the palms of the hands and the soles of the feet, and are innervated cholinergically by the sympathetic nervous system with the primary purpose of thermoregulation (Dawson, Schell, & Filion, 2000). There has been, however, some uncertainty whether the eccrine sweat glands function solely to regulate heat transfer, and some have suggested the eccrine sweat glands on the Palmer and plantar surfaces may be involved in grasping behavior more than thermoregulation (Edelberg, 1972). Also, the eccrine sweat glands may not receive only sympathetic innervation, but may be supplied by parasympathetic nerve fibers as well (Shields, MacDowell, Fairchild, and Campbell, 1987).

A distinction can be made between skin conductance level (SCL), the tonic absolute level of conductance in the absence of a sensory stimulus, and phasic skin conductance responses (SCRs), which ride along the tonic levels and may vary moment to moment. A long history of psychophysiological research has shown that SCRs are sensitive to novel, unexpected, potentially important sensory stimuli, or the absence of an anticipated stimulus, and that this response habituates with repeated stimulation (Dawson, Schell, and Filion, 2000). Exercise can clearly have an effect on tonic levels of skin conductance, as sweat reduces the resistance to current flow between the sensors. For this reason, the examination of changes in skin conductance level during or after acute exercise requires careful consideration. Similarly, the measurement of SCRs after acute exercise may be confounded by the prior activation of the sweat glands by the exercise. However, if the effect of increased sweating on SCL is controlled, then an interpretation of SCRs may be possible.

Despite the methodological challenges that face exercise psychologists who choose to measure SCRs, their measurement can provide important information regarding psychological constructs, and there are important questions that can be addressed regarding the effects of physical activity on responses to motivationally relevant stimuli. Skin conductance responses can be evoked by a wide variety of stimuli characterized as novel, surprising, emotionally arousing, or motivationally significant. For this reason, the SCR is not clearly interpretable as reflecting any single psychological process (Landis, 1930). However, specificity regarding the meaning of a SCR can be inferred based on the rigor of the experimental design employed and the nature of the stimulus. For example, in studies of emotion using affective pictures as stimuli, larger SCRs occur in the context of viewing more arousing pleasant and unpleasant pictures compared to during the viewing of neutral pictures (Lang, 1995). It is not known if acute or chronic exercise alters this effect of emotional arousal on SCRs. The measurement of SCRs to exercise- or
activity-related stimuli (such as pictures or sounds) could be useful to characterize those who begin an exercise program, and whether these responses predict adherence or withdrawal.

There are many individual differences in electrodermal responding that have the potential of being modified by physical activity. For example, patients with schizophrenia show a bimodal distribution in skin conductance responding; some exhibit hypo-responsiveness and some exhibit hyper-responsiveness. Hypoactive SCR activity has been associated with emotional withdrawal and disorganized thought processes, and hyperactive SCR activity has been associated with psychotic episodes (Bernstein et al., 1982). Physical activity has been shown to improve the quality of life (Callaghan, 2004), and have therapeutic effects (Martinsen, 2000) in patients diagnosed with schizophrenia. The mechanisms of the physical activity-related improvements are not known. It is plausible that the activation of the sympathetic nervous system during exercise could provide a stimulus for adaptation in skin conductance responding. It remains to be determined if these adaptations could be associated with improvements in positive and negative symptoms of schizophrenia. Consistent with negative symptoms of schizophrenia, depressed patients also show a hypo-responsive electrodermal system. Exercise has been shown also as an effective treatment for clinical depression (Martinsen, Hoffart, & Solberg, 1989). The effects of exercise training on the sympathetic nervous system make it biologically plausible that afferents to the eccrine sweat glands may be affected, but it is not known to what extent normal electrodermal responding is restored after an exercise intervention and remission from depression.

An example of an experiment that examined skin conductance was conducted by Steptoe, Moses, Mathews & Edwards (1990). Groups of sedentary low fit and moderately fit adults were compared on skin conductance level during and after a visual problem-solving task. The groups did not significantly differ in SCL during the task, but the moderately fit showed a lower SCL during the recovery period. These groups were then randomly assigned to one of four conditions that lasted 10 weeks, four sessions per week: 30 minutes of moderate intensity walking/jogging (70-75% of maximum heart rate); 20 minutes of low intensity walking/jogging (60-65% of maximum heart rate); 20 minutes of discontinuous stretching exercise (below 50% of maximum heart rate); and a wait-list control group. Despite a significant change in fitness for only the moderate intensity exercise group, there were no differences post-training in skin conductance level between the exercise groups during or after the problem-solving task. However, those in the wait-list control group showed smaller changes in skin conductance during and after the task compared to the three experimental groups.

