Motor Variability and Gait Complexity Responses to Load Carriage and Imposed Locomotion Patterns

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Military occupational tasks are often characterized by the addition of load carriage. Further, during loaded gait tasks, warfighters are required to maintain a walking gait despite ambulating at velocities individuals would normally jog at. The latter locomotion pattern is colloquially known as forced-marching. Not surprisingly, warfighters incur a high incidence of lower extremity musculoskeletal injuries (MSI). Load carriage has been implicated as a primary culprit in the incidence of MSI. However, it remains unclear what effect load carriage and forced-marching have on motor control. **PURPOSE:** The primary purpose was to determine the interactive effects of load magnitude and locomotion patterns have on motor control. The secondary and tertiary purposes were to determine the influence of sex and task completion ability on motor control respectively. **METHODS:** Twenty-six recruit aged men and women executed 10-minute trials of running and forced-marching with no load (BW) and with an additional 45% (+45%BW) and 55% (+55%BW) of body weight separately. Trials were executed at a velocity 10% above gait transition velocity. 3x2 (load*locomotion) RMANOVA were conducted on each outcome separately. Significance set at α=.5. **RESULTS:** As load increased, relative variability increased independent of locomotion pattern. Forced-marching exhibited more relative variability than running. Stride length and stride time exhibited long-range correlation and pink noise fractal dimension regardless of condition. By contrast stride speed exhibited stochasticity (white noise). Women had greater long-range correlation and lower fractal dimension compared to men. Non-completers only had significantly greater stride length variance compared to completers.

DISCUSSION: Healthy recruit individuals were able to adapt load carriage and forced-marching by regulating the coupling parameter (stride speed) more strictly. Motor variability was expanded in the null space with structured temporal variance to achieve a prescribed velocity. The expanded range of motor variability may come at a penalty of various cost functions such as kinetic and postural stability costs. As a result, prolonged execution of forced-marching with load may exacerbate MSI risk. Further in a dimensionally rich 'natural' setting, an individual may not be able to optimize motor control to handle load carriage.

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Preface

"The most intense conflicts, if overcome, leave behind a sense of security and calm that is not easily disturbed. It is just these intense conflicts and their conflagration which are needed to produce valuable and lasting results."

-Carl Jung

The pursuit of a PhD was not one I had ever envisioned for myself when I was younger, and it has been a circuitous journey to this point. The last four years were fraught with difficulty, burdened by sacrifice and plagued by self-doubt. But significant achievements are rarely accomplished alone. Honest introspection requires the humility to recognize the important contribution others have made in one's own success. Importantly, thanks must be given, and I hope these words serve as a sincere token of my appreciation and gratitude.

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Lastly and most importantly I want to thank my wife, Katie. You have been my biggest supporter since day one and sacrificed much for it. Your own success, work ethic, ambition and integrity have served as a great motivation for me to be better. You were always there to listen to me vent and tell me what I needed to hear to keep going. Without you my life would be on a very different path altogether. Thank you for all the support and everything you have done for me!

1.0 Introduction

Warfighter (i.e., military personnel in combat related roles) readiness, lethality and resilience is a continued priority of the United States (U.S.) Armed forces as evidenced by the recently enacted "Military Readiness and Injury Prevention Act of 2019" (S.1860) and the Holistic Health and Fitness (H2F) action. Both initiatives define the optimal warfighter as one who is healthy enough to operate on short notice, resilient enough to overcome external stressors while maintaining performance, and robust against musculoskeletal injury (MSI). Important to the success of the warfighter is the execution of occupational tasks. A major component of tactical training and operations for the warfighter are tasks involving load carriage, with magnitudes of load carried increasing substantially over the last decade[1-4]. Deployed warfighters spend considerable amounts of time wearing some type of load carriage, upwards of 52 hours/week while conducting military operations[5]. Furthermore, of the 52 hours/week, as much as 32 hours a week can be spent performing dismounted patrols where soldiers are ambulating load carriage of smaller magnitudes[5].

Load carriage refers to any additional mass that is carried by an individual and represents a perturbation to the locomotor system. In military populations it is typically composed of personal protective equipment (PPE) [i.e., body armor and helmet], weapons/munitions (small arms holstered to the waist, legs and chest, while rifles are slung on the shoulder or held) and/or rucksacks (large reinforced backpacks capable of holding sizable loads)[6]. As load carriage technologically has advanced the load magnitude commonly used has increased[1-4]. Load magnitudes are dictated by mission requirements and range from 20-60kg[6], with 20kg representing a 'combat load' that consists of PPE and weapons/munitions, while 60kg represent approach marches that include rucksacks as well[5, 6]. The load magnitude ranges are absolute and defined solely by mission requirements and not altered based on the size of an individual[7, 8]. Smaller load magnitudes such as ‘combat loads’ are often situated anterior-posterior, with the load distributed as evenly as possible (about 60:40 posterior-anterior). Greater load magnitudes require the use of rucksacks and therefore position the load more posteriorly to the warfighter[7]. Although load magnitudes of 20-60kg may be substantial for some, warfighters are still expected to execute a range of physical tasks while under load.

Warfighters typically perform load carriage ambulation tasks at velocities of 0.67-1.48m/s for long periods of time to traverse large distances[7]. During enemy engagements, regardless of load magnitude warfighters will be required to find cover and advance position thus requiring them to run with load carriage. In addition, ‘ditching’ of rucksacks during combat engagements, is common practice, where the warfighters drop the large rucksack which contains equipment not necessary for the immediate ‘engagement’, while retaining their combat load. As such running locomotion patterns are rarely used for sustained periods of time, but rather short bursts. When ‘mission’ priorities dictate, and time is a factor during approach marches, marching velocities (upwards of 2.28m/s) can increase beyond the point of gait transition velocity where individuals typically transition from a walking to running locomotion pattern[7], while carrying heavier loads. However, warfighters are encouraged to maintain a walking locomotion pattern known as forced-marching. Not surprisingly, an occupation characterized by arduous physical tasks (i.e., substantial load carriage and prolonged, high velocity ambulation) also experience a high incidence of MSI, even outside of theaters of operation[9-11].

Despite many advances in injury prevention research, MSI remain a serious threat to warfighter operational readiness and impose a significant burden on individuals in combat roles[9-

11]. Currently, MSI impose a financial burden on military populations leading to health care cost estimates between \$110 million-\$700 million each year [9, 10, 12, 13], 40% of which is attributed to outpatient care[9]. A stark example of the impact of MSI comes from data from a group of only 302 recruits attending basic training, which resulted in \$1.4 million in healthcare cost due to MSI alone[14]. Importantly, 21-31% of all observed MSI during basic training are attributed to load carriage activities for new recruits[12, 15]. Likewise, loaded road marching accounts for a significant portion of observed MSI in the entire U.S. Armed forces[15, 16]. The latter findings suggest that load carriage during bipedal ambulatory tasks is one of the main culprits to the development of MSI in warfighters. Therefore, load carriage as a perturbation on bipedal ambulation must be better understood to determine its effect on the locomotor system when executing bipedal ambulatory tasks. But firstly, the problem of MSI in the military must be clearly defined, culprits identified, injury perspectives explored and lastly the current state of the science regarding culprits addressed.

1.1 Definition of the Problem

In the United States (US) Military across all branches (Army, Navy, Air Force and Marines), 5% of all hospitalizations are due to MSI with ~2% for lumbar spine and ~28% for lower extremity (LE)[10]. 'Noncombat' MSI (not the result of ballistic weapons, improvised explosive devices [IED], parachuting or vehicular accidents [i.e., helicopter crashes]) are the leading cause for outpatient medical care in the U.S. Army of active duty personnel with two million encounters annually[16]. 'Noncombat' designated MSI account for 80% of all observed MSI[16, 17], 50-75% being lower body (includes lumbar/sacral spine, pelvis and lower extremity) MSI[9-12], with the lumbar spine (~18%) and knee (~17%) representing the most common sites for MSI occurrence[10, 14]. In addition, the hip, ankle and foot account for 14%, 12% and 12% of all lower body MSI respectively[9]. The majority (~85%)[9, 18, 19] of the injuries suffered are categorized as mechanical; ~75% due to cumulative microtrauma (repetitive mechanical stress) and ~10% as non-contact acute trauma (non-ballistic)[9, 10, 12, 18, 19]. Injuries observed range from stress fractures[9-12, 20], sprains/strains[9-12, 20] and even soft tissue degenerative diseases like osteoarthritis[21-23]. Furthermore, of all secondary MSI; ~28% are classified as spinal and ~23% are classified as lower extremity soft tissue degeneration [19]. Importantly, 60% of limited duty days and 65% of non-deployable soldiers has been attributed to 'noncombat' related MSI[16, 17]. While deployed, in theater, 30% of medical evacuations are the result of a noncombat related MSI[16]. Moreover, 85% of those medically evacuated soldiers are never re-deployed[16] reducing operational effectiveness of deployed units. Furthermore, ~70% of medical discharges and ~90% of first-year enlisted service disability discharges were due to MSI.

'Noncombat' MSI are not just limited to soldiers on deployment as many observed MSI are sustained during basic training[9, 12, 14]. An epidemiological study by Lovalekar et al (2020)[14] observed 78% and 66% of all MSI during physical training for female and male Marines respectively. Among new recruits in basic combat training, 61 MSI per 100 recruits were observed, with recruits on average suffering two separate MSI[9]. Indeed, even elite military training observed ~25% of all dropouts were due to MSI[24]. Additionally, MSI also resulted in a mean range of 8.3 to 20.8 days of restricted training[24]. During basic combat training, 21% of all reported musculoskeletal injuries were attributed to load carriage activities for infantry soldiers[15]. Moreover, Jensen et al (2019)[12] investigated the prevalence of MSI in Marine Corps basic training and observed 31% of injuries occurring during load carriage conditioning specifically. Thus, load carriage has become a focal point as a major contributor to the high prevalence of MSI observed in the armed forces, especially among new recruits[12, 20, 22, 25-28].

1.2 Culprits

Given that such a large percentage (~80%) of MSI can be attributed to 'Noncombat' scenarios such as physical training[16, 17], it is suggested that many observed MSI in the armed forces may be avoidable by modifying risks. However, MSI risks must be clearly delineated before prevention strategies can be implemented. As mentioned earlier 21-31% of lower body MSI occur during load carriage activities for new recruits[12, 15]. Load carriage activities such as loaded road marches have been associated with MSI, especially MSI involving the lower body[5, 16, 29-35]. A study by Schuh-Renner et al (2017)[36] of U.S. Army infantry demonstrated that ~50 % of all lower body MSI were attributed to road marching and running, with road marching under load carriage being the greater contributor of the two ambulatory tasks[36]. Specifically, loaded road marching accounts for ~25% of lower body MSI in the entire U.S. Armed forces[15, 16]. The latter finding indicating that individuals in infantry roles are at greater risk, potentially due to the greater load carriage requirements of their basic occupational task requirements. Additionally, when considering the occurrence of low back pain (LBP) alone, 35-75% is classified as load induced[5]. The two largest factors for MSI risk involving loaded road marching were considered the magnitude of load (>25% of body weight significantly increasing risk) and the frequency of loaded marches per month (≥5 loaded marches significantly increasing risk)[36].

In addition to load carriage, sex is a strong indicator of MSI risk in the armed forces. Whilst both sexes are affected by high incidences of noncombat related MSI, female trainees (new recruits) experience nearly twice as many injuries as male trainees[16]. During basic combat training females exhibited more than 10% greater incidence in MSI compared to their male counter parts[14]. Specifically, women experience 82 MSI per 100 recruits versus 54 MSI per 100 recruits for men during basic recruit training[9]. When assessing load carriage sustained MSI specifically, females exhibited a significantly greater risk of MSI with a female to male incident rate ratio (IRR) of 1.02 (95% CI 0.74 to 1.41)[37]. In conjunction, 15% of sustained MSI due to load were classified as serious personal injuries (SPI) for women with the leading site being lumbar spine (43%) compared to only 6% SPI and only 29% at lumbar spine for men[37].

1.3 Injury Perspectives

There are a couple of leading culprits (risk factors) such as load carriage and sex implicated in the observed noncombat MSI occurrence; however, the mechanisms of injury are still not fully understood. As previously stated, ~85%[9, 18, 19] of the injuries suffered are categorized as mechanical, either cumulative microtrauma (repetitive mechanical stress) or non-contact acute trauma (non-ballistic)[9, 10, 12, 18, 19]. While these findings allude to the etiology of observed MSI in the military, they don’t fully elucidate the mechanisms associated with those risks, especially those that are potentially modifiable. To characterize the risks of these potential culprits of MSI more thoroughly there are two important perspectives on injury mechanisms to consider: i) biomechanical and ii) motor control.

A biomechanical perspective on musculoskeletal injury is governed by a set of basic principles: i) threshold tolerances, ii) magnitude of force, iii) line of action/direction, and iv) repetitive loading (frequency). Threshold tolerances are the ultimate strength of a structure obtained by tissue mechanics research, representing the maximum force an anatomical structure in the body can experience before failure (i.e., rupture or fracture)[38]. Threshold tolerances are dictated not only by the magnitude of force but its loading rate as well[38-43]. When conducting human movement research in regard to MSI, one aim is to determine if forces experienced exceed, or near established failure thresholds. Likewise, the line of action or direction the force is applied is important to whether a structure can withstand the force exposure[44-46]. Thus, limb positions at the instance of peak force application may dictate the amount of MSI risk certain activities have. Lastly, mechanical properties are not constant and can be altered with repetitive loading acutely[41, 47] (i.e., changes in stiffness) and chronically[47-50] (i.e., cumulative microdamage). Therefore, a biomechanical perspective of MSI risk may assess how forces change during prolonged activities, the frequency of the exposures and the time/rest/ recovery apparent between exposures.

A motor control perspective utilizes some principles of biomechanics and expounds on them by evaluating the execution of movement using biomechanical outputs (i.e, stride length and stride time specifically for ambulation tasks). Furthermore, the locomotor system (collection of subsystems that globally yield movement) function is assessed in terms of the perception-action cycle (a component of dynamical systems theory)[51, 52]. The perception-action cycle, sometimes referred to a loop or coupling, is the continuous interaction between an individual’s perception of themselves/environment (obtained through sensory information) and the execution of actions that are available to them (affordances)[51, 53]. Affordances are defined by the action capabilities of the individual, their calibration to their action boundaries (the limits of their capabilities) and their perception of the environment, and relevant objects/events within it[54-58]. Therefore, an optimally functioning locomotor system will correctly perceive perturbations, (re)calibrate the system dynamics and make the appropriate affordance selection based on accurately coupling the correct action response to the appropriate information source, while acting within their action capabilities (at that instant in time)[59, 60]. Furthermore, the locomotor system will exhibit stability (maintain task execution strategy in presence of perturbation) and adaptability/flexibility (utilize new strategy to execute same task in presence of perturbation)[61-65]. Under a complex systems theoretical framework there are three specific theories that guide MSI risk assessment from a motor control perspective: i) the variability-overuse hypothesis, ii) movement regulation and iii) locomotor system organization.

The variability-overuse hypothesis relates to, and encompasses repetitive loading, stating that reliance on too few appropriate motor solutions (lack of motor variability) to execute a movement task can lead to site-specific (over)loading[66, 67]. Moreover, greater motor variability can demonstrate greater movement solution capacity (stability and adaptability/flexibility to perturbation)[68-70]. Thus, when executing a movement task there is a goal referred to as the goal manifold which represents the spectrum of solutions (motor variability) that will successfully execute the task goal[69-72]. Movements tangential to a goal manifold (also known as the null space or task irrelevant) can be considered ‘good’ variability (δ_T) and those perpendicular (also known as the task space or task irrelevant) are ‘bad’ variability (δ_P)[71, 73-76]. Therefore, the ratio of ‘good’ to ‘bad’ variability (relative variability = $\sigma\delta_T / \sigma\delta_P$; σ represents standard deviation)[71, 73-76] quantitates motor variability with ratios greater than 1 indicating an individual leveraging their motor solution capacity (available movement patterns) reducing the likelihood of overuse injury (due to reduced site specific loading[67]). However, assessment of motor variability alone is insufficient due to its parabolic nature and should be paired with other measures to clarify locomotor system function. For example, relative variability can increase during novel tasks as individuals explore state-space (abstract representation of task and null space that is interlaced with cost functions and their cost gradients to define various motor solutions) to learn how to better complete the task which can indicate less stability (i.e., greater risk of slip, trip, fall) and use of deleterious/maladaptive movement patterns (overloading certain anatomical structures).

Movement regulation, such as stride to stride regulation, is another tenet of a motor control perspective on MSI. Assessing the movement regulation can evaluate an individual's ability to effectively mitigate movement errors without overloading the locomotor system[59, 60]. Specifically, regulation is determined by statistical persistence assessment (alpha coefficients[77]) of deviations tangential (good variability) [δ_T] and orthogonal (bad variability) [δ_P] to the goal manifold[77]. The quantification of the regulation strategy of the system used for stride to stride fluctuations acts as an indirect assessment of the perception-action loop function namely: (i) the ability to (re)calibrate information-action in a dynamic environment, (ii) (re)weighting the relative importance of information sources as they become available, and (iii) modulate actions based on the relative importance in relation to the successful maintenance of a functionally useful action-response[59, 60].

Lastly, system organization or complexity is used to determine the stability and adaptability/flexibility of an individual to a perturbation[62]. Specifically, in terms of ambulation, spatiotemporal parameters (stride length and stride time) that display fractal dynamics in their variance structure of consecutive stride series display self-similarity indicating subsystems of the locomotor system are operating at different time scales (heterarchical)[78]. The independence of one subsystem operating at a different time scale from another subsystem means that global system (locomotor system) function is less likely to be affected by a perturbation in the local subsystem (stable or more error-tolerant)[78]. System organization is assessed utilizing a set of fractal analysis methods (frequency and time domains) on each of the gait variables[60, 79-83]. The variance structures of a movement signal complexity such as, pink noise (fractal) will indicate that the individual's system complexity is optimal, representing a heterarchical system organization, functioning efficiently and effectively, and stable against perturbations[62]. White or brown noise (nonfractal) may indicate potential risk [74, 81, 82]. Ultimately, any research conducted from a motor control perspective must analyze a movement task in the presence of a perturbation to elucidate locomotor system function and ascertain MSI risk.

1.4 State of the Science

1.4.1 Load Carriage Biomechanics

Load carriage represents a perturbation to the locomotor system that can constrain the system (individual) altering their performance capabilities and has been implicated in the mechanism of many observed MSI in warfighters[12, 20, 22, 25-28]. With the biomechanical constraint of load carriage, the metabolic demands of basic ambulatory tasks such as walking and running significantly increase[84-87]. At relative locomotion velocities, load carriage increases heart rate, oxygen consumption (VO_2), carbon dioxide expulsion (VCO_2) and ratings of perceived exertion (RPE)[84-87]. Therefore, for relative activity durations, loaded ambulation will result in greater levels of fatigue quicker than unloaded[84-87]. Fatigue, in particular neuromuscular fatigue, potentially increases MSI risk[88], thus fatigue due to load carriage may contribute to the high incidence of observed MSI. The increased metabolic demands are a result of the increased biomechanical demands of load carriage because active components of the locomotor system must generate more force to displace the now greater mass of the system[8, 20, 89-97].

During ambulatory tasks the magnitude of load changes the mean behavior of the trunk (greater forward trunk lean) and LE; greater sagittal joint excursions, peak frontal knee adduction; greater peak moments (sagittal plane and frontal plane)[20, 85, 91, 92, 98-105]. Moreover, during prolonged load carriage ambulation, vertical ground reaction forces (vGRF) increase as time progresses[89]. Likewise, in females specifically, the knee frontal plane moment is greater with load carriage (25-45% of bodyweight) compared to unloaded[93]. The increase in energy transmission through the kinetic chain (greater forces/moments) coupled with maladaptive joint positions (peak frontal plane knee angles) at the instance of force application is believed to be responsible for the increase in observed lower body MSI in the armed forces[93]. However, most research has focused on male samples[8], neglecting female sex specific responses and utilized loads that are less than military-relevant loads (<45%)[106].

1.4.2 Load Carriage Motor Control

Whilst there is a substantial amount of research regarding the effects of load carriage on ambulation from a biomechanical perspective[20, 85, 91, 92, 98-105], there is a dearth of information exploring this phenomena from a motor control perspective[105, 107, 108] with the majority of work focused on spatiotemporal gait parameters[105, 107, 108]. Specifically, there is a paucity of research investigating several aforementioned tenets of motor control: i) motor variability (variability-overuse hypothesis), ii) regulation, and iii) system organization (complexity)[109]. An exploratory analysis by Krajewski et al (2020) observed load magnitude and use of ‘unnatural’ locomotion patterns like forced-marching reduced motor variability and impaired regulation and system organization[109] but due to the sample being only women it remains unclear if these observed responses are sex specific. This latter point is important given the anatomical differences (especially at the hip joint [q angle]) and physiological differences (i.e., muscle tissue composition) between men and women which might influence movement strategies[110]. Likewise, the absence of neurophysiological differences between men and women suggests that certain aspects of motor control should not be altered by sex[111, 112], thus direct sex comparisons are needed. Moreover, previous findings with load carriage were exhibited with ~130 consecutive data points, while sufficient for motor variability and regulation analysis, complexity analysis typically requires a minimum of 512 data points, but ~1000 is ideal (~10 minutes)[113]. Therefore, further research is needed with longer time-series data to confirm complexity change responses to load magnitude and locomotion pattern.

1.5 Purpose

The primary purpose of this investigation is to determine the interactive effects of load magnitude and locomotion pattern on lower extremity motor variability, system regulation and system organization during gait tasks in recruit aged individuals. The secondary purpose is to determine if there is a sex specific response to the interactive effects of load magnitude and locomotion pattern on lower extremity motor variability, system regulation and system organization. Lastly, the tertiary purpose is to conduct an exploratory analysis to determine the influence of completion ability (ability to complete full ten-minute trial) response to the interactive effects of load magnitude and locomotion pattern on lower extremity motor variability, system regulation and system organization.

1.6 Specific Aims and Hypothesis

Specific Aim 1.1: Use a goal equivalent manifold (GEM) decomposition method of spatiotemporal parameters to determine the interactive effects of load magnitude and locomotion pattern on motor variability.

Hypothesis 1.1a: As load magnitude increases there will be a reduction in relative variability indicating that individuals have to limit their motor solution capacity[109, 114, 115].

Hypothesis 1.1b: Utilization of an 'unnatural' locomotion pattern such as forced-marching will result in greater relative variability compared to running demonstrating an exploration of state-space due to the novelty (unfamiliarity) of the task[109, 116].

Hypothesis 1.1c: When examining the GEM subspace variability (tangential and perpendicular); tangential variability will be greater than perpendicular variability[109].

Hypothesis 1.1d: As load carriage increases perpendicular variability (or the amount of stride solutions that fail to achieve the trial velocity) will increase[109].

Hypothesis 1.1e: Forced-marching will result in greater perpendicular variability compared running since it is an 'unnatural' locomotion pattern for the given velocity.

Specific Aim 1.2: To determine if sex alters the interactive effects of load magnitude and locomotion pattern on motor variability.

Hypothesis 1.2a: Due to anatomical differences of the lower extremities, women will demonstrate less relative variability compared to men.

Hypothesis 1.2b: Women will exhibit differences in relative variability due to locomotion pattern whereas men will not because of lower extremity anatomical/physiological differences[109].

Specific Aim 1.3: To determine if trial completion ability alters the interactive effects of load magnitude and locomotion pattern has on motor variability.

Hypothesis 1.3a: Individuals who are unable to complete the full ten-minute trial will exhibit less relative variability compared to those who can complete the full ten minutes because non-completers will have a limited motor solution capacity at their disposal.

Specific Aim 2.1: Use detrended fluctuation analysis on task subspace coordinate time series (tangential and perpendicular) derived from GEM decomposition to determine the interactive effects of load magnitude and locomotion pattern on task execution regulation.

Hypothesis 2.1a: Tangential (null space) coordinates will exhibit weak regulation ($\alpha \sim 1.0$) and remain unchanged for all conditions[109].

Hypothesis 2.1b: As load carriage increases, the locomotor system will utilize stricter regulation of task execution specific motor variability ($\alpha \leq .5$ for tangential and perpendicular variability)[59].

Hypothesis 2.1c: Forced-marching will result in stricter regulation of task execution specific motor variability compared to running ($\alpha \leq .5$ for tangential and perpendicular variability)[59].

Specific Aim 2.2: To determine if sex alters the interactive effects of load magnitude and locomotion pattern on task execution regulation.

Hypothesis 2.2a: Motor regulation will be unaffected by sex due to lack of neurophysiological differences between sexes[111, 112].

Specific Aim 2.3: To determine if completion ability alters the interactive effects of load magnitude and locomotion pattern on task execution regulation.

Hypothesis 2.3a: Individuals who are unable to complete the full ten-minute trial will loosely/weakly control stride variations that do not achieve the task goal ($\alpha \sim 1.0$ for perpendicular variability).

Specific Aim 3.1: Use detrended fluctuation analysis and Higuchi box counting methods on spatiotemporal parameters to determine the interactive effects of load magnitude and locomotion pattern on locomotor system gait complexity.

Hypothesis 3.1a: Increases in load magnitude and a forced-marching locomotion pattern will exhibit reductions in gait complexity ($\alpha \leq .5$) independently[109] indicative of a less error-tolerant system[117].

Hypothesis 3.1b: Increases in load magnitude and forced-marching will independently result in increases in fractal dimension equal to stochasticity (FD ≥ 2.0).

Specific Aim 3.2: To determine if sex alters the interactive effects of load magnitude and locomotion pattern on locomotor system complexity.

Hypothesis 3.2a: There will be no significant differences in alpha coefficients (long-range correlation) between sexes[110].

Hypothesis 3.2b: There will be no significant differences in fractal dimension between sexes[110].

Specific Aim 3.3: To determine if completion ability alters the interactive effects of load magnitude and locomotion pattern on motor system complexity.

Hypothesis 3.3a: Individuals who cannot complete the full ten-minute trial will exhibit over regularity (brown noise) [$\alpha \leq 1.3$] in their spatiotemporal parameter time series.

Hypothesis 3.3b: Individuals who cannot complete the full ten-minute trial will exhibit lower (over regular) fractal dimension (FD≤1.5) compared to those can.

1.7 Study Significance

An investigation by Roos et al. (2015)[118] demonstrated that an adapted load carriage conditioning protocol during basic military training significantly reduced the occurrence of acute injuries but failed to significantly reduce overuse injuries and attrition. Despite the advances in training protocols in the literature and the proliferation of biomechanics investigations for load carriage, lower body MSI persists in military populations. Importantly, the leading mechanisms of MSI in military populations are cumulative mechanical stress (overuse injuries), and slips, trips and falls suggesting that alterations in or deficits to motor control strategies for load carriage execution play an important role in the high incidence of MSI in the military. Specifically, the high incidence of slips, trips and falls demonstrates a disruption to the perception-action cycle that cannot properly calibrate to systemic perturbations[51, 52]. This indication is further evidenced by the observed differences in motor variability structure (good versus bad variability ratio)[77], stride to stride regulation/control[76, 77], and gait complexity[80, 81, 83, 119-121] between impaired/at risk populations compared to healthy controls.

Utilizing new analysis techniques motor variability and system complexity of gait can be assessed to define the state, and potential ‘health’ of the movement system, and motor control of an individual. A lack of motor variability may portend cumulative mechanical stress (overuse) injuries due to site-specific tissue overloading[50, 67]. In addition, reduced motor variability can indicate a shortage of movement flexibility, making the individual more unstable to perturbation (because of the lack of movement solutions)[71, 72]. In fact, healthy elderly individuals with low fall risk display similar magnitudes of motor variability and stride regulation to young healthy controls[77]. Likewise, amongst healthy individuals, motor variability is the same regardless of locomotion pattern (running versus walking)[76]. As gait velocity increases regulation remains the same but relative variability increases (good::bad variability)[76]. Thus, these previous findings[75-77] indicate that compromised motor variability (more 'bad' variability versus 'good' variability) and changes in stride regulation control potentially increases slip, trip and fall risk. However, there is limited research indicating how the addition of load carriage effects motor variability structure (good::bad variability) and stride regulation control[109].

Complexity assessment through the use of fractal analysis methods has demonstrated great utility in other disciplines (cardiac health) serving as a biomarker of health status[122-124]. Current research has demonstrated a correlation between white noise variance structures (complexity structure) from gait dynamics (S_L & S_T) to impaired populations[80-82]. Interestingly, only central neurological impaired populations demonstrated negative alterations to complexity of gait dynamics compared to peripherally impaired populations[125]. The correlation of the central nervous system (CNS) to changes in gait complexity implicates its impact on system organization[126]. Moreover, the role of the CNS indicates that disruption to the perception-action loop manifests in alterations in gait complexity. Conversely, healthy populations demonstrate a pink noise variance structure in gait parameters[80-82]. However, research is needed to further understand how load carriage and imposed locomotion patterns (forced-marching) alters system complexity due to their impacts on the perception-action loop (fall risk)[109]. Thus, the results of the present investigation provide the opportunity to identify a non-invasive marker of injury risk.

2.0 Literature Review

Warfighters experience a range of ‘noncombat’ related MSI that have been attributed to mechanical etiologies[21-23]. Moreover, recent findings have indicated that load carriage may be one of the main culprits to the development of these observed MSI[12, 20, 22, 25-28]. To better identify potential modifiable risks of MSI, basic mechanical principles of MSI mechanisms must first be fully understood. The study of load carriage during ambulatory tasks is not necessarily a new phenomenon and has been investigated thoroughly from a biomechanical perspective and in terms of metabolic demands (physiological). However, there is a paucity of information regarding the underlying elements of motor control and the perturbation of load carriage[105, 107, 108]. To best present a comprehensive review of the extant literature, requires that we first better understand the mechanisms of injury from a biomechanics perspective followed by the biomechanics of load carriage specifically. Next principles from a motor control perspective must be explained and how they have been applied to the understanding of motor control during ambulatory tasks to identify future directions and methods of analysis to be applied to load carriage research.

2.1 Injury Mechanisms

Due to the breadth of lower body MSI observed in the military and the anatomical complexity, the etiology of each specific injury will not be discussed. However, common mechanical principals that play a role in injury regardless of the specific anatomical structure will be elaborated on. The distinction between non-contact acute trauma and cumulative microtrauma is important due to the mechanistic differences in injury mechanism. Acute traumas are characterized by large magnitude loads (force or moment), high strain rates and misalignment of supportive structures. The supportive tissues (ie: bone, cartilage, ligaments, tendons and muscle) that make up the body to serve as supportive/motor structures, all have specific stress tolerance failure thresholds[38] commonly referred to as ultimate stress[47]. The soft tissue structures in the body have anisotropic characteristics or different properties in different directions[41, 44-47, 50, 127, 128]. Specifically, when assessing the microstructures of bone, ligaments or cartilage, the supportive 'struts' (trabecular bone, collagen fibers) are oriented in a certain direction[44, 129]. Depending on the specific material properties the orientation of 'struts' alters the capability of that structure to tolerate load in certain directions[39, 40, 44, 46, 47, 129-131]. Application of an external load at a magnitude greater than ultimate stress will result in the failure of that specific supportive tissue [127, 132]. For many of the more pliable soft tissues (cartilage, ligaments and tendons), plastic (unrecoverable) deformation occurs before complete rupture of the tissue [38, 41]. Ligaments in particular are capable of withstanding ~4-6% of deformation with ability to recover, but even at those values, the ligaments have the potential to demonstrate residual effects (compromised ultimate strength)[38, 41].

Although substantial absolute external load magnitudes are possible, like those experienced at landing from static line parachuting[133-135], most external load magnitudes are below ultimate stress values[38]. The strain rate (velocity) at which the load is applied reduces the ultimate stress failure magnitude[38-43]. Furthermore, nonlinear increases in strain rate when applying external loads to soft tissues yields more deleterious results such as absolute failures[41-

43] or microdamage[39-41]. At the microstructure level, the elastic properties of soft tissues allow for larger load magnitude tolerance at low strain rates [38, 41]. At the macro structure level (limb as a whole), the soft tissues can withstand greater loads because of the distribution of energy (mechanical stress) across multiple structures[136, 137]. Importantly, from a neuromuscular perspective, this is achieved through the greater motor recruitment resulting in eccentric action of muscles to ameliorate energy as it travels up the kinetic chain[136, 137]. Therefore, at high strain rates it is not unlikely to observe tissue failure at a load magnitude below the absolute stress failure threshold observed in some tissue mechanics studies[38].

Alignment of supportive structures is another important factor in non-contact acute trauma. Supportive tissues of the human body have anisotropic characteristics[44-46] and their mechanical properties are not uniform across all positions[46, 130]. Moreover, position can alter the characteristics of external loads, resulting in soft tissues experiencing multi-directional external loads (forces and moments) which yield greater percentages of maximum principle strain[130] and stress failure[42, 127]. Therefore, if a limb is positioned improperly (force line of action and orientation of structure to handle greatest force are misaligned) at the moment of external load application then it could lead to the structure failing. Indeed, mechanical models have demonstrated that when joints deviate into extreme frontal knee plane positions injury threshold is reduced [138]. For example, the fiber alignment of intervertebral discs and the circular shape of the disc (annulus fibrosis) itself yields large tensile force tolerance, as long as the force is distributed equally amongst the whole disc[127, 139, 140]. This is achieved when the nucleus pulposus is in the center of the intervertebral disc and spreads out evenly during compression pushing against the annulus fibrosis, bending the fibers creating a tensile force[127, 139, 140]. However, if the spine is misaligned (rounding of the back), the reduction of joint spacing at one end shifts the nucleus pulposus out of center. Thus, while experiencing a large compressive force, significant tensile forces are applied to one specific region of the disc, exceeding its threshold tolerance and resulting in tissue failure (ie: prolapse, extrusion, sequestration).

Unlike acute trauma which occur as the result of a singular incident, cumulative microtrauma is due to the chronic over-loading of soft tissues, with insufficient periods of rest allowing repair and adaptive responses. When partaking in any physical activity such as running, repetitive mechanical stress results in microdamage at the microstructural level of soft tissues (destruction of myofibrils in muscle[136, 141], microcracks in trabecular and cortical bone[47-50]). Physiological responses to mechanical strain yield the upregulation of specific pathways that result in new tissue generation so that the body is better suited to handle the mechanical stress in the future (adaptation)[47, 142, 143]. However, natural (unaided through exogenous substances) regeneration processes take some time[142, 143]. The body is never solely in a catabolic (breakdown) or anabolic (build up) state, but both occur simultaneously. During adequate recovery and nutritional intake [144, 145] , the ratio favors anabolic processes, resulting in positive adaptation. But when assiduously exposed to mechanic stress, absent of adequate recovery, injury risk increases [145-147] potentially due to a ratio shift in favor of greater catabolism [144]. Overtime, this heightened catabolic state compromises the structural integrity of many supportive soft tissues, leading to injuries such as stress fractures (micro-fractures that don’t result in an absolute threshold failure immediately).

While repetitive mechanical stress over a larger time scale (multiple days/weeks/months) may result in the negative impact on mechanical properties of soft tissues, acute (minutes/hours) repetitive mechanical stress can affect them as well[38-41, 48, 49, 148-150]. With cyclical (repetitive) loading there is a progressive loss of modulus (stiffness) and an accumulation of strain[41, 47]. Therefore, as repetitive loading prolongs, the ultimate stress (threshold) of the tissue is reduced requiring a smaller magnitude to achieve failure[41, 42, 47]. In fact, an investigation by Bowman et. al (1998)[39] demonstrated that trabecular stiffness could be reduced up to 70% after 10^6 cycles, even at minimal external loads. Likewise, with repetitive mechanical stress, as microdamage accumulates, soft tissue material heterogeneity increases[50]. At high levels of heterogeneity strain is concentrated to specific sites and ultimate stress is reduced[50].

Additionally, another important component to cumulative microtrauma is variability of mechanical exposure [66, 67]. If the exact same movement pattern is utilized then, the same structures are exposed to mechanical stress repeatedly. Regularity in movement (lack of motor variability) in conjunction with maladaptive movement patterns further exacerbates the loading of specific structures[67]. Moreover, if limb coordination is replicated with minimal degree of difference stride to stride then a specific site will be exposed to an even greater total mechanical stress load. Thus, the mechanical stress load is not distributed to other structures over a prolonged period of time, resulting in quicker breakdown[50].

2.2 Biomechanics of Load Carriage

2.2.1 Trunk Kinematics

Compared to unweighted marching, loaded marching produces greater sagittal plane forward trunk lean, that increases as load magnitude increases [20, 85, 98, 99]. Moreover, in a fatigued state, trunk sagittal plane excursion increases while walking with a load[151]. While the forward leaning torso has been attributed to the load magnitude itself, these studies utilized traditional ruck sacks and did not account for alterations in center of mass (COM) displacement [20, 98]. Thus, changes in torso position are most likely not due to the magnitude of load itself, but by the degree at which it moves the COM posteriorly. Therefore, to maintain a center of gravity (COG) within the base of support or slightly anterior while ambulating horizontally, individuals must adjust for the posteriorly translated COM by leaning the torso forward in the sagittal plane. An investigation by Caron et al.(2013)[99] found that sagittal plane forward trunk lean increases linearly with linear load increases, however the vector orientation between the COM and the ankle/knee remains unchanged up to 40% of body weight[99]. Moreover, a study by Simpson et al (2012)[152] observed more upright trunk posture with 30% of body weight packed in a high position compared to mid and low packs, suggesting COM changes modulate trunk position. Interestingly, most prescribed loads (20-60kg) exceed 50% of an average individual's body weight, indicating potentially deleterious effects of forward trunk lean for 'military relevant' loads. Additionally, this information was obtained utilizing infrared motion capture systems. While currently considered the 'gold standard', these reflective marker-based systems can still demonstrate large ranges of error ~1-35°[153, 154], 2-40mm[155], which are further exacerbated by skin artifact[155-158]. Likewise, many of these studies utilized custom marker plates on extended rods to avoid marker occlusion from the ruck sack, potentially increasing the error through mechanical vibration[89, 91].

2.2.2 Joint Kinematics

In addition to trunk position changes, greater peak hip flexion angles[100-103] and knee/ankle joint sagittal plane excursions[91, 104] are observed compared to unloaded marching tasks. Moreover, with the addition of fatigue, loaded marching further increases hip/knee joint sagittal plane excursions[151]. Posterior situated loads as low as 15% of body weight elicit increased ankle joint excursions[104]. For both sexes, knee frontal plane excursions increased when comparing unloaded and loaded marching[8]. Females had greater frontal plane hip excursions when compared to males directly[8]. Interestingly, women also exhibited reduced sagittal plane knee excursions as load increased[8], an antithetic finding compared to previous research regarding males[91, 92, 105]. These findings have been specifically attributed to the female sex; however, absolute loads and walking velocities were used in the study by Loverro et al. (2019)[8]. The females in that sample were significantly smaller than the males, so the relative load females carried were substantially greater than the males. Conversely, a study by Silder et al (2013)[102] observed no sex differences in sagittal plane joint kinematics when utilizing relative loads. Therefore, the changes in frontal plane hip excursions may have been due to the load magnitude and not sex, but a direct comparison of males and females with relative loads of three-dimensional kinematics would be needed to further elucidate the differences.

2.2.3 Gait Spatial-temporal characteristics

Loaded marching decreases step length but increases stride frequency[20, 89, 105, 107, 109, 159-161]. However, Caron et al. (2015)[162] observed for posterior packed loads up to 30% of body weight did not affect stride frequency, but that loads greater than 30% and up to 40% significantly increased stride frequency. For running and walking locomotion patterns, ground contact time increases as load increases[103]. Interestingly, unlike unloaded conditions, during loaded conditions while in a fatigued state step width variability increases significantly compared non-fatigued[151]. When comparing sexes while using relative loads (anterior-posterior loaded) up to 30% of body weight, there were no significant differences in gait spatiotemporal characteristics[102].

2.2.4 Ground Reaction Forces

Loaded bipedal ambulation significantly increases absolute vertical ground reaction forces (vGRF) compared to unloaded ambulation[102, 163]. Interestingly, vGRF increases are not equal to the amount of load added, investigations have demonstrated ~6% increase in vGRF for every 10% increase in load[102, 103]. Load position itself within a posterior loaded pack has no effect on average absolute vGRF[152]. Loads packed lower in the pack do however yield greater peak vGRF in the early deceleration phase of early contact vGRF compared to mid and high packed loads[152]. In addition to increased vGRF, load carriage alters experienced plantar pressures, increasing pressure (~20 - 88kPa) in the rearfoot, medial midfoot and medial forefoot when compared to unloaded walking[163]. When preferred stride frequency is decreased by 15% a significant increase in anterior-posterior ground reaction forces (GRF) is observed[89].

2.2.5 Joint Kinetics

From a joint kinetics perspective, loaded marching produces greater absolute net external joint moments, mechanical work and physiological work[20, 98, 102]. As loads increase peak joint external moments (hip, knee and ankle) during early stance phase significantly increase compared to unloaded and 15kg loads[91]. In conjunction with decreases in stride frequency, peak extension moments at the knee during early stance phase significantly increase[89]. Additionally, decreases in stride frequency yield ~25% increase in ankle joint power absorption during early stance phase[89]. Marching with ‘operational loads’ resulted in significantly greater vertical ground reaction forces (vGRF) that increased over the duration of the task when compared with no load carriage[98]. Furthermore, load carriage reduced knee moments in the sagittal and frontal planes, while hip moments increased as load magnitude increased during a walking task[8]. Additionally, sex comparisons have only revealed significant greater peak hip abduction moments (during stance phase) [8].

Joint work, at the knee and ankle, significantly increases with load during walking, demonstrating a linear relationship[84]. More specifically, knee positive joint work exhibits significant increases with load compared to unloaded walking during the rebound phase of gait (12-30% of the stride)[84]. Likewise, ankle positive joint work exhibits significant increases with load carriage compared to unloaded walking during the propulsive phase of gait (45-60% of stride)[84]. In addition to increased joint positive work, the positive work exerted onto the COM increases linearly with posterior load magnitude[84]. As load and locomotion velocity increase, positive joint work increases[92]. Typically, as load increases, the greatest contributor to positive work shifts from the ankle to hip[92]. However, which joint contributes the most to positive work is largely dependent on the load configuration[92].

Limited research has assessed joint power in favor of analyzing joint stiffness. The lone study observed increases in hip positive power as load magnitude increased indicating the hip is responsible for task execution as task demands of load carriage increase[92]. For dimensionless or global leg stiffness (peak vGRF divided by the change in COM to center of pressure [COP] distance) resulted in greater stiffness for loads of 10-30% of body weight during running locomotion patterns[103, 162]. When calculating vertical stiffness, which represents the collective forces of the lower extremities that resist the COM collapsing (falling), increasing loads increase vertical stiffness[162].