Despite the initial cross-sectional differences based on aerobic fitness, there was no effect of increased fitness on SCL during a mental stress task. Rather, a Hawthorne-like effect was evident, in that any of the treatments produced an increased SCL during the task. This finding, though counter intuitive to the hypothesis that physical fitness results in reduced reactivity to stress (Crews & Landers, 1987), is consistent with the notion of a greater capacity to respond sympathetically with increased physical activity (Dishman & Jackson, 2000). This study also bears on the effect Lacey & Lacey (1958) termed
individual response stereotypy, and highlights the importance of assessing multiple indices of autonomic function in order to fully understand how exercise affects reactions to stress, with the expectation that correlations within individuals between physiological systems will vary.

**ELECTROMYOGRAPHY**

The use of electromyography (EMG) can be dated back to the work of Carlo Matteucci in 1833 (Fulton, 1926), and in modern psychophysiological research in humans to the works of Edmund Jacobson in 1927 and 1932 (Tassinary & Cacioppo, 2000). The output of a surface EMG measurement reflects the difference in voltage detected between two electrodes in relation to a common ground or reference, usually expressed in microvolts. In contrast to the measurement of skin conductance, where a constant voltage is applied between two electrodes, EMG involves the passive detection on the skin surface of the voltage produced by muscular contraction. This voltage is the result of the depolarization of motoneurons, which release acetylcholine, and cause a transient excitatory potential resulting in the depolarization of the resting membrane potential of the muscle cell. A portion of the changes in the electromagnetic field associated with the calcium dependent interaction between actin and myosin passes through the extra-cellular fluid to the skin, and this comprises the majority of the surface EMG signal (Tassinary & Cacioppo, 2000). The surface EMG does not reflect the activity of a single motor unit or action potential, but rather is the summation of the activity of many motor units firing at different rates (Suzuki, Conwit, Stashuk, Santarsiero, & Metter, 2002).

Among the most important methodological considerations for EMG research are: 1) to employ a sampling rate at least twice that of the fastest EMG frequency of interest; 2) to employ a very precise analog-to-digital converter in order to detect weak muscular actions that may only occupy fractions of a microvolt (e.g., a 16-bit converter offers a resolution of 1 part in 65,536); 3) to employ an appropriate, and calibrated, integration time constant to rectified raw EMG signals (unless one performs spectral analysis of the raw EMG signal), which will vary with the specific muscle and the time course of the response of interest; 4) to calibrate the amplifier gain regularly; 5) to express the data in the appropriate units of measurement (which will depend on the type of integration performed); 6) to specify to time interval over which the EMG signal is averaged; and 7) to give careful consideration to the baseline EMG level, which physiologically represents the noise floor, but psychologically may be influenced by factors (e.g., anxiety, movement) that could increase EMG activity above the electrical silence typically exhibited by a muscle "at rest" (see Fridlund & Cacioppo (1986) for detailed research guidelines). Also, because the placement of any type of sensors on research participants can be psychologically intrusive and potentially anxiety provoking (Smith et al., 2002), the researcher should have both good technical and interpersonal skills, be well trained to interact with the research participants in a relaxed but straightforward and professional manner, establish "standard" conversation topics with participants, provide a comfortable room temperature and dim lighting, avoid unnecessary exposure
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to recording equipment, and refer to the equipment using neutral words such as "sensors" rather than "electrodes".

The measurement of EMG has been used in many areas of research relevant to physical activity, so our focus will be necessarily limited to questions related to psychological constructs. EMG measurements are appropriate in order to answer questions related to the effects of physical activity on muscular tension or relaxation, fatigue, muscle soreness, and perceived exertion (Lagally, Robertson, Gallagher, Goss, Jakicic, et al., 2002). In addition, researchers have utilized EMG in biofeedback studies to help control chronic pain (Flor & Birbaumer, 1993), and urinary incontinence (Hirsch, Weirauch, Steimer, Bihler, Peschers, et al., 1999).

One hypothesis regarding exercise-related effects on anxiety is that reduced muscular tension, or reductions in indices of motor neuron excitability, contributes to reduced subjective anxiety (e.g., Bulbulian & Darabos, 1986). Importantly however, these studies have assessed EMG in muscles with no direct link to brain systems that are related to anxiety or emotion (such as in the gastrocnemius, frontalis or temporalis muscles) (Canter, Kondo, & Knott, 1975), and/or have failed to assess concurrent reports of subjective anxiety at the time of EMG measurement (Bulbulian & Darabos, 1986). Recent EMG research employing reflex-probe paradigms involving muscles known to be influenced by brain systems of emotion have provided insight into the effects of acute exercise on the relationship between decreased in muscular tension and reports of state anxiety.

Two studies examined the acoustic startle eye blink reflex during the viewing of affective pictures in order to examine the effects of physical activity on emotional responsiveness. Smith and O’Connor (2003) examined the startle reflex (the eye blink component, known to be larger during the processing of an unpleasant stimulus, and smaller during the processing of a pleasant stimulus; see Lang, 1995 for an overview) while female participants simultaneously performed low intensity (40% of peak power output) cycling exercise and viewed pleasant, neutral, and unpleasant pictures from the International Affective Picture System (IAPS; NIMH Center for the Study of Emotion and Attention (CSEA), 1999). The magnitude of the startle reflex was not affected by light, non-painful exercise, and the effect of picture valence (i.e., larger startle magnitude during unpleasant compared to pleasant and neutral pictures) was similar during both the exercise and seated rest condition. In addition, EMG responses from the corrugator supercilii muscle (a measure of facial frowning) during the viewing of unpleasant pictures were not altered during exercise compared to during seated rest.

Smith, O’Connor, Crabbe, and Dishman (2002) examined the relationship between changes in mood after acute exercise (e.g., Morgan, 1985) and changes in emotional responsiveness during affective picture viewing indexed using the startle paradigm. Twenty-four healthy females completed 25 minutes of low (40% VO_{2peak}) and moderate (70% VO_{2peak}) intensity cycling exercise and seated rest. Startle and corrugator supercilii responses, as well as baseline corrugator supercilii EMG activity, were measured immediately before and 20 minutes after each condition while participants viewed pleasant, neutral, and unpleasant pictures. Both state anxiety and startle magnitude

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during each type of picture were significantly reduced 20 minutes after each condition. The fact that reductions in the magnitude of the startle reflex did not differ between pleasant, neutral, and unpleasant pictures suggests that the motivationally relevant reflex pathways in the brain known to modulate the startle reflex were relatively unaffected. However, there were significant exercise intensity-dependent decreases in baseline corrugator supercilii tension after exercise. Nevertheless, corrugator EMG responses during unpleasant pictures were not different between or after the exercise and rest conditions, even though baseline tension was lower after exercise. These findings suggest that anxiolytic conditions of low and moderate intensity cycling and seated rest are related to decreased startle amplitude but do not reflect changes in appetitive or defensive responsiveness, even in the presence of changes in muscular tension.

Motl & Dishman (2004) examined the relationships among acute exercise, anxiety, and the H-reflex by experimentally increasing and then decreasing anxiety and measuring the concurrent effects on the H-reflex. The soleus H-reflex and state anxiety were measured in 16 individuals whose anxiety was experimentally manipulated by consumption of a large dose of caffeine. The soleus H-reflex and state anxiety were measured before and 1 hour after consuming caffeine or placebo and then again 10 minutes after 30 minutes of cycling at 60% VO$_{2peak}$ or quiet rest. The results indicated that caffeine consumption did not influence the amplitude of the soleus H-reflex, but it did increase state anxiety. However, acute exercise reduced the soleus H-reflex after consumption of either caffeine or placebo. Moreover, exercise reduced state anxiety only after consumption of caffeine; and there was no evidence of a relationship between changes in the soleus H-reflex and state anxiety. These findings provide no evidence that increased or decreased anxiety were associated with concurrent changes in the soleus H-reflex.

**Electroencephalography**

Hans Berger first demonstrated the measurement of brain electrical activity in humans about 75 years ago (Berger, 1929). Since that time there have been rapid advances in the technology for recording electroencephalographic (EEG) signals, and in the analysis of the vast amounts of EEG data that can be acquired in a relatively short amount of time. Based on comparison of intracellular and scalp recordings, most researchers agree that ongoing EEG measured at the scalp reflects the summation of post-synaptic potentials, not summated action potentials, and that glial cells contribute very little to the variance in the EEG signal (Davidson, Jackson, & Larson, 2000). The EEG signal can be described by its amplitude (in the range of microvolts) and frequency.

The amplitude of the EEG signal can be measured across different time periods, and also in response to a discrete event (such as an auditory or visual stimulus) as an event related potential (ERP; see Picton, Bentin, Berg, Donchin, Hillyard, et al., 2000, and Fabiani, Gratton, & Coles, 2000, for research guidelines). The frequency of the change in positive and negative going voltage has been characterized in adults as the delta (0-4 Hz), theta (5-7 Hz), alpha (8-12 Hz), beta (13-20 or 30 Hz), and gamma (36-44 Hz) bands. More recently, researchers have divided the alpha and beta bands
to examine the higher and lower frequency ranges within each of the traditional bands. The power in the different frequency components within a time epoch (selected after artifact removal with a Hanning or Hamming window, a cosine taper function designed to minimize the impact of spurious frequencies at the edge of the epoch) are determined most commonly by a fast Fourier transformation, and can expressed in squared-microvolts per unit frequency. Detailed guidelines for recording and analyzing EEG data can be found in Pivik, Broughton, Coppola, Davidson, Fox, and Nuwer (1993).