2.2.6 Energy Expenditure

Metabolic cost significantly increases with load carriage, even with smaller loads of 15% of body weight as much as a 12% increase in metabolic cost can be observed[104]. The relationship between metabolic cost and load has been shown to be linear up to 40% of body weight; as load increases, metabolic cost increases[84, 85]. This finding is further evidenced via heart rate and rating of perceived exertion (RPE), which increase significantly with loaded walking compared to unloaded[85]. However, there are no significant heart rate differences between loads of 20-40% of body weight[85], but this finding is most likely due to trials being conducted at a self-selected walking speed which observed decreasing walking speeds as load increased.

2.3 Motor Control

2.3.1 Bipedal Ambulation

Bipedal ambulation or gait requires the complex integration of multisensory information (optical, somatic and vestibular) that is used to coordinate actions within specific environments in order to achieve goal-directed movement[164-168]. Perceptions of continuously obtained multisensory information yield opportunities to act (affordances) resulting in a perception-action coupling, with a specific movement's success predicated on the modulation (tuning and weighting) of the afferent signals that provide (or fail to) appropriate affordances for the task[51, 53, 165, 168-172]. In conjunction with sensory ‘reafference’, feedforward mechanisms stimulate coordinative structures or muscle synergies that produce a desired movement that achieves a locomotion task goal[173-175]. Collectively, the reciprocal cooperation of feedback (afferent) and feedforward (efferent) subcomponents executing a locomotion task is known as the locomotor system. The function of the locomotor system reflects the emergent properties of the organization of the degrees of freedom during locomotor tasks, with specific macroscopic pattern of organization being influenced by the confluence of cost functions (i.e. metabolic efficiency and energy dampening), task, organism (including feedback and feedforward processes) and environmental constraints (i.e. gravity, uneven terrain)[114, 176-180]. Optimal locomotor system function is represented by a global biomechanical output that is both stable and adaptive to perturbation[56, 59, 114, 181].

Human gait is commonly associated with two distinct biomechanical models of locomotion: i) the inverted pendulum model for walking where individuals essentially vault from one stiff leg to the next[182-185], and ii) the spring mass model for running where individuals utilize elastic energy to spring forward[164, 183, 186-189]. The joint specific actions are altered by locomotion pattern despite shared prime movers for horizontal translation across different patterns[167]. While the inverted pendulum model and spring mass model represent the observed biomechanical outcomes of different locomotion patterns, they fail to thoroughly describe motor control of bipedal ambulatory tasks. Dynamical Systems Theory (DST), sometimes referred to as complex systems, is a frame work encompassing many disciplines (mathematics, physics, biology, psychology and chemistry) that explains the organization of multiple systems that are in constant flux and evolving over different timescales (chaos theory)[63, 64, 190]. Organization of subsystems are never strictly hierarchical, and they evolve with the changes in cost functions, task constraints, system (individual) constraints, and environmental constraints[114, 176, 177]. During goal directed movement, interaction between these components yield numerous potential movement solutions (affordances) based on our internalized representation of the environment (perception) [51, 52]. Unlike cognitive control theories which focus on individual and a single cause, DST emphasizes the role of all components including the environment and accounts for the entire perception-action cycle[191]. Thus, the dynamical system of human movement (locomotor system) is the interplay between the subsystems of the human body and the environment.

Subsystem interaction in a dynamical system occurs through recurrent processing that uses multisensory feedback information (proprioceptive, interoceptive and visual) to provide real-time data on the state of the system constraints (proprioceptive and interoceptive) and the environmental constraints (visual) and help calibrate movement to continually execute tasks successfully[164-168]. Specifically for gait, stepping or stance phase execution, is a mostly ballistic activity that relies on the active modulation of synergistic muscle activations, while concurrently leveraging the passive dynamics of the system (i.e., elastic energy, inertia of COM)[75, 192]. Perturbations can be understood passively or actively (controller) and often depend on the state of the system and the magnitude of perturbation[117, 193]. For example, individuals utilizing appropriate coordination patterns and possessing optimal mechanical properties of lower extremity elastic tissues can handle unexpected step-downs of only a few percent of leg length without controlled responses[117, 194]. Likewise, to actively nullify perturbations an individual must correctly calibrate to a perturbation (multisensory modulation [perception]), utilizing an appropriate action response (affordance choice) and possess capable (strong, fast, precise) actuators to carry out action[117]. During swing phase (pre-step) visual information is utilized to 'plan' (in a recurrent processing capacity of calibrating the system dynamics [i.e., subconsciously]) the next step by predetermining foot placement[75, 166, 195-198].

It is important to understand that systems are never constant and change over time (evolving)[63, 64, 190]. Take for example steady-state gait, locomotion velocity may be constant but the physiological state of the individual changes as the task progresses. Firstly at the start of gait, reliance will be on adenosine triphosphate (ATP) utilization from quick yielding energy substrates of creatine phosphate and glucose (phosphagen and glycolytic pathways)[199]. As the gait task prolongs, the individual can begin to rely on larger energy yielding substrates such at fats (krebs cycle and electron transport chain)[199]. However, as the activity progresses more fatigue inducing by-products (H+ ions) are generated altering the muscular function[199]. Moreover, changes will occur beyond physiological fatigue, such as the ligamento-muscular reflex arc, where cyclical loading (repetitive impact forces) desensitize ligament embedded mechanoreceptors altering proprioceptive accuracy and reducing reflexive activation of associated muscle fibers[200-209]. Therefore, sensory obtainment and recurrent processing is continuous to ascertain (or at least attempt to) the current state of the system and thusly dynamical systems must (re)calibrate to conform to the ever-evolving changes in these systems and possess enough system redundancy to avoid destabilization (i.e., task failure).

No system is perfect and there will always be inherent noise within the system that either must be filtered out or leveraged to improve performance[210]. Considering the complexity of the locomotor system, the nervous system anatomical complexity alone can generate exponential amounts of noise through a few steps of sensory information obtainment from stimulation of nonessential neurons[210]. Greater force requirements, like those during load carriage tasks, could increase neuronal and somatosensory noise[210] making it harder for the system to filter and regulate appropriate dynamics of gait, thus the perturbation overwhelms the system producing a non-fractal structure to gait variability. This is further exacerbated by constraints imposed on the system in terms of kinetic requirements; muscles acting at the limit of their action capabilities and are unable to leverage an appropriate range of movement solutions (affordances) to successfully execute a task. However, the system is attempting to maintain complexity and still function optimally, so it is trying to leverage the entire appropriate affordance workspace, but noise interferes with this regulation process resulting in stochastic gait dynamics (white noise). Moreover, as the system fatigues, additional noise is introduced into the system (chemical by-products such as H+ ions) and further limits the action capabilities. This disruption to action capability that is becoming increasingly constrained may manifest in over-regularity (brown noise) of the system to maintain task execution. Ultimately, state of system health determination must be made through measurable outputs of the system, as it is currently impossible to measure internal noise of a human.

To determine the state of a system during gait tasks, dynamic spatiotemporal parameters are often assessed. Gait dynamic variables along the anterior-posterior axis such as stride length (S_L) and stride time (S_T) impact the anterior-posterior base of support (BOS) while modulating gait velocity (or horizontal locomotion velocity). Variables along the medial-lateral axis such as step width (S_W) and lateral position (L_P) [medial-lateral position of COM relative to a walking path/treadmill] play a role in medial-lateral COM stability[126]. To successfully execute a goal directed bipedal ambulation task, individuals must simultaneously regulate anterior-posterior and medial-lateral gait dynamics[75]. This multi-objective model of gait execution means regulating propulsive torques, exploiting passive dynamics during load acceptance and controlling COM excursions all while leveraging the redundancy of the system to utilize 'good' variability to execute the task optimally for a prolonged period of time[75].

2.3.2 Postural Stability

As mentioned earlier one aspect of ambulation is the regulation of COM[182-185] and therefore an important component to bipedal ambulation is postural control or postural stability. Postural control and stability refer to the ability to manipulate one's COM through three-dimensional space in a manner that results in optimal task execution[75, 192]. Furthermore, postural control is considered stable if the desired posture can be maintained even in the presence of perturbation. Ineffective management of the COM in regard to the base of support (BOS) and the movement goal can lead to an inability to maintain balance and translate through three-dimensional space efficiently. Simplistically, postural control has been defined as retaining the COM location within the BOS, however this an inaccurate statement from a dynamic perspective as walking is essentially 'controlled falling' due to the COM positioned anterior to the BOS. Utilizing this strategy leverages the conservation of angular momentum increasing mechanical efficiency. Considering the importance of postural control there are several mechanisms or strategies to control posture. Moreover, there are many ways to assess the postural control of an individual including static assessments of limits of stability (LOS) and dynamic assessments[211].

Postural control is one of the few aspects of motor control that has been investigated more in regard to load carriage. When assessing static postural control, stability reduces linearly with load increases as evidenced by posterior loads ranging from 12-30kg increasing sway path, sway area and sway velocity in the anterior-posterior and medio-lateral directions[212]. Load carriage as discussed previously results in a multitude of biomechanical changes that result from the COM displacement from the load placement and altered joint kinematics/kinetics due to the load magnitude itself. These two factors act as additional perturbations to a system of postural control that must be readily overcome during ambulatory tasks.

During dynamic tasks, the outcomes of postural control can be different than those observed in static assessments. When assessing local dynamic stability in the medio-lateral and vertical directions, load magnitudes of 20kg had a deleterious effect on Lyapunov exponents, indicating less gait stability[213]. However, loads less than 20% of body weight appear to have no effect on gait postural stability as long as the load is packed evenly (in the posterior position)[214]. The distribution of the load itself tends to have a more profound influence on the gait stability, with asymmetrical loads of 20% body weight across the back yield decreased gait stability in the anterior-posterior, medio-lateral and vertical directions[214]. The strategy utilized and the consequence on stability is dependent on the perturbation itself. During medio-lateral pendular torques applied to the trunk to represent undulating loads, individuals maintain postural control by reducing COM excursions[215]. Furthermore, compared to a fixed load, unstable or undulating loads that impose mediolateral trunk perturbations resulted in less postural stability[215]. Load magnitude also negatively impacts the dynamic postural stability of an individual when jumping over small obstacles[211]. Thus, a perturbation of load carriage does impair the postural control of the locomotor system, but much is unclear in regard to other aspects of motor control (motor variability, regulation and system organization).

2.3.3 Motor Variability

Gait for sustained periods of time is associated with greater variability. Much debate has arisen over the variability observed in gait, with some researchers postulating that movement variability occurs as the consequence of noise or error, while others believing it to be a natural, ‘healthy’ phenomenon. Due to the number of joints and muscles in the lower extremities there is a large number of degrees of freedom that contribute a multitude of movement patterns to accomplish a successful step.

When analyzing human movement, given the number of muscles and degrees of freedom (DOF) there becomes a nearly infinite array of movement solutions available to solve a movement problem [216]. Navigation and selection of the appropriate coupling structures of movement became known as Bernstein’s “Degrees of Freedom problem”[216]. According to DST, sub-systems act as a set of feedback machines, a slight alteration of input information (ie: sensory information) recalibrates the sub-system which in turn alters the organization of other systems[63]. Ultimately, the system desires to organize all the subsystems based on real-time information and by mitigating specific cost functions to achieve the task goal. The system therefore searches for synergies which yields an organization of sub-systems that work in conjunction together optimally[56, 114, 177, 191, 217-220]. Initial stages of learning involve a state-space exploratory period in search of synergies that result in optimal system organization[114, 176, 191, 219, 221]. From a biomechanical perspective, synergies could be the optimal coupling patterns (orientations of body segments in three-dimensional space) that produce desired movement[69, 73, 222-224]. However, the vast number of DOF allow for more than one solution, and when plotted geometrically you create a topology of movement or manifold[69].

At one point in time variability was thought to be errors (noise) produced by the system[61, 69, 72]. As discussed earlier, the DOF problem eventually gave way to the "Principle of Abundance" paradigm[68], which places emphasis on the notion that, a well-organized system leverages the full capabilities of the human body[69, 72]. This has given rise to the principle of equifinality; there are an infinite number of solutions for a single outcome[73-76]. But not all movement patterns (solutions) for a given task are created equal, as some will result in the same output, while others will cause deviations that negatively impact the ability to execute a task goal effectively. Thus, variability should be contextualized as variability that accommodates goal execution (good) and variability that hinders it (bad). The ratio of good::bad variability is known as relatively variability[73-76].

Variability is commonly assessed only through linear methods (Gaussian statistics) such as standard deviation of an entire time series[61, 73]. A linear method only provides the magnitude of variability, without any temporal correlation or contextualization in reference to the goal[73]. Given the broad spectrum of variability and the nuance of stability, a singular analytical outcome will fail to fully explain its context for a given observation. Therefore, a comprehensive set of analyses are required to expound on the meaning of stability/variability that is exhibited during a given task such as bipedal locomotion with load carriage. Furthermore, there are certain limitations and research design requirements that must be fulfilled for a valid interpretation of results. First and foremost, a large number of consecutive data points are needed to conduct most non-linear, equifinality and fractal methods of variability assessment[61, 69, 113, 225]. Moreover, specifically for long-range correlation analysis, the observed task must be a cyclical steady state behavior (ie: walking, running, forced-marching, etc.)[226]. Lastly, methods that consider the temporal component of movement variability is necessary to contextualize the state of the locomotor system.

Goal equivalent manifold (GEM) decomposition is an equifinality method of assessing motor variability temporally in relation to the execution of a task goal. GEM decomposition creates a topology that plots variable parameters either tangential to a goal manifold (‘good’ variability) or perpendicular to the manifold (‘bad’ variability [motor solutions that execute a task such as a step, but don’t meet the desired criteria such as gait speed exactly])[74-76]. Therefore, GEM is capable of assessing the redundancy and stability of the system simultaneously. Repetitive tasks executed with high relative variability indicate a system optimally organized to leverage the redundancy of the system (greater motor variability to execute a task), potentially giving it more flexibility to handle perturbations[74-76]. Additionally, the greater use of motor solution capacity is not only indicative of a more robust system, but a locomotor system using a greater range of coordinative patterns that will reduce site-specific loading (variability-overuse hypothesis).

During unloaded ambulation, S_L and S_T regulation is unchanged as velocity increased, relative variability (tangential::orthogonal variability) structure significantly changed as velocity increased independent of locomotion pattern[76]. Healthy adults leverage equifinality to a greater extent as locomotion velocity increases, as evidenced by the increase in tangential variability and decrease in orthogonal variability[76]. Meaning that at higher velocities, healthy individuals use a greater combination of S_L and S_T variations that results in maintaining the goal task execution and less combinations that negatively impact task execution. At higher locomotion velocities it can be postulated, that for healthy individuals the exploitation of variability along the goal manifold serves multiple purposes: i) reduce excessive mechanical loading of specific structures [67], ii) improve metabolic efficiency by dispersing work across a multitude of muscle fibers

[227], and iii) reduce likelihood of a movement pattern that may be maladaptive or deleterious in nature. In fact, an investigation by Gates et al, demonstrated that in the presence of muscle fatigue, relative variability of the fatigued limbs increased suggesting that redundancy of movement was exploited to maintain successful task execution[227].

The perturbation of load carriage and 'unnatural' locomotion patterns have been minimally investigated in terms of motor variability[109]. As load magnitudes increase (25% and 45% of body weight added with an anterior-posterior loaded weight vest) relative variability decreases, indicating that load constrains the system reducing the action capabilities of the individual (limiting motor variability)[109]. Thus, increases in load magnitude might be increasing site-specific loading because less movement variability can be utilized to successfully execute a task. Conversely, use of an 'unnatural' locomotion pattern like forced-marching increased relative variability[109]. The latter finding suggesting that the novelty of the task elicited a state-space exploratory behavior[223]. Therefore, utilizing an unnatural locomotion pattern may alter the locomotor system, impairing the ability to handle additional perturbations and potentially increasing the likelihood of slips, trips and falls[109]. Moreover, this finding demonstrates that motor variability alone cannot be considered when investigating the effects of different perturbations on the locomotor system.

2.3.4 System Regulation

Goal equivalent manifold (GEM) is a dynamical topology method of assessing motor control of steady state or cyclical behavior such as gait[59]. As mentioned previously, GEM leverages the concept of equifinality; there are multiple ways to complete the same task[59].

However, not all movement solutions are created equal, indicating that some movement patterns are ‘better’ than others. Furthermore, motor control theorizes that the optimal neuromotor system only corrects ‘bad’ movement pattern variants[59]. The quantification of the regulation strategy of the system used for stride to stride fluctuations acts as an indirect assessment of the perception-action loop function namely: (i) the ability to (re)calibrate information-action in a dynamic environment, (ii) (re)weighting the relative importance of information sources as they become available, and (iii) modulate based on the relative importance in relation to the successful maintenance of a functionally useful action-response[59, 60]. **Therefore, in a dynamical system the regulation of the subsystems defines the functional capacity to handle additional perturbations.** Most importantly, unlike other variability analysis tools (i.e., standard deviation, Uncontrolled Manifold [UCM] and Tolerance-Noise-Covariation [TNC]), GEM analyzes the fluctuations in conjunction with the temporal structure of the variance[59].

Conceptually, GEM utilizes a topology-based method where a manifold is plotted on a two-dimensional graph to represent the solution (which can be mathematically derived beforehand)[59]. Each axis represents a task variable that is necessary to execute the task. The goal is the most difficult aspect to define because it is an assumption on the part of the researcher, as we don’t know the specific goal or intention of the individual performing the movement task[59]. Thus, task choice is important when utilizing a GEM framework to improve the validity of the assumed goal function. Stride regulation is then determined by statistical persistence assessment (alpha coefficients[77]) of deviations tangential (good variability) [δ_T] and orthogonal (bad variability) [δ_P] to the goal manifold[77]. Optimal regulation is defined by a minimal intervention principle ($\delta_T\alpha>1$; $\delta_P\alpha <.5$)[77] which suggests that control of stride modulations is not priority, freeing up workspace capacity to potentially handle additional perturbations.

Conversely, a suboptimal regulation strategy is an absolute position control (POS) model [$\delta_T\alpha$ and $\delta_P\alpha$ <.5][59] where considerable workspace capacity is dedicated specifically to stride modulation control and may even be a determinant of fall risk[77].

While gait variability has been often associated with fall risk, interestingly older healthy adults with low fall risk demonstrated similar gait variability along the GEM compared to younger adults.[77]. Moreover, older healthy adults demonstrated no significant differences in stride-to-stride regulations compared to younger healthy adults[77]. Older adults employ the same error-correction and stride regulation strategy as younger healthy adults despite starting with more physiological noise[77]. Therefore, findings suggest that by modulating movement patterns (older adults utilized a shorter and quicker step) older individuals are able to filter noise to allow use of the same regulation method as the younger controls[77]. Thus, gait variability itself may not be the indicator of deteriorating health in individuals but the deterioration of step control in the presence of high physiological noise[77].

Gait (in terms of stride length [S_L] and stride time [S_T]) is regulated independent of locomotion velocity, exhibiting similar statistical persistence within a specific ambulation pattern at different velocities[76]. However, when utilizing different locomotion patterns (i.e., walking versus running) stride-to-stride regulation changed significantly, with running demonstrating a quicker and more direct correction to an errant stride than walking (indicated by the smaller scaling exponent for running) [76]. The tighter stride-to-stride regulation of a running locomotion pattern versus walking for the same velocity along the goal manifold may be a result of the increased physiological noise produced from increased muscle activation requirements[76, 228, 229]. Thus, to overcome or potentially leverage the greater physiological noise inherent to the neuromuscular system during running, healthy individuals impose more control stride-to-stride to reduce the probability of the noise initiating an errant step that may be disastrous (i.e., result in a slip, trip, fall or support tissue failure). Likewise, greater regulation during running infers that additional perturbations to the system could result in task failure (slips, trips and falls)[76, 77].

Gait evaluation has not only emphasized the anterior-posterior regulatory dynamics of gait (S_L and S_T), but on the medial-lateral aspects as well (step-width [S_W] or lateral position [L_P])[75]. While anterior-posterior gait dynamic variables are important to the execution of horizontal translation, medial-lateral variables play in important role in stabilization considering biped's are more unstable in the medial-lateral directions[75, 230-232]. Regulation of S_W is stricter than L_P which demonstrates weaker regulation stride-to-stride, L_P is just as critical otherwise individuals would drift off the intended path or treadmill[75]. Likewise, S_W is most likely controlled more tightly due to greater deviations having a more profound immediate impact (i.e., failure of task execution from a fall).

Ultimately, healthy human bipeds do not demonstrate optimum gait mechanics, but rather regulate stride-to-stride dynamics to correct for minor errors[74]. Moreover, gait regulation aims to maximize energy efficiency in parallel with postural control by leveraging movement redundancy to utilize more tangential or 'good' motor variability[74].

2.3.5 System Organization (Complexity)

As mentioned in earlier sections, complex systems are characterized by the organization of multiple subsystems. The interplay between multiple components inevitably leads to the concept of complexity. Complexity serves as a global representative of the state of a dynamical system by taking into account the self-organization, number of components, determinism and dimensional scaling[63-65, 233, 234]. **Therefore, complexity can be defined as the robustness of the system**

to function optimally in the presence of system perturbations or across multiple attractor states. From the perspective of human movement, the regulatory components (i.e., sensory information, bioenergetics, recurrent neural processing, etc.) add to the complexity of the motor system by modulating behavior optimally. A reduction in complexity can be interpreted as a decline in function of one or more regulatory components[123, 234]. Assessing complexity provides an opportunity to measure dynamical system states without having to measure each sub-system individually (which presently is infeasible). Choosing the correct output measure requires understanding the various tenets of complexity: fractals, criticality and dimensionality.