EEG has been employed by psychophysiologists to study a wide-range of processes related to brain activation, such as cognition, sleep, emotional reactivity, and psychopathology (Fabiani, Gratton, & Coles, 2000; Davidson, Jackson, & Larson, 2000). In the field of exercise psychology, the focus of most research has been on the examination of brain electrocortical activity related to cognitive function and to changes in mood. In research related to cognition and aging, Hillman and colleagues have measured ERPs during a cognitive decision making task and have reported a pattern of results indicating that being physically active may help to attenuate the natural decline in cognitive executive control with age, as measured by P3 amplitude and latency (Hillman, Belopolsky, Snook, Kramer, & McAuley, 2004).

Among the EEG experiments that have been conducted on the relationship between acute exercise and mood, much attention has been paid to the alpha frequency band. Previous work has shown that the increased power in the alpha frequency band (reflecting decreased cortical activation indicative of a relaxed state) was associated with decreases in self-rated anxiety (Petruzzello & Landers, 1994), and that hemispheric asymmetry in alpha power in the frontal lobes before acute exercise (reflecting a greater left frontal activation and a hypothesized disposition to respond more positively to pleasant stimuli) predicted decreased anxiety after exercise (Petruzzello & Tate, 1997). However, in a statistical synthesis of the effects of acute exercise on EEG, it was found that alpha power increased in other recording sites after exercise, and that across the literature, changes were not confined to the frontal lobes, or to the alpha frequencies (Crabbe & Dishman, 2004). In fact, exercise has been shown to globally increase cortical activity during and immediately after a single exercise session. Whether or not the changes in EEG during and after exercise reflect changes in the motivational systems that govern affective responses remains to be elucidated.

In a recent study to examine whether acute exercise affects reactions to emotion eliciting stimuli, EEG responses and self-rated valence and emotional arousal were measured during the viewing of pleasant, neutral and unpleasant pictures from the International Affective Picture System before and after subjects exercised or sat at rest for 30 minutes (Crabbe, Smith & Dishman, in review). Compared to after rest, self-reports of arousal during the viewing of unpleasant pictures decreased after exercise, but self-reports of valence and frontal alpha power were generally unchanged. The change in rated arousal was also accompanied by a shift from right to left dominance in the beta frequency bands in the parietal cortex. This change in EEG power reflects a decrease in right parietal activation. In previous work, right parietal and temporal activation has
been associated with increased emotional arousal (Nitschke, Heller, Palmieri, & Miller, 1999). Taken together, the changes in EEG after exercise indicate that exercise does not compromise the neural systems that respond to the motivational valence of a stimulus (as would be required to promote survival, such as escape from a predator), but may reduce emotional arousal to unpleasant stimuli. It remains to be determined if reductions in emotional arousal are at the source of the well-documented changes in mood after exercise (Smith et al., 2002).

**IMPEDANCE CARDIOGRAPHY**

The use of electrical impedance methods to study cardiovascular function dates back to work by Geddes and Baker (1968) and Nyboer (1970) who determined that changes in thoracic impedance could be due to several physiological processes including ventilation and cardiopulmonary blood flow. Following acceptance as a reliable and valid measure of cardiac function, impedance cardiography (ICG) has gained strong psychophysiological appeal. ICG provides a sensitive, non-invasive and continuous measure of autonomic nervous system activity and therefore is a potentially powerful method to study a host of disease-related problems consequent of a sedentary lifestyle. For the field of sport and exercise psychology, ICG provides a valuable method to determine some of the physiological bases that underlie psychological behaviors such as emotion and perception.

ICG is a measure of the mechanical function of the heart and is used to measure stroke volume and systolic time intervals (for detailed reviews and research guidelines see Miller and Horvath, 1978; Sherwood et al., 1990). Electrodes (“spot” or “band” configurations) that are placed in specific configurations along the neck and thorax of the body are used to measure the changes in electrical impedance that occur at rest or during a challenge (e.g., exercise or mental stress). In conjunction with heart rate (for measurement guidelines see Jennings et al. 1981), measures of cardiac output (product of heart rate and stroke volume) can be derived, and the addition of blood pressure (for measurement guidelines see Shapiro et al. 1996) can provide a more complete picture of autonomic nervous system activity. Because ICG is an index of myocardial contractility, time dependent measures of pre-ejection period, left-ventricular ejection time and estimates of contractility (the Heather Index) can also be derived. These variables can then be used to determine how the physiological properties of the cardiovascular system relate to central nervous system processes that underlie behavior.