Complex dynamical systems demonstrate seemingly chaotic (stochastic) output behavior that is temporally dependent. Most common exhibition of complex behavior is the presence of fractal patterns of a time series of a specific output measure such as heart rate fluctuations[122-124], finger tapping[235-239], visual perception[240], cognitive performance[241-244] and stride time fluctuations[81, 82, 245, 246]. Fractal patterns are derived from two-dimensional signals that are classified into three different noise structures: i) $1/f^0$, known as white noise, ii) $1/f^1$, known as pink noise (or flicker noise), and iii) $1/f^2$, known as brown or red noise[247]. White noise is a signal of uncorrelated series of data points, meaning a completely stochastic output (completely random)[247]. Thus, the probability of any behavior for a given output type is equal from one time point to the next. Pink noise represents a balance of stochasticity and determinism in behavior output (power of signal is inversely proportional to the frequency)[247]. Lastly, brown noise represents deterministic behavior of a signal output (dominated by powerful low frequencies)[247].

Fractals represent two specific underlying components of complexity: i) long memory and ii) self-similarity[234]. Long memory refers to the serial correlations or long-range correlations observed in the variance structure of a time-series[61, 234]. Self-similarity represents the scale invariance of the signal structure, meaning the structure (or shape) is similar across multiple scales[234, 248]. A good visual representation of self-similarity is a head of broccoli, if you zoom in to a smaller stock, the shape will be the same as the entire head[234]. Another example of self-similarity is the 'Sierpinski Triangle' which is formed by patterning triangles in a three-point formation and then replicating the same image which ultimately creates a large triangle[248]. Specifically, long memory and self-similarity are characteristics of pink noise. Theoretically, pink noise indicates a complex system organized to leverage regulatory component information to optimally calibrate output behavior to be stable against but adaptable enough to transition to other attractor states in the presence of perturbations.

By contrast, white noise and brown noise exhibit non-fractal patterns[234]. More specifically, white noise is characterized by a lack of long memory (serial correlations), indicating each output to the next is completely uncorrelated to the previous iteration. A lack of serial correlations indicates the regulatory components of the systems are unable to provide necessary information to appropriately calibrate future behavior. A system that can't rely on previous information is destined to potentially make the same mistakes over and over again. In terms of the perception-action loop, a choice must be made from a set of affordances, therefore, previous information helps inform the decision (action)[51, 52]. A white noise output potentially indicates that affordances will be chosen at random for each behavioral output. Likewise, the stability of the system is compromised in the presence of perturbation due to the regulatory systems sub-optimal input (resulting in no modulation of output behavior).

Similar to white noise, brown noise also represents a non-fractal pattern. However, unlike the stochastic nature of white noise, brown noise is highly deterministic. Brown noise specifically is characterized by a strong long memory (super serial correlation) sometimes referred to as infinite memory, that is persistent throughout the duration of the behavior[234]. Typically, brown noise results from the correlation to a single previous iteration and in most cases representative of some type of error signal (powerful low frequency). Whilst the behavior is not random, the error permeates through the system, inhibiting its ability to recalibrate or modulate the behavior. Theoretically it is believed that a specific feedback signal (of one of the regulatory components), auditory for example, imposes a behavioral calibration on the system that overrides or dominates all other information[249]. This is evidenced by the improved reception and processing of $1/ f^1$ (pink noise) structure sensory information[250]. Importantly, brown noise structured behavioral output may be indicative of an inflexible system that cannot adapt to new constraints.

Several methods are utilized to obtain a quantitative value of complexity known as a parameter[234]. Parameters define the state of the system by providing a numeric value that represents the structural pattern (white vs pink vs brown noise) of the signal[234]. Parameters should not be confused with estimators which are not numbers but functions[234]. Different methods assess different components of fractals, but ultimately produce comparable parameters. The most common parameters of complexity are power exponents (ß) derived from power domain assessments (power spectrum density analysis), scaling exponents (∝) derived from frequency domain assessments (detrended fluctuation analysis) and Hurst coefficient (H) derived from a probability analysis[113, 234, 247, 251]. Specifically, the following parameter values indicate each noise structure: i) white noise are ß = 0, ∝ = .5, $H = .5$; ii) pink noise are ß = 1, ∝ = 1, $H = 1$; and iii) brown noise are ß = 2, ∝ = 1.5, $H = .5$[234, 247].

Complexity analysis has been utilized across a multitude of disciplines including human bipedal ambulation or gait[79-81, 83, 125, 246, 249, 252]. Fractal analysis techniques have typically been performed on stride interval (S_L and S_T) time series data[80, 81, 125, 234, 246, 252]. Healthy adults typically display pink noise signal structures when performing walking trials, while individuals with Parkinson's Disease, Huntingdon's Disease and Multiple Sclerosis displayed a completely stochastic noise structure (white noise)[81]. These findings suggests that the central nervous system (CNS) plays a vital role in gait complexity and is further evidenced by individuals suffering from peripheral neuropathy demonstrate pink noise structures similar to healthy controls[125]. Moreover, while the CNS is important to gait complexity, it does not mean that it has a complex role in controlling gait complexity[253].

Impairments to the CNS do not need to be in the form of neurological disorders only. An imposed frequency, such as walking to a metronome, resulted in a shift in gait fractal dynamics from pink (correlated) to white noise (uncorrelated)[246]. This change in gait fractal dynamics postulates that the metronome interferes with the biorhythms at the supraspinal level overriding the natural processes. Therefore, the auditory information generates an interreference or error signal that propagates through the physiological noise disrupting rhythmic neuronal activation patterns. In fact, biomechanical models of walking have demonstrated that motor output noise when moderate to high yielded white noise signal structures[253]. Furthermore, high gain (amplification) noise altered fractal dynamics to a brown noise structure, or caused the model to fall[253]. Thus, the absence of peripheral sensory information has no effect on gait complexity[125], but the obtainment of too much sensory information or specifically high gain sensory information may inhibit the CNS to properly calibrate self-organization resulting in white and brown noise signal outputs[253]. Likewise, use of 'unnatural' gait patterns like forced-marching that override natural gait transitions yield reductions in complexity as well[109].

Disruptions in gait dynamics have also been linked to physiological aspects as well. Poor gait dynamics has been linked to cardiac output[120, 121]. These findings suggest that lack of complex heart rate variability in cardiac output influences the locomotor system, thus disrupting gait dynamics by increasing the stochasticity of the stride-to-stride fluctuations, negatively impacting balance and increasing fall risk[120, 121]. Moreover, the addition of load carriage presumably tasks the individually physiologically more than unloaded locomotion, evidenced by observed reduction in gait complexity with loads 25-45% of body weight[109].

3.0 Methods

3.1 Study Design

Male and female participants completed a within subject cross-sectional design consisting of one session to obtain anthropometrics, ratings of perceived exertion (RPE) and three-dimensional biomechanical data. Three-dimensional kinematic and kinetic data were captured of participants executing ten-minute trials of steady state bipedal ambulation utilizing two locomotion patterns and three load magnitude conditions.

3.1.1 Independent Variables

Independent variables were locomotion pattern (2 levels), sex (2 levels), load magnitude (3 levels) and completion ability (2 levels). Locomotion patterns included: Running and forced-Marching. Running is a 'natural' locomotion pattern utilized when locomotion velocity exceeds gait transition velocity. Warfighters in combat situations may require running to cover or to evacuate injured. Forced-marching is when a walking locomotion pattern is utilized at a locomotion velocity above gait transition velocity.

Load magnitudes included: Bodyweight or no additional load (BW), plus an additional 45% of their BW (+45%BW), and plus an additional 55% of their BW (+55%BW). +45% and +55% load magnitudes were chosen firstly as they adequately represent the range of loads commonly used in the military from 'combat loads' to 'approach march' loads. Furthermore, previous research has primarily focused on relative load magnitudes on the lower end of the spectrum (5-30%)[106]. Likewise, lower relative load magnitudes (≤25% of BW) don't always elicit biomechanical and motor control responses[109].

Completion ability was determined by an individual's ability to complete the full ten-minute trial. Participants who completed the full ten-minute trial for *all* six trials were categorized as 'completers'. Participants who failed to complete the full ten-minute trial for one or more trials was categorized as 'non-completers'. Load carriage tasks can persist for hours in military settings; thus, it is important to determine if there are differences between individuals who can and cannot complete even a ten-minute bout of the required task.

3.1.2 Dependent Variables

1. Spatiotemporal Gait Parameters:
    a. Stride length (m) [S_L]
    b. Stride time (seconds) [S_T]
    c. Stride speed (m/s) [S_S]
2. Variability Measures (unitless):
    a. Goal Equivalent Manifold Decomposition:
        i. Tangential variability (δ_T)
        ii. Perpendicular variability (δ_P)
        iii. Relative variability (RV)
    b. Long-range correlation (α)
    c. Fractal dimension (FD)

3.2 Participant Recruitment

Participants were recruited from local and university population using fliers and the “Pitt+Me” recruitment website. Interested individuals conducted a phone screen with a member from the Neuromuscular Research Laboratory or Biodynamics Laboratory to determine eligibility. If all inclusion criteria were met, then their testing session was scheduled to inform the individual of risks and benefits of study and then written consent was obtained prior to any data collection.

3.3 Participant Characteristics

3.3.1 Inclusion Criteria

Participants had to be 18-35 years of age to represent the age requirements for military enlistment. Additionally, included participants had to be physically active (operationally defined as engaging in moderate to intense physical activity for 1 hour three times a week). The lack of very strict physical activity requirements was to yield a sample similar to military recruits in basic training where there is a wide range of physical fitness levels. Participants should have limited previous exposure to load carriage to better represent the typical skill level of new recruits in military basic training. Furthermore, the relative novelty of load carriage will improve interpretations of motor control between subjects.

3.3.2 Exclusion Criteria

Criteria that excluded potential participants included:

1. Musculoskeletal injuries in the past 6 months
2. On medications that contraindicate physical activity or significantly impact performance parameters
    a. i.e., beta-blockers
3. Medical Conditions that preclude individuals from rigorous physical activity
    a. i.e., high blood pressure, heart disease, severe asthma, etc.
4. Neurological disorder that affects movement
    a. i.e., Parkinson's' Disease, Huntingdon's Disease, ALS, Cerebral Palsy, etc.
5. Pregnancy (due to radiation exposure)

3.4 Power Analysis

Effect sizes obtained from previous load carriage research[93, 109] utilizing a 2 x 3 repeated measures analysis of variance (RMANOVA) were used to estimate sample size. Power analysis was conducted specifically to ensure power for the locomotion by load magnitude interaction. Due to sex comparisons being a secondary purpose, the study is not adequately powered for a true sex comparison. After power calculations it was determined that 34 subjects would need to be recruited (factoring in a 5% attrition rate,) 16 males and 16 females (n = 32 after attrition) are needed to achieve 80% power and significance set to p = .05.

3.5 Instrumentation

3.5.1 Kinematics

Three-dimensional kinematic data was acquired at 100hz via 12 high-speed infrared cameras (Vicon Vantage, Vicon Motion Systems, Oxford, UK) in conjunction with retroreflective markers. Vicon motion systems have demonstrated relatively high accuracy, with 1% difference error for marker velocities[254] and within 2° for sagittal plane joint angles[154]. Frontal plane joint angles demonstrate moderate accuracy with rotation (transverse) joint angles exhibiting limited accuracy[255]. Infrared cameras were optimally positioned in a hemispherical array to generate a large capture space engulfing a split-belt treadmill to collect three-dimensional trajectories. Camera orientation and specific position was to ensure that at least three cameras could view a single retroreflective marker at any position within the capture volume. Global axis defined as i) anterior-posterior (y axis) with anterior set as positive, ii) medial-lateral (x axis) with movement to the right (when facing y axis positive) set as positive, inferior-superior (z axis) with superior direction representing positive.

3.5.2 Kinetics

Kinetic data were acquired at 1000hz via an instrumented dual belt treadmill (Bertec Corporation, Columbus, Ohio). The split-belt treadmill is equipped with two separate running surfaces approximately 152 centimeters long and 50 centimeters wide per belt. Each belt is capable of independently measuring force and torque in all three axes.[256]. Ground reaction forces (GRF) were captured in all three planes: vertical (superior-inferior), horizontal (anterior-posterior), and transverse (medial-lateral). The entire width of the treadmill was utilized as constraints on treadmill width can alter motor control[257]. Additionally, although treadmills reduce the natural variability in gait[258], terrain and locomotion velocity can be more strictly controlled as to not confound interpretations of findings.

3.5.3 Software

Kinematic and kinetic data collection was synchronized and collected simultaneously with Vicon Nexus 2.0 software. Basic kinematic and kinetic variable post processing was performed in Visual 3D (C-Motion, Germantown, MD, USA). GEM decomposition and complexity (detrended fluctuation analysis and Higuchi's box counting method) analysis was conducted with custom scripts in Matlab version 2019a (Mathworks, Natick, MA, USA).

3.5.4 Participant Equipment

Three different load magnitudes were investigated (Bodyweight [BW], plus an additional 45% of their BW [+45%BW], and plus an additional 55% of their BW [+55%]). Load carriage was added utilizing an anterior-posterior loaded weight vest distributed 40:60. The dual sided weight vest was used in favor of a traditional military ruck sack to reduce lumbo-pelvic marker occlusion and control for center of mass (COM) displacement[8, 91]. Due to the structure of ruck sacks, as load increases COM shifts (typically posteriorly), which alters joint kinematics and will confound the interpretations of the effects of load magnitude. Additionally, all individuals wore standardized combat boots (provided by the research team), as footwear has a profound effect on frontal knee plane kinematics[259]. Lastly, participants were instructed to wear tight fitting clothing (spandex, lycra, etc) and thick sports socks (mid-shank height) to reduce the likelihood of developing blisters. Females were required to tie hair up in a tight bun to reduce spinal marker occlusion.

3.6 Testing Procedures

3.6.1 Randomization

Experimental trials were randomized by load conditions to control for ordering effects. Within each load condition locomotion patterns were randomized.

3.6.2 Questionnaires

During the first visit following the obtainment of informed consent, participants filled out a series of questionnaires. Firstly, participants were required to complete a physical activity readiness questionnaire (PAR-Q) to ensure that they are allowed to engage in rigorous physical activity [see Appendix A]. Then basic demographics were collected for all participants. To further understand influences on motor control participants completed custom physical activity history questionnaires [See Appendix B]. These questionnaires ask a series of questions that ascertain the type of exercise that participants regularly engage in including: i) modalities, ii) intensity, iii) duration, and iv) frequency. Moreover, questionnaires determine previous sporting and exercise experience to further elucidate the motor experience of the participant. Lastly, load carriage experience was determined with a series of questions.

3.6.3 Body Composition

Body composition was determined as body mass index (BMI) which is calculated as body mass divided by height squared (kg/m^2). Although dual-energy X-ray absorptiometry (DXA) is the gold standard for determining body composition, it further exposes individuals to radiation (albeit a small dose). However, BMI serves as a suitable surrogate measure of body fat percentage in general populations[260, 261].

3.6.4 Familiarization/Warmup

A dual velocity steady state treadmill protocol (5 minutes of walking and 5 minutes of jogging) was executed to serve two purposes: i) familiarize the participant with the study equipment, and ii) prepare the participant physically for the experimental trials. Most participants are unfamiliar with the use of combat boots and ambulating on a split-belt treadmill (which can initiate an unnaturally wide stance at first due to the two belts). Furthermore, six ten-minute trials of running and forced-marching with and without load was being conducted which can be physically taxing and this protocol prepared the participant for the experimental trials. Participants were first required to walk at a velocity that elicited an RPE between 8-10. At the conclusion of the 5 minutes of walking the individuals immediately transitioned to jogging at a velocity that elicited an RPE of 10-12. Once the correct jogging velocity was obtained, they jogged for 5 minutes.

3.6.5 Marker Set-Up

A custom 41-reflective marker set was employed to capture trunk and lower extremity kinematics with markers on the: anterior superior iliac spine (ASIS) and posterior superior iliac spine (PSIS) to track pelvis movement, a marker at C7, markers on the anterior and posterior aspect of the shoulder, four-marker clusters on the thigh and shank to track each segment respectively, and markers on the calcaneus, 5th metatarsophalangeal joint (MTP) and 1st MTP to track the foot segment (see figure 1). Removable reflective markers were placed on prominent bony landmarks of the greater trochanters, medial-lateral femoral epicondyles and medial-lateral malleoli.

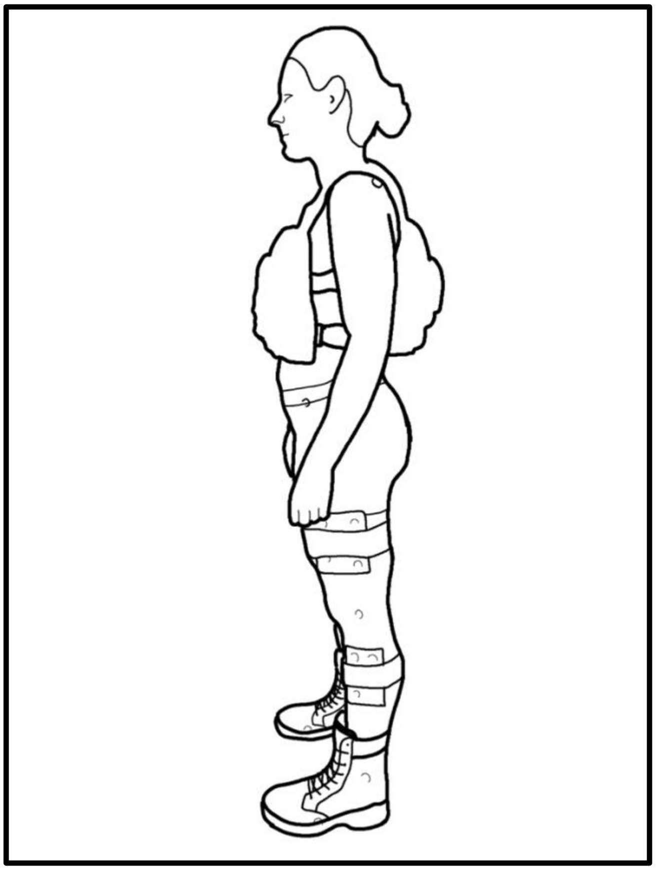


Figure 1. Participant Marker Set-up

3.6.6 Velocity Determination

Prior to data collection trials, gait transition velocity was determined as the mean gait transition velocity of 3 walk-to-run trials utilizing a ramped treadmill protocol with a constant acceleration of 0.05 m/s^2. Mean gait transition velocity was obtained for each load condition immediately prior the performance of that load condition. Gait transition velocity was utilized as a means to provide greater internal validity so that all participants are performing their trials at the same relative velocity effort. The difference in anthropometrics influences the velocity capabilities of individuals and therefore absolute velocities were determined to be inadequate to allow for comparison between participants.

3.6.7 Calibration/Static Trial

A static trial was captured with removable reflective markers for accurate 3D model construction in post processing for respective load conditions.

3.6.8 Experimental Trials

For each load condition, participants completed one trial each of running and forced-marching at a velocity 10% above their gait transition velocity. Following gait transition velocity determination, experimental data trials utilized a steady state treadmill protocol; the treadmill accelerated at 0.5 m/s^2 up to velocity 10% below gait transition velocity and then ten minutes at a velocity 10% above gait transition velocity to ensure adequate number of consecutive data points are collected for GEM and fractal analysis[71, 83]. RPE was assessed at minutes 0, 2, 4, 6, 8 and

10 [See appendix C for exemplar data collection sheets]. For the running trials, participants were instructed to adopt a 'natural', and comfortable gait pattern. During the forced-marching trials participants were instructed to maintain a walking gait irrespective of the treadmill velocity. A minimum of 2 minutes but up to 10 minutes rest was given between each trial to minimize fatigue and ensure full physiological recovery[262].

3.7 Data Reduction

3.7.1 General Data Reduction/Model Creation

Using the Vicon Nexus 2 software (Vicon Motion Systems, Oxford, UK), a custom labeling template was created for the marker configuration used in the study. Once all static and motion trials were reconstructed, the labeling template was used to auto label the static trials captured for each load condition which were then used to auto label their respective motion trials (running and forced-marching). Gap filling methods in Nexus 2 were used to correct any breaks in trajectory data due to marker occlusion. Data was then exported, and post processed in Visual 3D (C-Motion, Germantown, MD, USA).

Subject specific biomechanical models constructed in Visual3D from static trials utilizing a linked model for the trunk and lower extremities using 3D coordinates of reflective markers to calculate kinematics of the trunk, thigh, shank and foot. Segment position and orientation were generated using an optimized 6 degree of freedom (DOF) method that is more accurate than frame-by-frame methods[263]. The trunk and feet were modelled as single rigid segments. Although the spine is comprised of many articulations[127, 140, 264], intervertebral segment kinematics are beyond the scope of this investigation and would require substantially more markers with limited accuracy[265] that would be occluded with the weight vest. Likewise, it is well understood that the foot is made up of multiple segments[266], however specific foot kinematics are beyond the scope of this investigation and require intricate marker-sets that are difficult to apply with boots[266, 267]. For loaded conditions, added mass was applied to the trunk only. Load distribution was evenly distributed across the chest (anterior) and back (posterior), therefore COM is assumed to remain in the same relative location between load conditions[91]. Kinematic and kinetic data were filtered with a low-pass second order Butterworth filter at a cutoff frequency of 6 Hz and 40 Hz respectively as previously determined with residual analysis as the optimal cutoff frequencies[268].

3.7.2 Kinematics

Kinematics were derived from user defined segments of the link-based model in visual 3D. Segments defined by assigned markers (which represent the local coordinate system). Variables such as segment position, velocity and acceleration were calculated as vectors. The transformation of two segment coordinate systems was accomplished with the use of a rotation matrix to define joint angles as a Cardan sequence[269].