Impedance cardiography is based on properties of an electrical current, specifically the relationship between voltage and resistance and the known properties of blood as a conductor. Succinctly stated by Sherwood and colleagues (1990), “Impedance cardiograph systems induce a constant magnitude, alternating current field along the thorax, measure impedance (Z) changes occurring with each heart beat (ΔZ) and provide an output voltage that can be interpreted as reflecting stroke volume.” A formula is used to calculate SV, the most popular of which is the Kubicek formula (Kubicek et al., 1966):
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SV = \rho_b(L/Z_0)^2 \cdot \text{LVET} \cdot \frac{\text{d}Z}{\text{d}t_{\text{max}}}

Where SV is stroke volume (ml), \rho_b represents the resistance of blood (ohm·cm), L is a measure of the distance between the recording electrodes (cm), Z_0 is a measure of baseline impedance that occurs between the recording electrodes, LVET is a measure of the estimated left-ventricular ejection time (seconds) and \frac{\text{d}Z}{\text{d}t_{\text{max}}} represents the maximum slope of the impedance waveform per beat (ohm/second). It has been shown that changes in impedance (ΔZ) vary along with changes in left-ventricular (aortic) flow and thus reflects changes in SV.

There are several commercially available units for ICG data collection and one can be trained in the placement of electrodes. However, rigor in the placement of the electrodes, consistency of preparation within and between subjects and careful calibration of the device are critical for accurate signal collection and, ultimately, collection of interpretable data following experimental manipulation (Sherwood et al., 1990). The general equipment necessary for impedance cardiography includes commercially available Mylar “band” electrodes or standard electrocardiogram (ECG) electrodes, an impedance cardiograph system that utilizes a constant (1-4 mA) and an alternating current (100-200 kHz) and an amplifier. The use of “band” or “spot” (standard ECG) electrode configurations (see Sherwood et al., 1990) depends on the research question and the population being tested. If determination of SV is of primary interest tetrapolar “band” electrode configurations are preferable. If systolic time intervals are of primary interest, spot electrode configurations provide a fast and less invasive option. In addition to the impedance signal, measurements of the ECG and phonocardiogram (PCG) waveforms are essential for scoring the ICG signal. Major scoring points of the ICG signal include 1) the ECG Q-wave that denotes the onset of electromechanical systole on the ICG waveform, 2) the \frac{\text{d}Z}{\text{d}t} B-point corresponding to the rapid upslope of the ICG waveform and that denotes the onset of left-ventricular ejection, 3) \frac{\text{d}Z}{\text{d}t} \text{max} representing the peak value on the \frac{\text{d}Z}{\text{d}t} signal and corresponding to peak blood flow in the aorta and 4) the X-point that corresponds to the lowest point of the ICG waveform and denotes the end of left-ventricular ejection. These parameters provide the necessary information, in conjunction with the mathematical formula, to determine SV and several systolic time intervals.

As a research tool, ICG has been integral to our understanding of hemodynamic responses related to stress, fitness, personality, mood, gender, and risk for developing diseases of the cardiovascular system. In general, it has been shown that autonomic nervous system responses to stress are dependent on the degree of perceived control, the effort required for a given task and the appetitive or aversive nature of the stressor (Quigley et al., 2002; Weinstein et al., 2002). Fitness and gender have been demonstrated as important moderators of cardiovascular reactivity to stress with fit women, but not men, showing blunted cardiovascular responses to physical stress (Jackson and Dishman, 2002; Dishman et al., 2003). Normotensive individuals with a genetic risk for developing hypertension show augmented cardiovascular responses to stress in the form of enhanced reactivity, slow adaptation and delayed recovery for
pre-ejection period (Schneider et al., 2003) suggesting that non-invasive measures of hemodynamic function could be used as early warning signals for the development of essential hypertension. Mood has been shown to be related to measures of resting hemodynamic function with negative mood states being associated with lower stroke volume and cardiac output (Peckerman et al., 2003; Yu et al., 2001). Importantly, exercise is associated with improved mood (e.g., Martinsen et al., 1989) and exercise training has been demonstrated to improve cardiovascular responses to mental and physical stress (Claytor, 1991; Georgiades et al., 2000; Jackson and Dishman, 2002). Future research exploring the links between cardiovascular responses to stress, mood and exercise is necessary to determine whether improvements in mood following exercise are associated with improved hemodynamic function.

Jackson and Dishman (2002) used ICG, blood pressure and blood flow responses to determine the hemodynamic pattern of stress reactivity to physical (cold pressor) and mental (arithmetic) stress in normotensive black American women with and without a positive family history of hypertension. Participants also completed a maximal exercise test to determine cardiorespiratory fitness. Independent effects of fitness on several hemodynamic variables were reported. In general fitness was associated with enhanced total peripheral resistance (TPR) to both physical and mental stress and blunted systolic blood pressure increases and enhanced recovery of systolic blood pressure and mean arterial pressure to mental stress. The association between enhanced vascular (TPR and SV) recovery from stress was stronger for fit women with a negative family history of hypertension suggesting that the relationship between fitness and vascular reactivity may be further influenced by genetic risk. These results highlight the utility of comprehensively determining hemodynamic responses while concomitantly determining fitness and genetic risk factors when striving to understand mechanisms of cardiovascular disease. Future research utilizing ICG methods during exercise while obtaining perceptual measures of pain and exertion may help determine whether hemodynamic factors play a role in the decreased muscle pain during exercise that is experienced by black American women with a positive family history of hypertension (Cook et al., 2004).

Impedance cardiography has been used to study cardiac function in patients' with chronic fatigue syndrome (CFS); an illness characterized by debilitating symptoms of fatigue and extreme sedentary behavior (Fukuda et al., 1994). Based on the premise that symptoms of fatigue in CFS may be a consequence of impaired regulation of circulation to organs and skeletal muscle, Peckerman et al. (2003) compared ICG derived stroke volumes in CFS patients (n = 38) and healthy sedentary controls (n = 27) during conditions of quiet supine rest and quiet standing. Patients were stratified as having severe CFS (1988 case definition plus 7 symptoms rated as at least “substantial”) and less severe CFS (1994 case definition) to determine whether measures of stroke volume differed as a function of illness severity. Self-reported symptoms of energy, fatigue and depression were obtained to explore the relationship between cardiac output (product of SV and heart rate) and symptoms of CFS. Severe CFS patients were found to have lower resting SV than both the less severe CFS patients and healthy sedentary controls and exhibited
a smaller decline in SV during quiet standing than controls. Mean arterial pressure and heart rate increased during standing for all groups and were not significantly different. Lower cardiac output was significantly associated with greater self reported post-exertion fatigue and fever-chills and lower ratings of memory and concentration problems. These results show that patients who rate their illness as severe have lower cardiac output compared to patients with a less severe rating and controls, and suggest that symptoms in patients with severe CFS may be due circulatory insufficiency to vital organs and exercising muscles. ICG may provide a valuable method to test potential cardiovascular mechanisms that might underlie the reported elevations in perceived exertion during exercise for CFS (Cook et al., 2003). In addition, exercise training has the potential to improve both circulatory and affective problems in CFS patients. Matching subjects on fitness and physical activity level would be an important next step in research aimed at understanding the relationship between symptoms and cardiac output in CFS.

**Muscle Sympathetic Nerve Activity**

Microneurography is a technique used to directly measure sympathetic nerve activity to human skeletal muscle and skin. The procedure is invasive and requires the insertion of micro-electrodes percutaneously into peripheral nerves for the recording of action potentials from single or multi-unit fiber fascicles (for detailed reviews see; Ray & Mark, 1994; Seals & Victor, 1991; Wallin, 1994). The technique was developed by Hagbarth and Valbo (1968) and has been primarily used by exercise scientists to study sympathetic nervous system responses to exercise. However, the method has gained recent interest from exercise scientists interested in understanding the complex interaction between behaviors, physical activity and the sympathetic nervous system.

Sympathetic nerve activity measured by microneurography represents efferent sympathetic vasoconstrictor activity and reflects post-ganglionic activation of type III and IV afferent fibers (Wallin, 1994). Muscle sympathetic nerve activity (MSNA) in particular is an important regulator of blood flow and blood pressure and can be recorded during exercise to directly measure real-time changes in nerve activity. The primary afferent stimulus for MSNA has been termed the “exercise pressor” or “muscle metabo-” reflex and involves the accumulation of metabolites within the vasculature of the contracting muscle (Mitchell and Schmidt, 1996). Once accumulated, these biochemicals stimulate type III and IV afferent fibers, thereby sending a single to supraspinal systems involved in cardiorespiratory regulation. Several biochemicals are capable of stimulating the afferent fibers, but few have been systematically examined in relation to MSNA.

Measurement of MSNA is usually made from one or more of four nerves, the peroneal, tibial, median and radial, although activity from any peripheral nerve could conceivably be recorded. The procedures for measurement of MSNA require specialized equipment and extensive training. The equipment used includes the following:

- Tungsten micro-electrodes that are 30-40mm in length and 0.2 mm wide. The exposed tip of the needle electrode is 1 to 5 µM and is used to both stimulate and record from the innervated nerve.
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- A reference needle electrode that is placed ~2cm from the recording electrode.
- A nerve stimulator and isolation unit for electrode placement.
- A preamplifier and a loudspeaker for burst amplification and identification.
- A band-pass filter (700-2,000 Hz) and resistance-capacitance integrator (time constant 0.1 sec) for mean voltage display.