3.7.3 Spatiotemporal Gait Parameters

Heel strike was defined as when initial contact vGRF exceeded a 50N threshold and toe off when vGRF dropped below 50N. One stride was considered from heel strike to ipsilateral heel strike. Relevant walking variables calculated: Stride length (S_L) was computed as the distance covered from heel strike to ipsilateral heel strike. Stride time (S_T) was computed as the time elapsed from heel strike to ipsilateral heel strike. Stride speed (S_S) was computed as the quotient of S_L/S_T.

3.7.4 GEM Decomposition

Methods utilized for GEM decomposition have been described in detail by Dingwell et al (2010)[74]. However, to further elaborate the process, S_L and S_T time-series for each trial was normalized to unit variance (dividing by its own standard deviation [σ]). On the GEM a specific operating point was computed for S_T as:

$$S_T^* = \langle S_T \rangle_n$$

Where $\langle \blacksquare \rangle$ represents the average across all n strides of the time series. The specific operating point for S_L was computed as:

$$S_L^* = vS_T^*$$

The new centered operating point was then computed as:

$$S'_{T_n} = S_{T_n} - S_T^*$$

and

$$S'_{L_n} = S_{L_n} - S_L^*$$

Lastly, deviations tangential to the goal manifold are represented as δ_T and deviations perpendicular to the goal manifold are represented as δ_P. These deviations were calculated with a linear coordinate transformation:

$$\begin{bmatrix} \delta_T \\ \delta_P \end{bmatrix} = \frac{1}{\sqrt{1+v^2}} \begin{bmatrix} 1 & v \\ -v & 1 \end{bmatrix} \begin{bmatrix} S'_{T_n} \\ S'_{L_n} \end{bmatrix}$$

The σ of δ_T and δ_P were determined for each load and locomotion condition. Relative variability was calculated as the ratio between $\sigma\delta_T/\ \sigma\delta_P$. Additionally, DFA scaling exponents were computed for δ_T and δ_P (see section 3.7.5.1 for computational methods of DFA).

3.7.5 Complexity Analysis

‘Complexity’ is a subjective term that can indicate different fractal dynamics of a time series depending on the analysis method utilized. Moreover, many analytical methods can be sensitive to parameter settings, and/or are prone to false positive outcomes[270]. Recent evidence has suggested that singular gait outputs (i.e., stride time) possess multi-fractal characteristics, meaning a single signal has varying fractal dynamics yielding different interpretations, which are semantically important[271]. Therefore, multiple analysis methods are recommended to identify all indices of fractal dynamics confidently and comprehensively within a signal. The following series of analyses represent the methods used to examine various components of complexity and confirm results of spatiotemporal parameter time series (i.e., stride time, stride length, and stride speed).

3.7.5.1 Detrended Fluctuation Analysis

Complexity analysis was executed utilizing fractal methods, specifically DFA[113, 124, 234, 245] on spatiotemporal parameters (stride length, stride speed, and stride speed) (~800 consecutive strides Detrended fluctuation analysis (DFA) is a method utilized to determine the long-range correlation of a signal (sometimes referred to as long-range memory). Whilst not the only method of quantifying the auto-correlation function of a signal, it is commonly used for gait spatiotemporal variables due to it’s a reliability especially for group comparison[113]. Ultimately, DFA aims to determine the distribution of informational content in a time series and was developed to improve the root mean square analysis of nonstationary data[272, 273]. Signals with greater complexity will degrade at a quicker rate as the scale/window size decreases, thus less pertinent information is retained to define the system. Therefore, a complex system relies on smaller scales/windows to define its features. However, the size of the scale/window is not the important aspect to determining signal complexity, but rather the *rate* at which it decays as the scale/window sizes change.

The process of DFA has multiple steps and has been previously detailed thoroughly by Peng et al (1995)[272]. However, briefly DFA is accomplished through the following process:

Firstly, the time series data, referred to as the signal from here on out, is mean centered (subtracting the signal mean from each data point) and then cumulative summed (adding the previous interval data point to the present data).

$$X_i = \sum_{n=1}^{i} (x_n - \bar{x})$$

Following the integration of the signal, it is then broken down into a series of windows, also called scales or bins. Windows represent different lengths of time, steps, powers, etc. to separate the signal into epochs. As the rate, and not the window size is important to determining signal complexity, a series of window sizes is analyzed. Moreover, the spacing of these windows can alter the outcome of the analysis, and therefore great care should be taken in determining these windows[274]. The smallest window should retain enough intervals in a single epoch to accurately assess variance, while the largest window should yield at least two epochs (no greater than half the length of the signal) so that a variance average between epochs can be derived. Evenly spaced window sizes (log spacing) yield up to 36% less variation from the true alpha of the signal when compared to arbitrarily chosen window sizes (which is commonly executed)[274]. To obtain evenly spaced windows for a given signal the number of points (*k*) for a diffusion plot must be considered. Then the minimum (w_{min}) and maximum (w_{max}) window sizes are to be determined. Evenly spaced window sizes are then calculated from[274]:

$$\begin{cases} w_1 = w_{min} \\ w_i = \left[w_{i-1} 10^{\frac{log_{10}(w_{\max}) - log_{10}(w_{min})}{k-1}} \right] \end{cases}$$

Brackets indicated that all w_i are rounded to the closest integer. Furthermore, this investigation utilized a *k* = 18, to be consistent with other investigations[274]. The minimum size of the windows themselves should be based on the data and domain being analyzed (i.e., if analyzing a continuous signal collected at 500Hz that represents gait data, a window size of 4 would be far too small as this wouldn't even incapsulate the behavior of a single stride). For DFA conducted on the gait spatiotemporal parameters (i.e., stride length, stride time, and stride speed) the $w_{min} = 10$ and w_{max} = half the length of the time series.

Once the signal has been broken up into epochs for a single window size, each epoch is then detrended (i.e., the nonstationary trend of the signal is removed). The detrending process can be performed with different order polynomials. However, the order of detrending is also important and must be justified. For spatiotemporal data, a first order polynomial detrending process was utilized. For the COM positional data a second order polynomial detrending process was utilized.

After the signal is detrended, the root mean square (RMS) of each epoch is determined and then averaged together to generate the mean RMS for that specific window size. The above steps are then repeated for each window size to create a vector array of mean RMS for that specific window size. The log of each RMS value and the log of the window size is then taken and plotted on a log-log plot. A linear regression is then performed on the log-log plot with the slope (alpha coefficient [α]) representing the complexity of the original signal.

DFA ultimately yields a scaling exponent (α) which represent the correlational structure of the signal. White noise (uncorrelated or completely stochastic) is represented as $\alpha = 0.5$; Pink noise (positive long-range correlations) is represented as $\alpha = 1.0$; Brown noise (persistent long-range correlations or too much regularity) is represented as $\alpha = 1.5$[275]. Classifications based upon a range of α were employed to provide greater clarity as values are rarely the exact values listed above. 'Suboptimal self-organization' was represented by $\alpha < 0.75$; 'Optimal self-organization' was represented by $\alpha = 0.75 – 1.30$; 'Impaired self-organization' was represented by

$\alpha > 1.30$. These values were based upon previously established ranges that classified populations (healthy, elderly, and impaired) as either white, pink or brown noise.

3.7.5.2 Higuchi Fractal Dimension

The Higuchi method for fractal dimension derivation is similar to a box-counting method (such as the Minkowski fractal dimension), however it is generally considered more accurate and reliable, when compared to DFA and the Minkowski box-counting method[270]. Whilst, DFA measures the long-range correlation of a time-series (the auto-correlation function), fractal dimension is a measure of time-series roughness[276-278]. Specifically, roughness indicates the short-term irregularity or local variance compared to the global[276, 277]. The Higuchi method takes the sum of change in amplitude normalized by the time interval, but uses ‘boxes’ of ‘non-interger’ height[270]. The details of this method have been thoroughly outlined in other works (see:[279] [270]) but simply:

$$X_n^m = \{x(m), x(m+n), x(m+2n), \dots, x\left(m + \left[\frac{N-m}{n}\right] n\right)\}, \qquad m = 1, \dots, n,$$

Where x represents a time series of N length that is formatted into a new time series of X_n^m (m represents the initial time and n indicates the time interval. Then curve length is determined with:

$$L_m(n) = \frac{1}{n}\left\{\left(\sum_{j=1}^{\left\lfloor\frac{N-m}{n}\right\rfloor} |X(m+jn) - X(m+(j-1)n)|\right)\right.$$

$$\left.\frac{N-1}{\left\lfloor\frac{N-m}{n}\right\rfloor n}\right\}$$

These first two steps are then repeated for different interval lengths of *n*. The fractal dimension (FD) is then calculated as the slope of a linear slope of the log-log plot of $L_m(n)$ and *n*. For this analysis, the maximum interval length was set to four as it was previously determined to be the optimal length of data similar in nature and length[270]. The Higuchi fractal dimension was conducted separately for each spatiotemporal parameter (stride length, stride time, and stride speed).

Higuchi fractal dimension (FD) determines the amount of information required to describe a time series[270]. Moreover, it measures the *roughness* of the signal, specifically its temporal structure[270]. Fractal dimension (FD) derived from the Higuchi method are classified as follows: FD~1.8 represents pink noise; FD≤1.5 represents brown noise and FD≥2 represents white noise (unstructured randomness), thus the greater the FD value, the more *roughness* (short-term irregularity)[280].

3.8 Statistical Analysis

3.8.1 Subject Characteristic Analysis

Descriptive statistics (mean and SD) were reported for all the variables, with distributions assessed with Shapiro-Wilk tests of normality to ensure that assumptions of normality were not violated (indicated by $p>.05$). Additionally, to determine if there were significant differences between groups (men vs. women) independent t-tests were conducted for age, height, weight, body mass index (BMI), trial velocities and trial loads separately. Lastly, independent t-tests were conducted on all subject characteristic outcomes to determine if there was an effect of completion group (completers vs. non-completers).

For independent t-tests, if Levene's test for equality of variances was violated (indicated by a $p\leq.05$) then p values of unequal variances assumed were examined instead. Cohen's d (d= difference between groups/pooled standard deviation) effect sizes were calculated with $d\leq.2$ represents a small effect, $d=.2-.8$ ($d\sim.5$) represents a medium effect and $d\geq.8$ represents a large effect[281]. The alpha level was set at 0.05 ($p\leq0.05$).

3.8.2 Ratings of Perceived Exertion Analysis

Descriptive statistics (mean and SD) were reported for all the ratings of perceived exertion (RPE) variables (overall RPE and change in RPE). To determine interactive effects of load and locomotion on overall RPE (RPEO) and change in RPE (RPEC) over the entire trial a two-way repeated measure analysis of variance (RMANOVA) for Load x Locomotion (3x2) was conducted separately. If interactions were significant, simple main effects were performed (paired t-tests for locomotion stratified by load and RMANOVA for load stratified by locomotion). If no significant interaction was observed, only main effects were analyzed.

A secondary exploratory analysis (as the sample is not adequately powered for this specific outcome) was conducted to examine the influence of the group factor of sex on RPE outcomes. Therefore, a three-way Load x Locomotion x Sex (3x2x2) mixed factor RMANOVA was conducted on RPEO and RPEC separately. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only two-way interactions of Load x Sex and Locomotion x Sex were examined (as Load x Locomotion would be redundant from the primary analysis of RPE). If no significant two-way interaction was observed, only the main effect of Sex will be examined to determine if there was a difference between sexes.

The final tertiary analysis for RPE, also exploratory in nature (as group sizes were unequal), was executed to determine if there was a difference between participants who were able to complete the full ten-minute trial (completers) versus those who were unable to complete the full ten-minute trial (non-completers). Therefore, a three-way Load x Locomotion x Group (3x2x2) mixed factor RMANOVA was conducted on the RPE outcomes separately (RPEO and RPEC). If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Group and Locomotion x Group were examined (excluding Load x Locomotion to avoid analytical redundancy). If no significant two-way interaction was observed, only the main effect of Group will be examined to determine if there was a difference between groups.

For all RMANOVA, if sphericity was violated (indicated by a significant p value [$p \leq .05$] of the Mauchly's tests of sphericity) then Greenhouse-Geisser adjusted values were reported (denoted by the degrees of freedom). For mixed factor RMANOVA (those including a between-subjects factor) Box's M test and Mauchly's sphericity were executed to test equality of covariance and ensure assumptions of sphericity are met. Post-hoc analysis using Bonferroni-corrected pairwise comparisons were conducted when necessary. Partial eta squared (η^2_p) was calculated as a measure of effect size given the within-subject design[282, 283], with magnitudes of effect interpreted as: 0.01-0.085 (small effect); 0.09-0.24 (moderate effect); and >0.25 (large effect)[284]. The alpha level was set at 0.05 ($p \leq 0.05$).

3.8.3 Goal Equivalent Manifold Analysis

Descriptive statistics (mean and SD) were reported for all the variables assessed. Variables were assessed in a thorough and predetermined order. Firstly, each basic spatiotemporal parameter (stride length, stride time and stride speed) variance/mean was examined prior to the decomposition of the [stride length, stride time] plane. To determine interactive effects of load and locomotion on spatiotemporal parameter variance, a separate two-way repeated measure analysis of variance (RMANOVA) Load x Locomotion (3x2) was conducted on the variance of the entire trial for each spatiotemporal parameter (stride length, stride time and stride speed). If a two-way interaction was observed, then simple main effects were analyzed (RMANOVA for load and paired t-tests for locomotion). If no significant two-way interaction was observed, only main effects were examined.

A secondary exploratory analysis (as the sample is not adequately powered for this specific outcome) was conducted to examine the influence of the group factor of sex on spatiotemporal variance/mean. Therefore, separate three-way Load x Locomotion x Sex (3x2x2) mixed factor RMANOVA was conducted on the entire trial for each spatiotemporal parameter (stride length, stride time and stride speed). If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Sex and Locomotion x Sex were examined (as Load x Locomotion would be redundant from the primary analysis of the spatiotemporal variance). If no significant two-way interaction was observed, only the main effect of Sex will be examined to determine if there was a difference between sexes.

The final tertiary analysis for spatiotemporal variance, also exploratory in nature (as group sizes were unequal), was executed to determine if there was a difference between participants who were able to complete the full ten-minute trial (completers) versus those who were unable to complete the full ten-minute trial (non-completers). Therefore, separate three-way Load x Locomotion x Group (3x2x2) mixed factor RMANOVA was conducted on the entire trial for each spatiotemporal parameter (stride length, stride time and stride speed). If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Group and Locomotion x Group were examined (excluding Load x Locomotion to avoid analytical redundancy). If no significant two-way interaction was observed, only the main effect of Group was examined to determine if there was a difference between groups.

Following the analysis of basic spatiotemporal parameters, relative variability (the ratio of tangential variability to perpendicular variability $[\sigma\delta_T / \sigma\delta_P]$) was assessed (to address **Specific Aim**

1.1). Firstly, the entire trial was assessed and if significant findings were observed then the trial was broken down into a beginning (first 30% of the trial) and end (last 30% of trial) phase to further elucidate if relative variability changed over time. Thus, to determine interactive effects of load and locomotion on the relative variability a two-way RMANOVA for Load x Locomotion (3x2) was conducted. If interactions were significant, simple main effects were performed (paired t-tests for locomotion/direction stratified by load and RMANOVA for load stratified by locomotion/direction). If no significant interaction was observed, only the main effects were examined.

As stated previously, if the entire trial relative variability yielded significant findings, then beginning versus end phases were assessed. To determine interactive effects of load, locomotion and time on task execution a three-way Load x Locomotion x Time (3x2x2) RMANOVA was conducted on relative variability outputs. If a significant three-way interaction was observed, then simple main effects were assessed for each level. If no significant three-way interaction was observed, then only two-way interactions of Load x Time and Locomotion x Time were examined. If the two-way interactions were non-significant then only the main effect of time was examined.

After relative variability was analyzed for the entire sample, the secondary analysis was conducted to determine the effect of Sex (for the entire trial)[to address **Specific Aim 1.2**]. Therefore, a three-way Load x Locomotion x Sex (3x2x2) mixed factor RMANOVA was conducted on relative variability. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Sex and Locomotion x Sex were examined. If no significant two-way interaction was observed, only main effect of Sex was examined to determine if there was a difference between sexes.

An additional secondary analysis to further elucidate effects of sex in conjunction with time was executed. A three-way Load x Locomotion x Sex (3x2x2) mixed factor RMANOVA was conducted separately for the relative variability at the beginning and end phases to determine if time impacted sex differently. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Sex and Locomotion x Sex were examined. If no significant two-way interaction was observed, only main effect of Sex was examined to determine if there was a difference between sexes.

A final tertiary exploratory analysis was executed with a three-way Load x Locomotion x Group (completer vs non-completer) [3x2x2] RMANOVA was conducted on relative variability for the entire trial (to address **Specific Aim 1.3**). If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only the two-way interaction of Load x Group and Locomotion x Group were examined (excluding Load x Locomotion to avoid analytical redundancy). If no significant two-way interaction was observed, only main effect of Group was analyzed to determine any differences between groups.

Similar to the secondary analysis of sex, if significant effects of time were observed then a three-way Load x Locomotion x Group (completer vs non-completer) [3x2x2] RMANOVAs was conducted on relative variability separately for the beginning and end phase to determine if time impacted groups differently. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only the two-way interaction of Load x Group and Locomotion x Group were examined (excluding Load x Locomotion to avoid analytical redundancy). If no significant two-way interaction was observed, only main effect of Group was analyzed to determine differences between groups.

To further elucidate changes of relative variability a three-way Load x Locomotion x Direction (tangential variability vs perpendicular variability) [3x2x2] RMANOVA was conducted on the variance of geometric outputs. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Direction and Locomotion x Direction were examined. Lastly, the main effect of direction was also observed regardless of two-way interaction significance to determine if a true difference between directions existed.

If a significant difference between directions was observed in the primary three-way RMANOVA then the effect of time was assessed in regard to the variance of geometric outputs with a three-way Load x Locomotion x Time (3x2x2) RMANOVA, performed separately for each direction (tangential and perpendicular). If a significant three-way interaction was observed, then simple main effects were assessed for each level. If no significant three-way interaction was observed, then only two-way interactions of Load x Time and Locomotion x Time were examined. If the two-way interactions were non-significant then only the main effect of time was assessed.

A secondary and tertiary analysis of between subject factors of Sex and Group completion were not conducted on the geometric output variance as it would be redundant with the analysis of relative variability already performed. Moreover, four-factor RMANOVAs are strongly recommended against as their interpretation becomes very complicated and the probability of committing a type II error is very high[285], especially with the sample size apparent within the present study.

Once all geometric variables were assessed (spatiotemporal parameters, relative variability and directional variance), temporally related variance (alpha coefficients of tangential and perpendicular coordinates [$\alpha\delta_T$ and $\alpha\delta_P$]) were examined. To determine interactive effects of load and locomotion on stride regulation a two-way Load x Locomotion (3x2) RMANOVA was conducted on the stride regulation outputs separately (to address **Specific Aim 2.1**). If a two-way interaction was observed, then simple main effects were analyzed (RMANOVA for load and paired t-tests for locomotion). If no significant two-way interaction was observed, only main effects were analyzed.

To determine interactive effects of load and locomotion and time on stride regulation control strategies a three-way Load x Locomotion x Time (3x2x2) RMANOVA was conducted on stride regulation scaling outputs separately. If a significant three-way interaction was observed, then simple main effects were assessed for each level. If no significant three-way interaction was observed, then only two-way interactions of Load x Time and Locomotion x Time were examined. If the two-way interactions were non-significant then only the main effect of time was assessed.

Following the analysis of stride regulation for the entire sample, the influence of sex on stride regulation was examined (to address **Specific Aim 2.2**). Therefore, a three-way Load x Locomotion x Sex (3x2x2) mixed factor RMANOVA was conducted on the entire trial stride regulation outcomes separately. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Sex and Locomotion x Sex was examined (as Load x Locomotion would be redundant from the primary analysis of the spatiotemporal variance). If no significant two-way interaction was observed, only main effect of Sex will be analyzed to determine if there was a difference between sexes.

Further, to determine if time affected sexes different a three-way Load x Locomotion x Sex (3x2x2) RMANOVA was conducted separately for each stride regulation output at the beginning and end phases. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Sex and Locomotion x Sex were examined (as Load x Locomotion would be redundant from the primary analysis of the spatiotemporal variance). If no significant two-way interaction was observed, only main effect of Sex will be analyzed to determine if there was a difference between sexes.

A final tertiary exploratory analysis was executed with a three-way Load x Locomotion x Group (completer vs non-completer) [3x2x2] RMANOVA was conducted on stride regulation outputs for the entire trial separately (to address **Specific Aim 2.3**). If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Group and Locomotion x Group were examined (excluding Load x Locomotion to avoid analytical redundancy). If no significant two-way interaction was observed, only main effect of Group was analyzed to determine if there was a difference between groups.

Lastly, to determine if time impacted groups stride regulation differently three-way Load x Locomotion x Group (completer vs non-completer) [3x2x2] RMANOVAs were conducted on stride regulation outputs separately for the beginning and end phases. If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interaction of Load x Group and Locomotion x Group were examined. If no significant two-way interaction was observed, only main effect of Group will be analyzed to determine if there was a difference between groups.

For all RMANOVA, if sphericity was violated (indicated by a significant p value [$p \leq .05$] of the Mauchly's tests of sphericity) then Greenhouse-Geisser adjusted values were reported. For mixed factor RMANOVA (those including a between subjects factor) Box's M test and Mauchly's sphericity were executed to test equality of covariance and ensure assumptions of sphericity are met. Post-hoc analysis using Bonferroni-corrected pairwise comparisons were conducted when necessary. Partial eta squared (η^2_p) was calculated as a measure of effect size given the within-subject design[282, 283], with magnitudes of effect interpreted as: 0.01-0.085 (small effect); 0.09-0.24 (moderate effect); and >0.25 (large effect)[284]. Additionally, frequencies of complexity classifications and change classifications are reported to qualitatively examine individual responses. The alpha level was set at 0.05 ($p \leq 0.05$).

3.8.4 Complexity Analysis

Descriptive statistics (mean and SD) were reported for all the variables. To determine interactive effects of load and locomotion on spatiotemporal complexity a two-way repeated measure analysis of variance (RMANOVA) for Load x Locomotion (3x2) was conducted for stride time, stride length and stride speed alpha coefficients (α), and Higuchi fractal Dimension (FD) separately (to address **Specific Aim 3.1**). If interactions were significant, simple main effects were performed (paired t-tests for locomotion/direction stratified by load and RMANOVA for load stratified by locomotion/direction). If no significant interaction was observed, only main effects were analyzed.

A secondary exploratory analysis (the sample is not adequately powered for a true sex comparison) was conducted to determine if there is a difference between sexes when assessing the influence of load and locomotion on spatiotemporal (ST, SL & SS) complexity (α and FD) (to address **Specific Aim 3.2**). Therefore, a mixed two-way RMANOVA (3x2x2 [Load x Locomotion x Sex]) was conducted separately for ST, SL and SS. If a significant three-way interaction was observed, then simple main effects were examined for each level of the interaction. If no significant three-way interaction was observed, then only two-way interactions of Load x Sex and Locomotion x Sex were assessed. If no significant two-way interaction was observed, only the main effect of sex was examined.

The final tertiary analysis for spatiotemporal complexity, also exploratory in nature (as group sizes were unequal), was undertaken to determine if there was a difference between participants who were able to complete the full ten-minute trial (completers) versus those who were unable to complete the full ten-minute trial (non-completers) (to address **Specific Aim 3.3**). Therefore, a separate three-way Load x Locomotion x Group (3x2x2) mixed factor RMANOVA was conducted on the entire trial for each spatiotemporal parameter (stride length, stride time and stride speed). If a significant three-way interaction was observed, then simple main effects were assessed for each level of the interaction. If no significant three-way interaction was observed, then only a two-way interactions of Load x Group and Locomotion x Group were examined (excluding Load x Locomotion to avoid analytical redundancy). If no significant two-way interaction was observed, only the main effect of Group was examined to determine if there was a difference between groups.

For all RMANOVA, if sphericity was violated (indicated by a significant p value [$p \leq .05$] of the Mauchly's tests of sphericity) then Greenhouse-Geisser adjusted values were reported (denoted by the degrees of freedom). For mixed factor RMANOVA (those including a between subjects factor) Box's M test and Mauchly's sphericity were executed to test equality of covariance and ensure assumptions of sphericity are met. Post-hoc analysis using Bonferroni-corrected pairwise comparisons were conducted when necessary. Partial eta squared (η^2_p) was calculated as a measure of effect size given the within-subject design[282, 283], with magnitudes of effect interpreted as: 0.01-0.085 (small effect); 0.09-0.24 (moderate effect); and >0.25 (large effect)[284]. Additionally, frequencies of complexity classifications and change classifications are reported to qualitatively examine individual responses. The alpha level was set at 0.05 ($p \leq 0.05$).

4.0 RESULTS

4.1 Subject Characteristics

Refer to Table 1 for all means and standard deviations of all subject characteristic outcomes. All women were right leg dominant while only one male was left leg dominant. All participants were able to complete the full ten-minute trial for running and forced-marching at the BW condition (no load). However, two women were unable to complete any time of the +45%BW condition for either locomotion pattern. One subject after completing the forced-marching +55%BW informed the study team they were unable to complete any more loaded conditions. The second subject upon completing the +55%BW conditions reported low back pain. Moreover, the subjects heart rate remained high and did not recover even after a ten-minute rest period. Therefore, at the study team's discretion the subject was withdrawn from the remainder of the study.

Table 1. Participant Characteristics

Variable		Total (n=26)	Men (n=13)	Women (n=13)
Age		26.2±4.0	24.9±4.3	27.4±3.3
Height (m)		1.73±0.10*	1.80±0.07	1.66±0.08
Weight (kg)		72.50±16.49*	81.41±14.14	63.59±13.94
BMI		23.94±3.52	24.96±3.60	22.93±3.26
Trial Velocity (m/s)	BW	1.70±0.22	1.72±0.23	1.68±0.23
	+45%BW	1.65±0.20	1.58±0.14	1.73±0.23
	+55%BW	1.57±0.25	1.66±0.27	1.48±0.21
Load (kg)	+45%BW	32.01±7.55*	36.26±6.23	27.76±6.38
	+55%BW	39.30±9.01*	44.29±7.76	34.30±7.41

**=indicates a significant difference (p<.01) between men and women*

BMI=Body Mass Index (calculated as kg/m^2*), 20-25 considered optimal, >25 overweight, >30 obese, <20 underweight*

See Table 2. for all independent t-test outcomes regarding sex comparisons of subject characteristics. There was no significant difference between men and women in terms of age or trial velocities. However, the men were significantly larger (height and weight) than the women. Despite differences in stature, men and women did not differ in body mass distribution (as indicated by body mass index [BMI]) suggesting that the sexes had similar body compositions. Not surprisingly, the men had significantly greater trial loads.

Table 2. Sex Comparison of Participant Characteristics

Variable		T value	DF	P value	Effect Size
Age		1.64	24	.110	0.65
Height (m)		4.94	24	<.001	1.94
Weight (kg)		3.24	24	.004	1.27
BMI		1.50	24	.150	0.59
Trial Velocity (m/s)	BW	0.51	24	.620	0.20
	+45%BW	1.98	24	.060	0.81
	+55%BW	1.94	24	.070	0.76
Load (kg)	+45%BW	3.44	22	.002	1.35
	+55%BW	3.36	24	.003	1.32

DF=degrees of freedom; Effect size=Cohen's d.

See Table 3. for all independent t-test outcomes regarding completion group comparisons of subject characteristics. There were no significant differences between completers and non-completers for any of the characteristics (height, weight, BMI, trial loads or trial velocities) with the exception of +45%BW trial velocity where non-completers (1.80±0.27 m/s) had greater (p=.03) treadmill velocity than completers (1.60±0.14 m/s) [indicating that non-completers transitioned from walking to running at a higher velocity compared to completers].

Table 3. Completion Group Comparison of Participant Characteristics

Variable		T value	DF	P value	Effect Size
Age		1.17	24	.25	0.50
Height (m)		0.08	24	.94	0.04
Weight (kg)		0.11	24	.91	0.05
BMI		0.20	24	.84	0.09
Trial Velocity (m/s)	BW	0.85	24	.40	0.36
	+45%BW	2.34	24	.03	1.10
	+55%BW	0.14	24	.89	0.06
Load (kg)	+45%BW	0.16	22	.87	0.07
	+55%BW	0.03	24	.97	0.01

DF=degrees of freedom; Effect size=Cohen's d.

4.2 RPE

Refer to Table 4 for all means and standard deviations of overall RPE values (segregated by the total sample, men and women). When assessing overall RPE there was no significant interaction between load and locomotion ($F_{2,46}$=1.55, p=.22, η^2_p=.06). As load increased, overall RPE increased independent of locomotion pattern as confirmed by the significant main effect of load ($F_{2,46}$=87.99, p<.001, η^2_p=.79) [See Figure 2]. Post hoc analysis revealed that BW (10±2) was significantly (p<.001) less than +45%BW (15±3) and +55%BW (16±3). Moreover, running resulted in a greater overall RPE compared to forced-marching independent of load as confirmed by the significant main effect of locomotion ($F_{1,23}$=6.47, p=.02, η^2_p=.22) with running having a greater (14±2) overall RPE than forced-marching (13±3).

Table 4. Ratings of Perceived Exertion (RPE)

Variable	Group	RBW	FBW	R45	F45	R55	F55
RPE Change	Total*	3±2	3±2	6±3	6±4	6±3	6±3
	Women	3±1	3±2	6±3	5±3	6±4	6±3
	Men	3±2	3±2	6±4	6±4	6±3	5±3
RPE Overall	Total*†	10±1	10±2	15±3	14±3	16±3	15±3
	Women	10±1	9±2	16±2	15±3	17±2	15±3
	Men	10±1	10±2	15±3	14±3	16±3	15±3

[mean ± standard deviation]
RPE = Borg scale (6-20); R=Running; F=Forced-marching
**=Significant main effect of load; †=Significant main effect of locomotion*

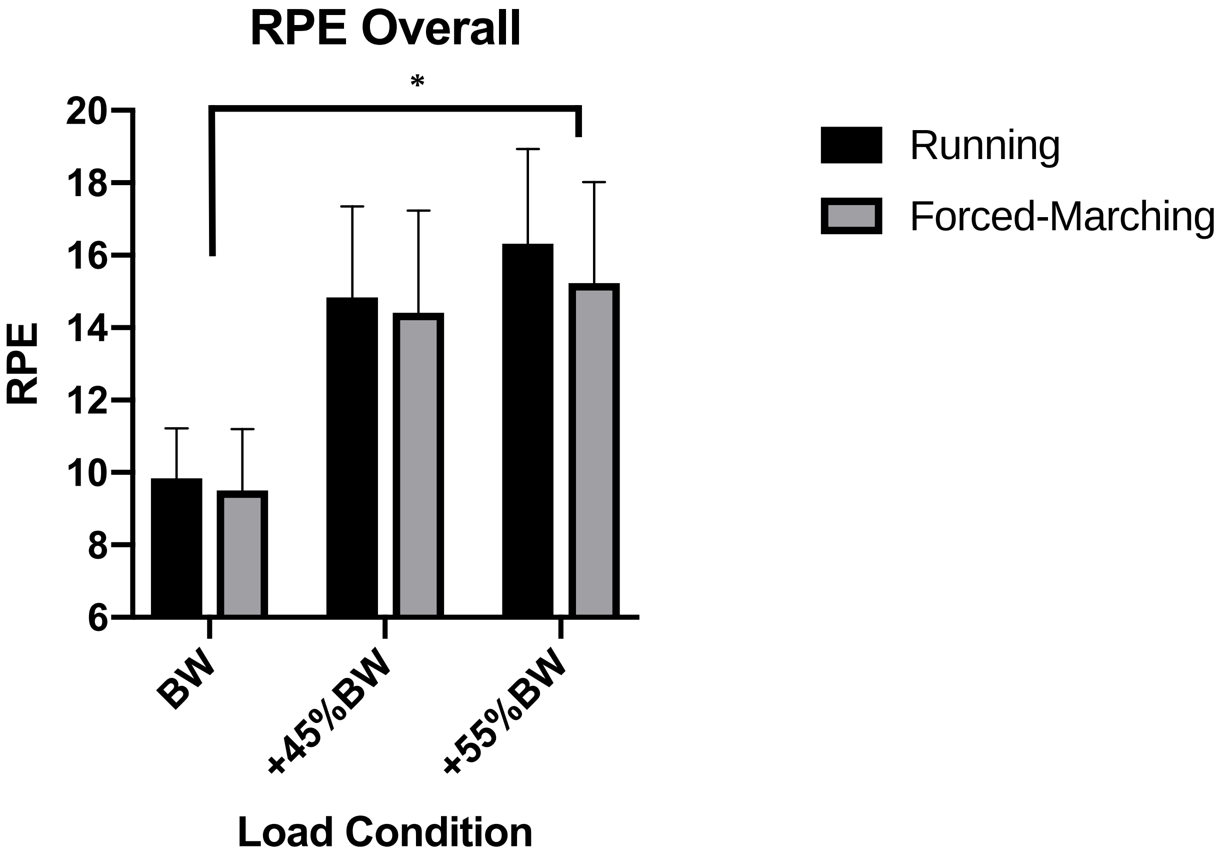


Figure 2. RPE Overall
**=Significant main effect of load*

When comparing the effect of sex on overall RPE (RPEO) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,44}$=0.01, p=.99, η^2_p=.00). There were no significant two-way interactions of load and sex ($F_{2,44}$=0.65, p=.53, η^2_p=.03) or locomotion and sex ($F_{1,22}$=0.07, p=.79, η^2_p=.00). Additionally, there was no main effect of sex ($F_{1,22}$=0.05, p=.83, η^2_p=.00).

When comparing the effect of completion group on overall RPE (RPEO) there was a significant three-way interaction between load, load and sex ($F_{1.48,32.51}$=5.84, p=.01, η^2_p=.21).

There was a significant simple main effect of load for completers during each load condition: running ($F_{2,21}$=64.60, p<.001, η^2_p=.86) and forced-marching ($F_{2,21}$=37.13, p<.001, η^2_p=.78); with RPE always increasing with load magnitude. There was a significant simple main effect of load for non-completers during each load condition: running ($F_{2,21}$=38.32, p<.001, η^2_p=.79) and forced-marching ($F_{2,21}$=36.98, p<.001, η^2_p=.78); with RPE always increasing with load magnitude. There was a significant simple main effect of locomotion for non-completers at BW ($F_{1,22}$=7.56, p=.01, η^2_p=.26) and +55%BW ($F_{1,22}$=7.52, p=.01, η^2_p=.26); with running having a greater RPE than forced-marching for both load magnitudes. Lastly, there was a significant simple main effect of group during running at +55%BW ($F_{1,22}$=4.34, p=.05, η^2_p=.17) with non-completers reporting higher overall RPEs than completers. Additionally, there was a significant simple main effect of group during forced-marching at +45%BW ($F_{1,22}$=9.14, p=.01, η^2_p=.29) with non-completers reporting higher overall RPEs than completers.

When assessing change in RPE (RPEC) from minute 0 to minute 10 there was no significant interaction between load and locomotion ($F_{2,50}$=0.03, p=.97, η^2_p=.001). However, with the addition of load carriage, change in RPE increased independently of locomotion as confirmed by the significant main effect of load ($F_{2,50}$=18.34, p<.001, η^2_p=.42), with BW (3±2) being significantly less (p<.001) than +45%BW (6±4) and +55%BW (6±3) [See Figure 3]. In contrast with overall RPE, there was no significant main effect of locomotion ($F_{1,25}$=3.11, p=.09, η^2_p=.11).

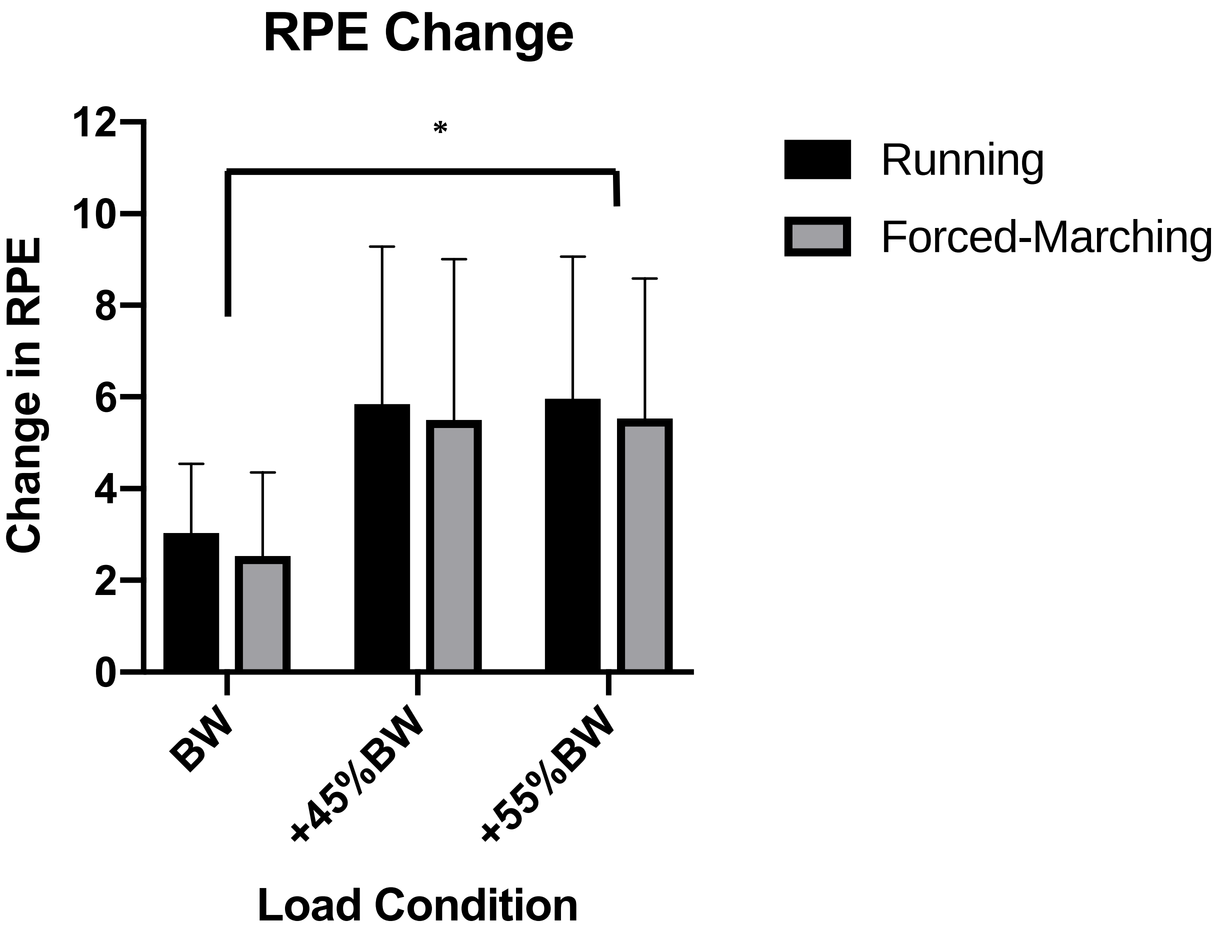


Figure 3. RPE Change
Change calculated as reported RPE at minute 0 subtracted from final reported RPE
**=Significant main effect of load*

When comparing the effect of sex on RPE change (RPEC) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,48}$=0.01, p=.99, η^2_p=.00). There were no significant two-way interactions of load and sex ($F_{2,48}$=0.15, p=.86, η^2_p=.01) or locomotion and sex ($F_{1,24}$=0.00, p=.96, η^2_p=.00). Additionally, there was no main effect of sex ($F_{1,24}$=0.00, p=.96, η^2_p=.00).

When comparing the effect of completion group on RPE change (RPEC) there was no significant three-way interaction between load, locomotion, and group ($F_{2,48}$=2.59, p=.11, η^2_p=.10). There were no significant two-way interactions of load and group ($F_{2,48}$=0.10, p=.91,

η^2_p=.00) or locomotion and group ($F_{1,24}$=2.06, p=.16, η^2_p=.08). Additionally, there was no main effect of group ($F_{1,24}$=0.00, p=.96, η^2_p=.00).

4.3 Goal Equivalent Manifold

4.3.1 Spatiotemporal Parameters

Refer to Table 5 for all mean and standard deviations of each spatiotemporal parameter's mean at each condition respectively. Additionally, four subjects (2 females and 2 males) were removed from the analysis due to data loss regarding the stride length. Moreover, two female subjects were unable to complete the +45%BW condition. One subject vocalized they were unable to complete the loaded conditions after executing forced-marching at +55%BW. The second subject reported low back pain and research staff observed that her heart rate was not dropping during the ten-minute rest periods. Therefore, at the discretion of the study team the subject was not required to perform the +45%BW load condition to avoid incurring an injury. Thus, n=20 (men=11, women=9) for the entire sample. When stratified by trial completion there were 5 non-completers and 15 completers (due to completion group factor being an exploratory analysis and Levene's Test of Equality of Error Variances rarely being violated the tertiary analysis was executed regardless of unequal group size).

Table 5. Spatiotemporal Parameter Means

Load	Locomotion	Variable	Total	Men	Women
BW	Running	Stride Length	1.33±.14	1.37±.13	1.27±.14
		Stride Time	0.76±.05	0.79±.05	0.76±.05
		Stride Speed	1.71±.21	1.74±.22	1.67±.20
	Forced-Marching	Stride Length	1.65±.14	1.72±.12	1.56±.11
		Stride Time	0.97±.08	1.00±.09	0.94±.06
		Stride Speed	1.71±.21	1.73±.22	1.67±.20
+45%BW	Running	Stride Length	1.32±.12	1.31±.09	1.34±.15
		Stride Time	0.80±.06	0.82±.05	0.77±.06
		Stride Speed	1.66±.20	1.59±.10	1.75±.26
	Forced-Marching	Stride Length	1.58±.09	1.61±.08	1.54±.08
		Stride Time	0.96±.10	1.02±.06	0.89±.10
		Stride Speed	1.66±.20	1.59±.10	1.75±.26
+55%BW	Running	Stride Length	1.25±.15	1.31±.11	1.17±.17
		Stride Time	0.81±.06	0.83±.06	0.79±.06
		Stride Speed	1.55±.22	1.60±.22	1.48±.21
	Forced-Marching	Stride Length	1.52±.13	1.59±.07	1.43±.14
		Stride Time	0.99±.10	1.01±.11	0.97±.08
		Stride Speed	1.55±.22	1.60±.22	1.49±.21

mean ± standard deviation
Stride length in m; stride time in seconds; stride speed in m/s

When assessing the mean of stride length there was a significant interaction between load and locomotion ($F_{2,38}$=7.19, p=.002, η^2_p=.27). There was only a significant simple main effect of load for forced-marching ($F_{2,18}$=6.94, p=.006, η^2_p=.44), with BW (1.65±0.14 m) having greater (p=.003) stride length than +55%BW (1.52±0.13 m). Additionally, there was a significant simple main effect of locomotion for every load condition with forced-marching having longer strides than running: BW ($F_{1,19}$=484.58, p<.001, η^2_p=.96); +45%BW ($F_{1,19}$=172.25, p<.001, η^2_p=.90); and +55%BW ($F_{1,19}$=356.27, p<.001, η^2_p=.95).