Identification of muscle nerve innervation is confirmed by several criteria being met based on known MSNA characteristics (Ray et al., 1994; Seals and Victor, 1991). The main criteria are that 1) innervation of the nerve by the electrode elicits a muscle twitch without the experience of paresthesias (tingling or pins-and-needles sensation); 2) activity is elicited by tapping or stretching the muscle or tendon; 3) activity is not elicited by light brushing of the overlying skin; 4) the Valsalva (forced breath-hold) maneuver elicits an increase in activity; 5) a sudden arousal stimulus (yell or clap) does not elicit an increase in sympathetic activity; and 6) the bursts are characterized by a spontaneous pulse-synchronous and irregular pattern. The integrated MSNA signal is scored from a mean voltage neurogram and the data are expressed in terms of burst frequency, burst amplitude and total integrated activity (burst amplitude per unit time X burst frequency per unit time). Concomitant measures of heart rate, blood pressure and respiration are standard and many studies have also included measures of blood flow to determine vascular resistance.

Although burst occurrence is variable, MSNA is pulse synchronous with the cardiac cycle and influenced by arterial baroreceptor activity. Sympathetic bursts occur during diastole when the baroreceptors are unloaded. MSNA also exhibits an independent respiratory rhythm, with increased frequency during expiration compared to during inspiration. Increases in MSNA during exercise are dependent on several factors including the mode, intensity and duration of the exercise, the degree of muscle fatigue, the size of the contracting muscle, the physical environment, body posture, and physical training status (Wallin, 1994; Ray and Mark, 1994; Seals and Victor, 1991).

Aside from basic questions regarding sympathetic adjustments to exercise, MSNA has been used to study several behaviors and conditions that are associated with physical activity. Sympathetic nervous system function has been assessed using MSNA in chronic disease conditions that are associated with sedentary behaviors such as heart disease, obesity, and hypertension (Elam et al., 2003; Fagius, 2003; Floras, 2003; Mary and Stoker, 2003; Monroe et al., 2000; Yucha, 2000). The relationships between MSNA and psychosocial variables such as race and gender also have been examined (Calhoun and Mutenga, 1997; Dishman et al., 2003; Ettinger et al., 1996; Ray, 2000; Ray and Monahan, 2002; Vozarova et al., 2003). Mental and physical stressors have been used to help determine sympathetic adjustments to stress and MSNA has been used to aid in the understanding of perceptual variables such as pain and fatigue during exercise (Carter et al., 2004; Cook et al., 2000; Dishman et al., 2003; Lambert et al., 2002; Middlekauff et al., 2001; Saito et al., 1989). Insights gained from these investigations are relevant for understanding issues related to exercise adherence, mechanisms of pain perception, and risk factors associated with diseases of the cardiorespiratory system.
Thus, MSNA is a useful psychobiological tool to examine the relationship between the physiology of sympathetic nervous system regulation and the psychology of behavior.

The relationship between MSNA and physical activity has been addressed in a number of investigations (for review see: Ray and Hume, 1998). In general, both cross-sectional and longitudinal data suggest that resting MSNA does not appear to be a function of exercise training status. The lone exception appears to be in bodybuilders. Sinoway et al. (1992) reported that bodybuilders had significantly higher resting levels of MSNA than sedentary subjects suggesting that chronic and intense resistance training may alter resting sympathetic nervous system activity to muscle. Contrary to MSNA at rest, MSNA responses to exercise are reduced following exercise training. The training adaptations are specific to the exercising muscle, thus implicating local metabolic adaptations as the primary mechanism. Additionally, the adaptations appear to be specific to the mode of exercise and do not generalize to other types of non-exercise stressors (e.g., cold pressor, lower body negative pressure or head-up tilt).

MSNA has been used to help understand risk factors associated with development of hypertension. In an extension of their work on cardiorespiratory fitness and blood pressure reactivity, Dishman and colleagues (2003) used MSNA to examine the effect of mental and physical stressors on blood pressure and sympathetic nerve responses among normotensive men and women who varied in level of fitness. Twenty-eight (n = 14 women; n = 14 men) healthy normotensive participants underwent fitness testing and measurement of cardiovascular responses to the hand cold pressor and mental arithmetic tests. For the hand cold pressor test, participants submerged their right hand into a bucket of ice water (2°C) for two minutes. For the mental arithmetic test, participants performed serial subtraction of the number 13 from a four-digit start value. During testing, measures of blood pressure (finger plethysmography), heart rate (ECG), respiration, calf blood flow (venous occlusion plethysmography) and MSNA (peroneal nerve) were obtained. Additionally, measures of trait anger and anxiety, and ratings of aversion and discomfort were obtained to determine associations between emotional traits and subjective feelings on the physiological measures. Consistent with their previous findings, cardiorespiratory fitness was inversely related to systolic and diastolic blood pressures and MSNA responses to the hand cold pressor stimulus in women but not men. Fit women displayed a blunted blood pressure and muscle sympathetic nerve response during the hand cold pressor task compared to less fit women. The authors concluded that the pattern of responses were consistent with a decreased central sympathetic signal in fit women and that decreased sympathetic outflow is a potential mechanism for the observed blunting of blood pressure responses during the cold pressor task. Further, the results suggest that cardiorespiratory fitness may be an important modifier of risk factors associated with the development of hypertension among women.