When assessing the mean of stride time there was no significant interaction between load and locomotion ($F_{2,38}$=1.95, p=.16, η^2_p=.09). There was no significant main effect for load

($F_{1.55,29.46}$=1.81, p=.19, η^2_p=.09). However, there was a significant main effect of locomotion ($F_{1,19}$=418.64, p<.001, η^2_p=.96), with forced-marching (0.97±0.09 s) having longer stride times than running (0.80±0.06 s). Additionally, there was a significant simple main effect of locomotion for every load condition with forced-marching having greater stride speed than running: BW ($F_{1,19}$=336.16, p<.001, η^2_p=.95); +45%BW ($F_{1,19}$=146.14, p<.001, η^2_p=.89); and +55%BW ($F_{1,19}$=239.04, p<.001, η^2_p=.93).

When assessing the mean of stride speed there was no significant interaction between load and locomotion ($F_{2,38}$=.93, p=.40, η^2_p=.05). There was no main effect of load ($F_{2,38}$=2.99, p=.06, η^2_p=.14). There was no main effect of locomotion ($F_{1,19}$=.42, p=.53, η^2_p=.02).

When analyzing the between group effect of sex for stride length mean there was no significant three-way interaction between load, locomotion and sex ($F_{2,36}$=2.81, p=.07, η^2_p=.14). Additionally, there was no two-way interaction between load and sex ($F_{2,36}$=1.81, p=.18, η^2_p=.09). However, there was a significant two-way interaction between locomotion and sex ($F_{1,18}$=5.91, p=.03, η^2_p=.25). There was a significant simple main effect of locomotion for women ($F_{1,18}$=210.06, p<.001, η^2_p=.92) and men ($F_{1,18}$=385.95, p<.001, η^2_p=.96) with forced-marching having longer strides than running for both sexes. Lastly, there was a significant simple main effect of sex for running ($F_{1,18}$=6.22, p=.03, η^2_p=.26) and forced-marching ($F_{1,18}$=20.74, p<.001, η^2_p=.54), with men having longer strides than women for both locomotion patterns.

When analyzing the effect of sex for mean stride time there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=2.36, p=.11, η^2_p=.12). There was no significant two-way interaction between load and sex ($F_{2,36}$=2.10, p=.14, η^2_p=.10). However, there was a significant two-way interaction between locomotion and sex ($F_{1,18}$=473.47, p<.001, η^2_p=.96). There was a significant simple main effect of locomotion for women ($F_{1,18}$=176.97, p<.001,

η^2_p=.91) and men ($F_{1,18}$=314.35, p<.001, η^2_p=.95) with forced-marching having longer stride times than running for both sexes. Lastly, there was only a significant simple main effect of sex for forced-marching ($F_{1,18}$=5.77, p=.03, η^2_p=.24) with men (1.01±0.07 s) having longer stride times than women (0.93±0.07 s).

When analyzing the effect of sex for mean stride speed there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.42, p=.66, η^2_p=.02). There was no significant two-way interaction between load and sex ($F_{1.83,32.91}$=2.49, p=.10, η^2_p=.12). However, there was no significant two-way interaction between locomotion and sex ($F_{1,18}$=2.46, p=.13, η^2_p=.12). There was no main effect of sex ($F_{1,18}$=0.01, p=.92, η^2_p=.00).

Lastly, when examining the between groups effect of ability to complete the entire ten-minute trial for mean stride length there was no significant three-way interaction between load, locomotion and group ($F_{2,36}$=1.01, p=.37, η^2_p=.05). There were no significant two-way interaction between load and group ($F_{2,36}$=1.08, p=.35, η^2_p=.06) or locomotion and group ($F_{1,18}$=0.82, p=.38, η^2_p=.04). There was no significant main effect of group ($F_{1,18}$=0.01, p=.93, η^2_p=.00).

When examining the effect of completion group on mean stride time there was no significant three-way interaction between load, locomotion and group ($F_{2,36}$=0.85, p=.43, η^2_p=.05). There were no significant two-way interactions between load and group ($F_{2,36}$=1.44, p=.25, η^2_p=.07) or locomotion and group ($F_{1,18}$=0.60, p=.45, η^2_p=.03). There was no significant main effect of group ($F_{1,18}$=0.48, p=.50, η^2_p=.03).

When examining the effect of completion group on mean stride speed there was no significant three-way interaction between load, locomotion and group ($F_{2,36}$=0.02, p=.98, η^2_p=.00). There were no significant two-way interactions between load and group ($F_{2,36}$=1.38, p=.26,

η^2_p=.07) or locomotion and group ($F_{1,18}$=2.43, p=.14, η^2_p=.12). There was no significant main effect of group ($F_{1,18}$=0.68, p=.42, η^2_p=.04).

Refer to Table 6 for all mean and standard deviations of each spatiotemporal characteristic's variance at each condition respectively. When assessing the variance of stride length there was no significant interaction between load and locomotion ($F_{1.18,22.49}$=0.88, p=.38, η^2_p=.04). Furthermore, there were no significant main effects for load ($F_{1,13,21.52}$=1.16, p=.30, η^2_p=.06) or locomotion ($F_{1,19}$=0.49, p=.50, η^2_p=.03).

Table 6. Spatiotemporal Parameter Variance

Load	Locomotion	Variable	Total	Men	Women
BW	Running	Stride Length	.034±.010	.038±.009	.031±.010
		Stride Time	.016±.007	.017±.007	.015±.007
		Stride Speed	.040±.010	.044±.012	.036±.005
	Forced-Marching	Stride Length	.026±.009	.028±.010	.024±.006
		Stride Time	.016±.005	.016±.004	.015±.006
		Stride Speed	.023±.010	.027±.012	.019±.004
+45%BW	Running	Stride Length	.038±.017	.041±.016	.034±.018
		Stride Time	.019±.012	.022±.014	.016±.008
		Stride Speed	.042±.014	.045±.017	.038±.010
	Forced-Marching	Stride Length	.037±.048	.029±.007	.046±.073
		Stride Time	.021±.021	.019±.005	.024±.032
		Stride Speed	.021±.021	.019±.005	.024±.032
+55%BW	Running	Stride Length	.036±.011	.038±.012	.034±.009
		Stride Time	.020±.009	.021±.010	.020±.009
		Stride Speed	.036±.007	.039±.007	.033±.004
	Forced-Marching	Stride Length	.035±.019	.038±.020	.032±.017
		Stride Time	.024±.013	.024±.009	.024±.017
		Stride Speed	.026±.010	.030±.012	.022±.005

mean ± standard deviation
Stride length in m; stride time in seconds; stride speed in m/s

When assessing the variance of stride time there was no significant interaction between load and locomotion ($F_{1.31,24.81}$=0.99, p=.35, η^2_p=.05). As load increased, stride time variance increased independent of locomotion pattern evidenced by the significant main effect of load ($F_{1.10,20.98}$=6.50, p=.02, η^2_p=.26); with post-hoc pairwise comparisons revealing that +55%BW (0.022±0.011 seconds) had greater (p<.001) stride time variance than BW (0.015± 0.006 seconds). However, there was no significant main effect of locomotion ($F_{1,19}$=1.08, p=.31, η^2_p=.05).

When assessing the variance of stride speed there was no significant interaction between load and locomotion ($F_{1.42,26.96}=2.83$, $p=.09$, $\eta^2_p=.13$). Additionally, there was no significant main effect of load ($F_{1.52,28.91}=0.01$, $p=.98$, $\eta^2_p=.00$). In contrast with stride time, running (0.039±0.010 m/s) exhibited greater stride speed variance than forced-marching (0.024±0.014 m/s) confirmed by the significant main effect of locomotion ($F_{1,19}=63.20$, $p<.001$, $\eta^2_p=.77$).

When analyzing the between groups effect of sex on spatiotemporal variance, there were no significant findings for any of the spatiotemporal outcomes. For stride length the three-way interaction between load, locomotion, and sex was non-significant ($F_{2,36}=1.95$, $p=.16$, $\eta^2_p=.10$), the same for two-way interactions of load and sex ($F_{2,36}=0.99$, $p=.38$, $\eta^2_p=.05$), and locomotion and sex ($F_{1,18}=1.22$, $p=.29$, $\eta^2_p=.06$). Lastly, the main effect of sex was not significant either ($F_{1,18}=0.21$, $p=.65$, $\eta^2_p=.01$).

For stride time no significant findings were observed for: the three-way interaction between load, locomotion, and sex ($F_{2,36}=1.60$, $p=.22$, $\eta^2_p=.08$); two-way interactions of load and sex ($F_{2,36}=0.13$, $p=.88$, $\eta^2_p=.01$) or locomotion and sex ($F_{1,18}=1.31$, $p=.27$, $\eta^2_p=.07$); or main effect of sex ($F_{1,18}=0.18$, $p=.68$, $\eta^2_p=.01$).

For stride speed no significant findings were observed for: the three-way interaction between load, locomotion, and sex ($F_{2,36}=2.05$, $p=.14$, $\eta^2_p=.10$); two-way interactions of load and sex ($F_{2,36}=1.07$, $p=.35$, $\eta^2_p=.06$) or locomotion and sex ($F_{1,18}=.06$, $p=.47$, $\eta^2_p=.03$); or main effect of sex ($F_{1,18}=3.13$, $p=.09$, $\eta^2_p=.15$).

When assessing the between groups effect of ability to complete the entire ten-minute trial on spatiotemporal variance there were many significant findings. For stride length variance there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}=2.33$, $p=.11$, $\eta^2_p=.11$). There was also no significant two-way interaction between load and group ($F_{2,36}=1.56$,

$p=.22$, $\eta^2_p=.08$). However, there was a significant two-way interaction between load and locomotion ($F_{1,18}=6.77$, $p=.02$, $\eta^2_p=.27$). There was no significant simple main effect of locomotion for either completers ($F_{1,18}=3.96$, $p=.06$, $\eta^2_p=.18$) or non-completers ($F_{1,18}=3.44$, $p=.08$, $\eta^2_p=.16$). There was a significant simple main effect of group for forced-marching only ($F_{1,18}=7.40$, $p=.01$, $\eta^2_p=.29$) with non-completers (0.052±0.046 m) having greater stride length variance than completers (0.026±0.007 m).

For stride time variance there was no significant three-way interaction between load, locomotion, and group ($F_{1.36,24.49}=3.33$, $p=.07$, $\eta^2_p=.16$). There were no significant two-way interactions between load and group ($F_{1.11,19.88}=2.16$, $p=.16$, $\eta^2_p=.11$) or locomotion and group ($F_{1,18}=3.48$, $p=.08$, $\eta^2_p=.16$). There was no significant main effect of group ($F_{1,18}=2.68$, $p=.12$, $\eta^2_p=.13$).

For stride speed variance there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}=1.46$, $p=.25$, $\eta^2_p=.08$). Additionally, there were no significant two-way interactions between load and group ($F_{2,36}=1.35$, $p=.27$, $\eta^2_p=.07$) or locomotion and group ($F_{1,18}=2.87$, $p=.11$, $\eta^2_p=.14$). Additionally, there was no significant main effect of group ($F_{1,18}=2.32$, $p=.15$, $\eta^2_p=.11$).

4.3.2 Relative Variability

Refer to Table 7 for all mean and standard deviations for all GEM related outcomes. When analyzing relative variability of the entire trial length (**Specific Aim 1.1**) there was no significant interaction between load and locomotion ($F_{2,38}=0.41$, $p=.67$, $\eta^2_p=.02$). However, as load increased relative variability increased independent of locomotion pattern as evidenced by the significant main effect of load ($F_{1.38,26.15}=7.65$, $p=.01$, $\eta^2_p=.29$); with post-hoc analysis revealing that

+55%BW (2.07±0.74) had significantly (p=.01) greater relative variability than BW (1.72±0.35). In addition, forced-marching (2.08±0.57) exhibited greater relative variability compared to running (1.67±0.63) as confirmed by the significant main effect of locomotion ($F_{1,19}$=14.40, p<.001, η^2_p=.43).

Table 7. Relative Variability

Load	Locomotion	Portion	Total	Men	Women
BW	Running	Total	1.56±.24	1.53±.18	1.58±.30
		Beginning	1.52±.16	1.52±.14	1.53±.18
		End	1.49±.31	1.42±.15	1.56±.41
	Forced-Marching	Total	1.91±.45	1.73±.50	2.09±.32
		Beginning	1.95±.45	1.71±.45	2.20±.29
		End	1.73±.52	1.57±.58	1.89±.42
+45%BW	Running	Total	1.66±.25	1.72±.24	1.60±.25
		Beginning	1.62±.36	1.76±.41	1.46±.22
		End	1.56±.38	1.60±.40	1.51±.36
	Forced-Marching	Total	2.00±.37	1.95±.37	2.07±.39
		Beginning	1.85±.41	1.91±.33	1.79±.51
		End	1.87±.39	1.83±.38	1.92±.41
+55%BW	Running	Total	1.90±.60	1.79±.58	2.01±.62
		Beginning	1.66±.25	1.67±.25	165±.27
		End	1.83±.73	1.78±.72	1.88±.78
	Forced-Marching	Total	2.29±.88	2.04±.42	2.55±1.15
		Beginning	2.18±.63	2.03±.45	2.33±.77
		End	2.09±1.01	1.92±.46	2.27±1.37

mean ± standard deviation
BW=Body weight (no load); +45%BW=An additional 45% of BW; +55%BW=An additional 55% of BW;
Total=Entire trial; Beginning=First 30% of the trial only; End=Last 30% of the trial only;

Following the assessment of relative variability of the entire trial the influence of time on relative variability (first 30% of the trial vs. the final 30% of the trial) was analyzed. There was no three-way interaction between load, locomotion, and time ($F_{2,38}$=0.89, p=.42, η^2_p=.05). Furthermore, there were no two-way interactions between load and time ($F_{2,38}$=0.64, p=.53, η^2_p=.03) or locomotion and time ($F_{1,19}$=0.53, p=.48, η^2_p=.03). Moreover, time did not affect relative variability ($F_{1,19}$=1.07, p=.31, η^2_p=.05).

A secondary exploratory analysis was conducted to determine if sex effects task execution (**Specific Aim 1.2**). For the entire trial relative variability there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.36, p=.70, η^2_p=.02). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=1.82, p=.18, η^2_p=.09) or locomotion and sex ($F_{1,18}$=4.10, p=.06, η^2_p=.19). Additionally, there was no significant main effect of sex ($F_{1,18}$=2.11, p=.16, η^2_p=.11).

For the beginning phase of the trial relative variability there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.52, p=.60, η^2_p=.03). There was a significant two-way interaction between load and sex ($F_{2,36}$=4.18, p=.02, η^2_p=.19). There was significant simple main effect of load for women ($F_{2,17}$=4.78, p=.02, η^2_p=.36); with +55%BW (2.06±0.52) having significantly (p=.02) greater relative variability than +45%BW (1.62±0.37). There was also a significant simple main effect of load for men ($F_{2,17}$=3.86, p=.04, η^2_p=.31); post-hoc pairwise comparisons failed to reveal any significant differences between load conditions. There was only a significant simple main effect of sex at BW ($F_{1,18}$=5.08, p=.04, η^2_p=.22); with women (1.83±0.24) having greater relative variability than men (1.61±0.30). There were no significant simple main effects of sex for +45%BW ($F_{1,18}$=2.98, p=.10, η^2_p=.14) or +55%BW ($F_{1,18}$=1.80, p=.20, η^2_p=.09). Additionally, there was a significant two-way interaction between locomotion and sex ($F_{1,18}$=4.41, p=.05, η^2_p=.20). There was no significant simple main effect of sex for running ($F_{1,18}$=1.32, p=.27, η^2_p=.07) or forced-marching ($F_{1,18}$=3.26, p=.09, η^2_p=.15). There was a significant simple main effect for locomotion for women ($F_{1,18}$=22.57, p<.001, η^2_p=.56); with forced-marching (2.13±0.52) having greater relative variability than running (1.54±0.22). There was also a significant simple main effect of locomotion for men ($F_{1,18}$=4.51, p=.05, η^2_p=.20); with forced-marching (1.88±0.41) having greater relative variability than running (1.65±0.27).

For the end phase of the trial relative variability there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.24, p=.78, η^2_p=.01). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=0.69, p=.51, η^2_p=.04) or locomotion and sex ($F_{1,18}$=2.26, p=.15, η^2_p=.11). Additionally, there was no significant main effect of sex ($F_{1,18}$=0.72, p=.41, η^2_p=.04).

The final exploratory analysis examining the differences between participants who could complete the ten-minute trial versus those who could not (**Specific Aim 1.3**), showed no significant three-way interaction between load, locomotion and group when observing the entire trial ($F_{2,36}$=1.86, p=.17, η^2_p=.09). Additionally, there was no significant interaction between locomotion and group ($F_{1,18}$=0.58, p=.47, η^2_p=.03). However, there was a significant interaction between load and group ($F_{2,36}$=3.96, p=.03, η^2_p=.18). There was a significant simple main effect of load for completers ($F_{2,17}$=4.34, p=.03, η^2_p=.34); with BW (1.67±0.28) having less (p=.03) relative variability than +45%BW (1.85±0.30). There was also a significant simple main effect of load for non-completers ($F_{2,17}$=6.26, p=.01, η^2_p=.42); with BW (1.82±0.49) having less (p=.03) relative variability than +55%BW (2.42±0.92). There were no significant simple main effects of group at any load condition.

When assessing relative variability differences of completion groups during the beginning phase of the trial there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}$=1.73, p=.19, η^2_p=.09). There was no significant two-way interaction of load and group ($F_{2,36}$=2.13, p=.13, η^2_p=.11) or locomotion and group ($F_{1,18}$=0.15, p=.70, η^2_p=.01). There was also no main effect of group ($F_{1,18}$=0.09, p=.77, η^2_p=.01).

Lastly, when assessing relative variability differences of completion groups during the end phase of the trial there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}$=0.99, p=.38, η^2_p=.05). There was no significant two-way interaction of load and group

($F_{2,36}$=2.08, p=.14, η^2_p=.10) or locomotion and group ($F_{1,18}$=1.72, p=.21, η^2_p=.09). There was also no main effect of group ($F_{1,18}$=2.60, p=.12, η^2_p=.13).

4.3.3 GEM Subspace Variance

Refer to Table 8 for all means and standard deviations of variance tangential and perpendicular (subspace variance) to the goal manifold for each condition. When assessing the variance of coordinate distance from the goal manifold and the interactive effects of load and locomotion there was no three-way interaction between load, locomotion, and direction ($F_{2,38}$=0.07, p=.94, η^2_p=.00). There was a significant interaction between load and direction ($F_{2,36}$=10.39, p<.001, η^2_p=.35). There was a significant simple main effect of load for tangential variability ($F_{2,18}$=7.69, p=.004, η^2_p=.46); with post hoc pairwise comparisons revealing that +55%BW (1.25±0.06) had greater (p=.002) variability than BW (1.21±0.07). Additionally, there was a significant simple main effect of load for perpendicular variability ($F_{2,18}$=8.25, p=.003, η^2_p=.48); with post hoc pairwise comparisons revealing that BW (.73±0.11) had greater (p=.002) variability than +55%BW (.65±0.13). Lastly, there were significant simple main effects for direction at each load condition with tangential variability always being greater than perpendicular variability: BW ($F_{1,19}$=267.63, p<.01, η^2_p=.93); +45%BW ($F_{1,19}$=575.97, p<.001, η^2_p=.97); and +55%BW ($F_{1,19}$=508.32, p<.001, η^2_p=.96). There was a significant interaction between locomotion and direction ($F_{1,19}$=18.69, p<.001, η^2_p=.50). There was a significant simple main effect of locomotion for tangential variability ($F_{1,19}$=18.43, p<.001, η^2_p=.49), with forced-marching having more variability than running. There was also a significant simple main effect of locomotion for perpendicular variability ($F_{1,19}$=18.61, p<.001, η^2_p=.50), with running having greater variability than forced-marching. Moreover, tangential direction exhibited more variability than perpendicular for both locomotion patterns evidenced by the significant simple main effects of running ($F_{1,19}$=519.36, p<.001, η^2_p=.97) and forced-marching ($F_{1,19}$=18.43, p<.001, η^2_p=.49). Lastly, there was a main effect for direction ($F_{1,19}$=658.003, p<.001, η^2_p=.97) with variability tangential (1.23±0.06) to the goal manifold being greater than perpendicular to it (0.69±0.11).