MSNA has also been used to help determine potential mechanisms of the moderate to intense muscle pain that is often experienced during exercise (Cook et al., 1997). Cook et al. (2000), in a double blind, placebo controlled study, determined the influence of an opioid agonist (codeine) and antagonist (naltrexone) on the perception of forearm muscle pain and MSNA during exercise. The study was based on the rationale that the
type III and IV afferent fibers responsible for stimulation of the exercise pressor reflex are also involved in transmitting nociceptive information. Thus, a portion of MSNA to exercise could potentially be due to stimulation of nociceptors on primary afferent fibers from muscle. Twelve healthy and pain-free college aged males completed three graded, dynamic handgrip exercise tests to fatigue on separate days. Prior to each exercise test the participants received either 60 mg codeine, 50mg naltrexone, or placebo in identical capsules. The administration of the capsules was double-blind and the order was randomized and counterbalanced. MSNA was measured continuously from the peroneal nerve at rest, during exercise and during recovery. Forearm muscle pain (0 to 10 scale) and perceived exertion (0 to 10 & 6 to 20 scales) were measured during the last fifteen seconds of each exercise stage. Results demonstrated that neither codeine nor naltrexone altered the perception of muscle pain during exercise or recovery and that neither drug had an influence on MSNA in terms of burst frequency or total integrated activity (see Figure 1). The results suggest that endogenous opioid system, specifically the mu receptor, does not play a primary role in the perception of naturally occurring muscle pain during exercise and does not modulate sympathetic nerve activity to muscle during maximal handgrip exercise.

Figure 1. No effect of either codeine, naltrexone or placebo on pain perception (top) or MSNA burst frequency (bottom) during fatiguing handgrip. Data are expressed as a function of percent peak work (J) and are mean ± SE (n = 12).
SUMMARY & FUTURE DIRECTIONS

Psychophysiological indices, along with self-evaluative judgments, provide important complementary information regarding an individual's psychological state. Skin conductance provides an index of sympathetic arousal that is sensitive to novelty and emotional intensity. Electromyography, particularly in the facial muscles known to be related to facial expressions of emotion, provides indices of emotional reactivity. The acoustic startle reflex is known to be directly influenced by the output of subcortical brain systems that govern emotion, such as the amygdala. Electroencephalography provides information regarding the amplitude and frequency of electrical oscillations in the cerebral cortex with fine temporal resolution. Recent developments with high-density electrode arrays (e.g., 129- & 256-channel arrays) provide a powerful tool to examine brain mechanisms related to a host of psychological phenomena associated with acute and chronic physical activity in humans. Recent developments in analytical techniques permit estimates of the source of cortical dipoles that explain the distribution of voltage recorded on the scalp, as well as information regarding the distribution of different EEG frequencies across time and space (Gevins, Smith, McEvoy, Leong, & Le, 1999).

Impedance cardiography provides a fast and non-invasive index of autonomic nervous system activity that is sensitive to subtle changes in behavior and can provide important information relating hemodynamic function to several areas of physical activity research including stress and fitness, exercise and emotion, and physical activity and disease. Muscle sympathetic nerve activity is a powerful tool for determining sympathetic adjustments that accompany a physically active lifestyle as well as those that occur during an acute bout of exercise. Although classically employed as a physiological tool, recent interest in MSNA as a psychophysiological measure highlights the potential for future research aimed at determining the interaction between physiology and psychology as they relate to physical activity behaviors.

It is important that these methods be employed with technical rigor, not only among the healthy young adult, but more importantly among children, youth, older adults, and clinical groups of all ages. Future progress in the fields of exercise and sport psychology will require multi-disciplinary efforts, spanning the expertise of electrical engineers, computer programmers, physiologists, clinical and experimental psychologists, neurophysiologists, neuroscientists, exercise physiologists, biomechanists, and epidemiologists. These worthwhile pursuits represent a lasting tribute to Professor Morgan's vision of the fields of sport and exercise psychology.

REFERENCES


Methodology in Psychophysiological Studies: Applications in Physical Activity