Table 8. Subspace Variance

Load	Locomotion	Variable	Portion	Total	Men	Women
BW	Running	Tangential	Total	1.18±.05	1.18±.02	1.18±.06
			Beginning	1.08±.12	1.07±.15	1.10±.09
			End	1.16±.20	1.13±.11	1.20±.26
		Perpendicular	Total	0.77±.08	0.78±.06	0.76±.10
			Beginning	0.72±.11	0.71±.12	0.73±.11
			End	0.79±.11	0.80±.11	0.78±.11
	Forced-Marching	Tangential	Total	1.23±.08	1.20±.09	1.27±.04
			Beginning	1.17±.16	1.12±.16	1.23±.14
			End	1.16±.18	1.11±.20	1.21±.16
		Perpendicular	Total	0.67±.13	0.73±.15	0.62±.07
			Beginning	0.62±.12	0.68±.13	0.57±.07
			End	0.71±.18	0.77±.22	0.66±.10
+45%BW	Running	Tangential	Total	1.21±.05	1.22±.04	1.19±.05
			Beginning	1.05±.21	1.08±.22	1.00±.21
			End	1.19±.34	1.20±.33	1.18±.38
		Perpendicular	Total	0.73±.08	0.72±.07	0.76±.08
			Beginning	0.67±.17	0.65±.18	0.70±.15
			End	0.76±.12	0.76±.13	0.78±.10
	Forced-Marching	Tangential	Total	1.26±.05	1.25±.05	1.26±.05
			Beginning	1.14±.22	1.21±.09	1.05±.30
			End	1.21±.21	1.20±.15	1.22±.28
		Perpendicular	Total	0.64±.09	0.66±.10	0.63±.09
			Beginning	0.63±.15	0.65±.10	0.60±.20
			End	0.66±.10	0.67±.09	0.65±.12
+55%BW	Running	Tangential	Total	1.23±.07	1.21±.07	1.25±.06
			Beginning	1.01±.18	1.04±.15	0.97±.20
			End	1.26±.27	1.30±.21	1.21±.33
		Perpendicular	Total	0.69±.13	0.71±.13	0.66±.13
			Beginning	0.62±.14	0.64±.14	0.60±.14
			End	0.73±.14	0.77±.13	0.68±.13
	Forced-Marching	Tangential	Total	1.28±.05	1.26±.05	1.29±.05
			Beginning	1.12±.23	1.13±.24	1.10±.23
			End	1.20±.26	1.28±.31	1.12±.19
		Perpendicular	Total	0.60±.12	0.64±.10	0.56±.13
			Beginning	0.54±.12	0.56±.10	0.51±.13
			End	0.63±.16	0.69±.15	0.57±.17

mean ± standard deviation

Since a significant difference was observed between tangential and perpendicular variability 3x2x2 (load x locomotion x time) RMANOVA were performed for each direction separately. For tangential direction variability there was no three-way interaction between load, locomotion, and time ($F_{2,38}$=0.33, p=.72, η^2_p=.02). There were no significant two-way interactions between load and time ($F_{2,38}$=1.03, p=.37, η^2_p=.00) or locomotion and time ($F_{1,19}$=1.51, p=.23, η^2_p=.07). Lastly, there was no significant main effect of time either ($F_{1,19}$=4.19, p=.06, η^2_p=.18).

For perpendicular variability there was no three-way interaction between load, locomotion, and time ($F_{2,38}$=1.07, p=.35, η^2_p=.05). There were no significant two-way interactions between load and time ($F_{2,38}$=0.84, p=.44, η^2_p=.04) or locomotion and time ($F_{1,19}$=0.28, p=.61, η^2_p=.01). However, perpendicular variability was greater in the final 30% (0.72±0.14) of the trial compared to the beginning 30% (0.64±0.14) of the trial confirmed by the significant main effect of time ($F_{1,19}$=21.16, p<.001, η^2_p=.53). See Figures 4 and 5 for exemplar GEM plots of two participants, notice that the spread of points is much larger along the goal manifold (tangential) than perpendicular to it.

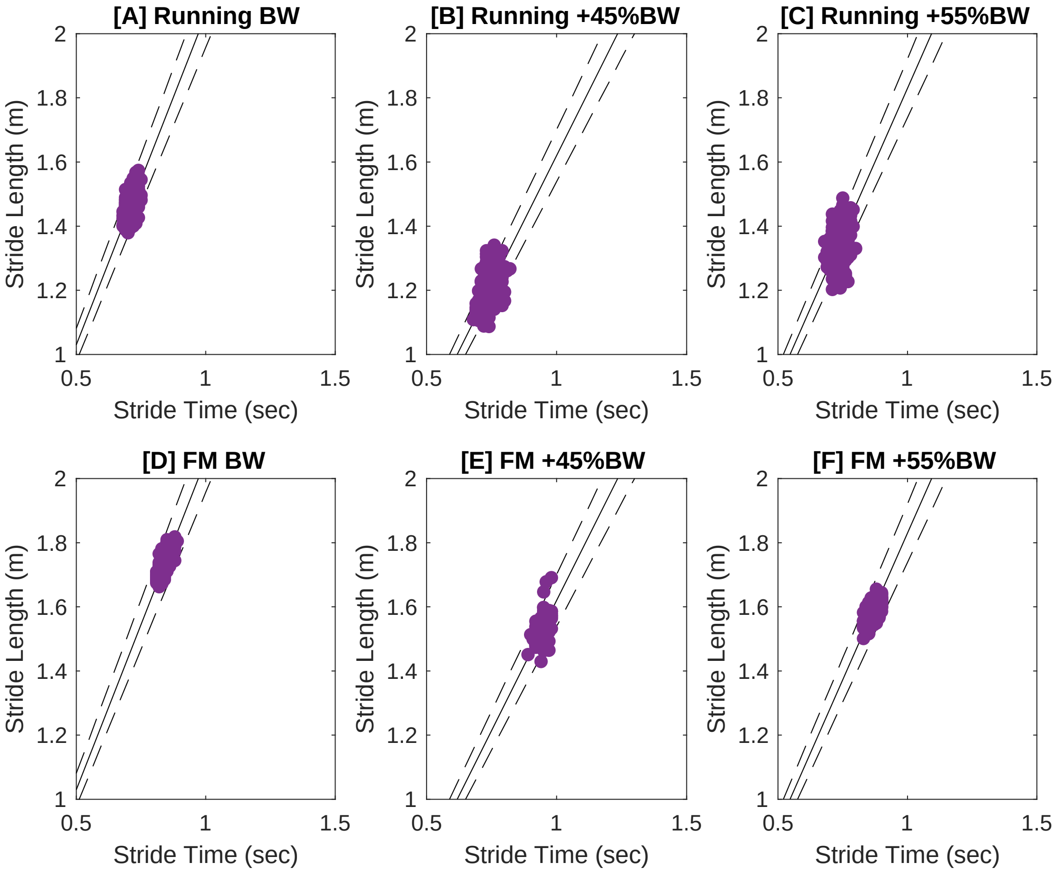


Figure 4. Exemplar GEM Plot (Completer)

The solid line represents the goal manifold (the trial velocity), with the dashed lines representing a +/- 5% error from the goal manifold. The above figures are those of an individual who completed the full ten-minute trial for each condition. In contrast with the participant from Figure 5, qualitative inspection reveals less variation along the goal manifold, especially for the loaded conditions (B, C, E, and F). Moreover, there was a greater spread perpendicular to the goal manifold during the loaded conditions, with more falling outside of the 5% error tolerances.

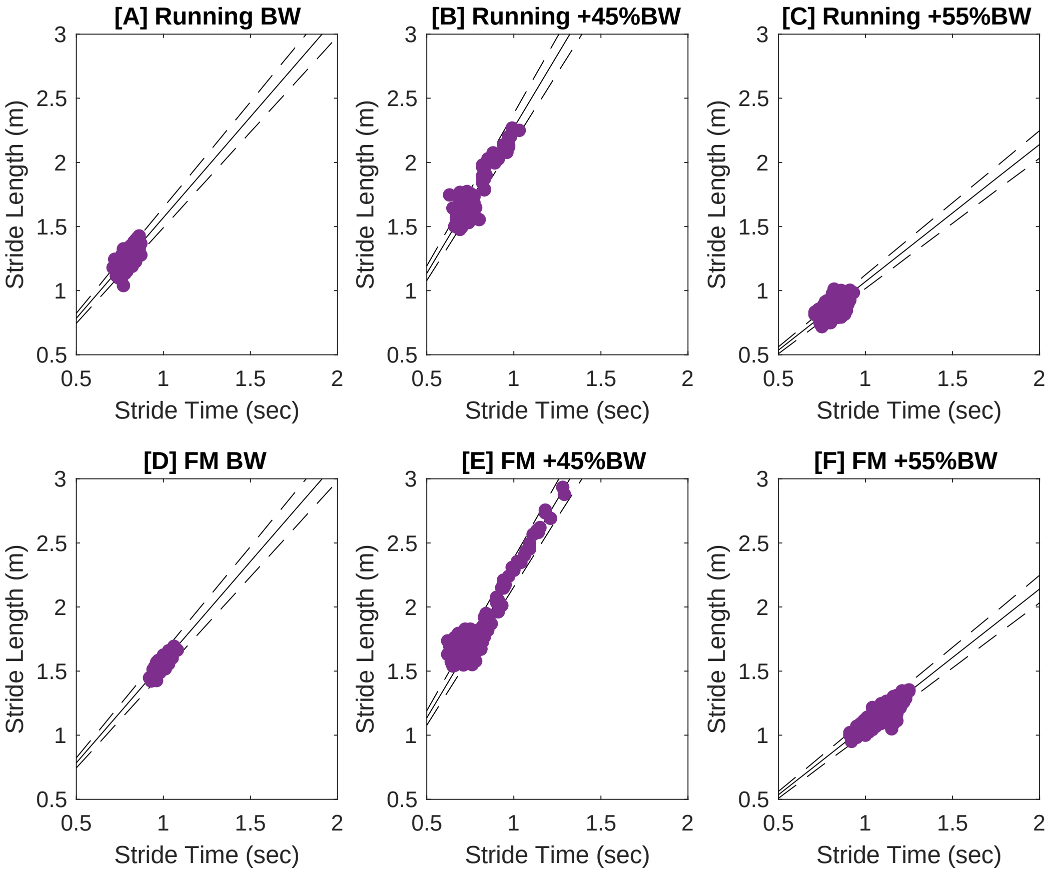


Figure 5. Exemplar GEM Plot (Non-completer)

The solid line represents the goal manifold (the trial velocity), with the dashed lines representing a +/- 5% error from the goal manifold. This specific participant was unable to complete the full ten-minute trials of the +45%BW load condition. As can be observed in B and E, there is greater variation tangential to the goal manifold (the solid line). Some of these variations are a result of the individual drifting towards the back end of the treadmill and essentially hopping forward to ensure they stay on (refer to E). Thus, this results in a new variation that may accomplish the task goal (maintaining velocity) but wouldn't meet the task constraints (maintaining a walking gait pattern).

4.3.4 Stride Regulation

Refer to Table 9 for all mean and standard deviations of alpha coefficients of GEM coordinate time series tangential and perpendicular to the goal manifold for each condition (**Specific Aim 2.1**). When assessing stride regulation of tangential variability there was no significant interaction between load and locomotion ($F_{2,38}$=0.59, p=.56, η^2_p=.03). Additionally, load had no impact on tangential variability stride regulation confirmed by no significant main effect of load ($F_{1.47,27.98}$=1.42, p=.25, η^2_p=.07). However, forced-marching (0.85±0.11) had a greater alpha coefficient than running (0.81±0.11) independent of load condition ($F_{1,19}$=4.59, p=.05, η^2_p=.19).

Table 9. Goal Equivalent Manifold Regulation Alpha Coefficients

Load	Locomotion	Variable	Portion	Total	Men	Women
BW	Running	Tangential α	Total	.78±.12	.77±.10	.79±.14
			Beginning	.77±.15	.72±.11	.81±.18
			End	.69±.17	.63±.13	.75±.19
		Perpendicular α	Total	.65±.09	.66±.09	.65±.09
			Beginning	.61±.10	.60±.10	.62±.10
			End	.56±.14	.54±.15	.58±.14
	Forced-Marching	Tangential α	Total	.85±.10	.86±.12	.84±.09
			Beginning	.82±.12	.82±.13	.83±.11
			End	.73±.12	.67±.12	.80±.10
		Perpendicular α	Total	.60±.12	.61±.11	.59±.13
			Beginning	.56±.12	.56±.14	.56±.10
			End	.55±.10	.55±.10	.55±.12
+45%BW	Running	Tangential α	Total	.82±.10	.81±.11	.82±.08
			Beginning	.77±.17	.78±.15	.76±.20
			End	.69±.13	.71±.16	.66±.09
		Perpendicular α	Total	.63±.10	.64±.13	.62±.06
			Beginning	.55±.18	.53±.16	.58±.21
			End	.51±.17	.54±.21	.47±.11
	Forced-Marching	Tangential α	Total	.86±.11	.80±.08	.92±.12
			Beginning	.86±.16	.86±.11	.87±.21
			End	.77±.19	.74±.10	.80±.26
		Perpendicular α	Total	.58±.10	.53±.09	.64±.08
			Beginning	.57±.10	.56±.10	.59±.10
			End	.50±.12	.45±.07	.56±.13
+55%BW	Running	Tangential α	Total	.83±.11	.79±.09	.88±.12
			Beginning	.77±.14	.74±.11	.80±.18
			End	.68±.20	.67±.17	.69±.24
		Perpendicular α	Total	.61±.10	.59±.10	.62±.10
			Beginning	.51±.15	.51±.13	.51±.18
			End	.46±.16	.49±.12	.43±.19
	Forced-Marching	Tangential α	Total	.87±.13	.78±.10	.95±.09
			Beginning	.86±.16	.82±.18	.89±.14
			End	.77±.17	.73±.18	.82±.14
		Perpendicular α	Total	.54±.10	.51±.11	.57±.09
			Beginning	.53±.11	.55±.15	.51±.06
			End	.47±.12	.51±.13	.43±.08

mean ± standard deviation
α = alpha coefficient derived from detrended fluctuation analysis;
BW=Body weight (no load); +45%BW=An additional 45% of BW; +55%BW=An additional 55% of BW;
Total=Entire trial; Beginning=First 30% of the trial only; End=Last 30% of the trial only;

α=.5 represents stochastic (random) control; α<.5 represents anti-persistent regulation (strict control); α>.5 represents persistent behavior (looser/weaker control); α≥1.5 represents over regularity (no control)

When assessing stride regulation of perpendicular variability ('bad' variability) there was no significant interaction between load and locomotion ($F_{2,38}$=0.09, p=.91, η^2_p=.005). As load increased alpha coefficients decreased as evidenced by the significant main effect of load ($F_{2,38}$=3.54, p=.04, η^2_p=.16); post-hoc pairwise comparisons failed to reveal any significant differences between the three load conditions. Additionally, forced-marching (0.58±0.11) yielded a lower alpha coefficient compared to running (0.63±0.10) confirmed by the significant main effect of locomotion ($F_{1,19}$=5.98, p=.02, η^2_p=.24).

When assessing the influence of time along with load and locomotion on tangential variability stride regulation there was no three-way interaction ($F_{2,38}$=0.23, p=.79, η^2_p=.01). Additionally, there were no two-way interactions either for locomotion by time ($F_{1,19}$=0.01, p=.94, η^2_p=.00) or load by time ($F_{2,38}$=0.07, p=.93, η^2_p=.004). However, there was a main effect of time ($F_{1,19}$=20.48, p<.001, η^2_p=.52), with the beginning (0.80±0.15) portion having greater alpha coefficients than the end portion (0.72±0.16).

When additionally assessing the influence of time for perpendicular variability stride regulation there was no three-way interaction ($F_{2,38}$=0.86, p=.43, η^2_p=.04). Additionally, there were no two-way interactions for load by time ($F_{2,38}$=0.89, p=.42, η^2_p=.05) or locomotion by time ($F_{1,19}$=0.14, p=.71, η^2_p=.007). Similar to tangential variability, there was a significant main effect of time on perpendicular variability ($F_{1,19}$=7.30, p=.01, η^2_p=.28), with the beginning portion (0.56±0.13) having greater alpha coefficients than the end portion (0.51±0.14).

A secondary exploratory analysis was conducted to determine if sex effects stride regulation (**Specific Aim 2.2**). For the entire trial tangential variability stride regulation there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=2.50, p=.10, η^2_p=.12). There was no significant two-way interaction between locomotion and sex ($F_{1,18}$=1.53, p=.23, η^2_p=.08). However, there was a significant two-way interaction between load and sex ($F_{2,36}$=6.48, p=.004, η^2_p=.27). There was a significant simple main effect of load for women ($F_{2,17}$=5.43, p=.02, η^2_p=.39), with BW (0.81±0.12) having significantly (p=.03, p=.01) lower alpha coefficients than +45%BW (0.87±0.10) and +55%BW (0.92±0.11) load conditions. By contrast there was no significant simple main effect of load for men ($F_{2,17}$=.42, p=.67, η^2_p=.05). The only significant simple main effect of sex was at +55%BW ($F_{1,18}$11.01, p=.004, η^2_p=.38), with women (0.92±0.11) having greater alpha coefficients than men (0.79±0.10). Simple main effects of sex were not significant for BW ($F_{1,18}$=0.01, p=.91, η^2_p=.001) and +45%BW ($F_{1,18}$=4.21, p=.06, η^2_p=.19).

For the entire trial perpendicular variability stride regulation there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=2.24, p=.12, η^2_p=.11). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=1.37, p=.27, η^2_p=.07) or locomotion and sex ($F_{1,18}$=1.59, p=.22, η^2_p=.08). Lastly, there was no significant main effect of sex ($F_{1,18}$=1.61, p=.22, η^2_p=.08).

For the beginning phase of the trial tangential variability stride regulation there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.62, p=.54, η^2_p=.03). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=0.87, p=.43, η^2_p=.05) or locomotion and sex ($F_{1,18}$=0.03, p=.89, η^2_p=.002). Additionally, there was no significant main effect of sex ($F_{1,18}$=0.90, p=.36, η^2_p=.05).

For the end phase of the trial tangential variability stride regulation there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.44, p=.65, η^2_p=.04). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=1.97, p=.16, η^2_p=.10) or locomotion and sex ($F_{1,18}$=1.68, p=.21, η^2_p=.09). Additionally, there was no significant main effect of sex ($F_{1,18}$=1.88, p=.19, η^2_p=.10).

For the beginning phase of the trial perpendicular variability stride regulation there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.02, p=.98, η^2_p=.001). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=0.51, p=.61, η^2_p=.03) or locomotion and sex ($F_{1,18}$=0.48, p=.50, η^2_p=.03). Additionally, there was no significant main effect of sex ($F_{1,18}$=0.17, p=.69, η^2_p=.009).

For the end phase of the trial perpendicular variability stride regulation there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=2.04, p=.15, η^2_p=.10). Moreover, there was no significant two-way interaction between load and sex ($F_{2,36}$=2.30, p=.12, η^2_p=.11) or locomotion and sex ($F_{1,18}$=1.67, p=.28, η^2_p=.07). Additionally, there was no significant main effect of sex ($F_{1,18}$=0.20, p=.66, η^2_p=.01).

Lastly, the tertiary exploratory analysis of group (completer vs non-completer) effects on stride regulation were examined for the entire trial (**Specific Aim 2.3**). For regulation tangential to the goal manifold there was no significant three-way interaction of load, locomotion, and group ($F_{2,36}$=2.56, p=.09, η^2_p=.13). Additionally, there were no significant two-way interactions between load and group ($F_{2,36}$=0.72, p=.49, η^2_p=.04) or locomotion and group ($F_{1,18}$=0.05, p=.82, η^2_p=.00). Finally, there was no significant main effect of group ($F_{1,18}$=0.79, p=.39, η^2_p=.04).

For regulation perpendicular to the goal manifold there was no significant three-way interaction of load, locomotion, and group ($F_{2,36}$=0.52, p=.60, η^2_p=.03). Additionally, there were no significant two-way interactions between load and group ($F_{2,36}$=1.15, p=.33, η^2_p=.06) or locomotion and group ($F_{1,18}$=1.97, p=.18, η^2_p=.01). Finally, there was no significant main effect of group ($F_{1,18}$=.21, p=.65, η^2_p=.01).

4.4 Complexity

4.4.1 Spatiotemporal Long-Range Correlation

Refer to Table 10 for all mean and standard deviations of spatiotemporal related alpha coefficients (α) [**Specific Aim 3.1**]. For stride time (ST) complexity there was no significant interaction between load and locomotion ($F_{2,46}$=1.59, p=.22, η^2_p=.07). Additionally, there were no significant main effects for load ($F_{2,46}$=0.30, p=.75, η^2_p=.01) or locomotion ($F_{1,23}$=1.65, p=.21, η^2_p=.07).

Table 10. Spatiotemporal Alpha Coefficients

Load	**Locomotion**	**Variable**	**Total**	**Men**	**Women‡**
BW (No additional load)	Running	SL	0.68±0.10	0.68±0.10	0.68±0.14
		ST	0.79±0.11	0.79±0.11	0.80±0.12
		SS	0.35±0.10	0.36±0.13	0.34±0.04
	Forced-Marching	SL	0.80±0.11*	0.80±0.14	0.81±0.08
		ST	0.82±0.10	0.81±0.10	0.83±0.11
		SS	0.41±0.11*	0.42±0.11	0.41±0.11
+45%BW	Running	SL	0.75±0.10	0.75±0.10	0.76±0.11
		ST	0.84±0.11	0.84±0.13	0.84±0.09
		SS	0.37±0.11	0.37±0.14	0.37±0.06
	Forced-Marching	SL	0.82±0.11*	0.76±0.08	0.88±0.12
		ST	0.79±0.10	0.74±0.09	0.86±0.07†
		SS	0.42±0.08*	0.40±0.08	0.44±0.07
+55%BW	Running	SL	0.76±0.11	0.73±0.09	0.80±0.13†
		ST	0.84±0.10	0.80±0.09	0.88±0.10†
		SS	0.29±0.04	0.30±0.03	0.29±0.04
	Forced-Marching	SL	0.83±0.14*	0.74±0.11	0.92±0.09†
		ST	0.81±0.14	0.71±0.10	0.90±0.11†
		SS	0.38±0.06*	0.39±0.04	0.38±0.08

mean ± standard deviation
SL=Stride Length; ST=Stride Time; SS=Stride Speed
BW=Body weight (No additional load); +45%BW=Plus an additional 45% of BW; +55%BW=Plus an additional 55% of BW;

White Noise = 0.5 ('Suboptimal self-organization' ['S'] represented as <0.75);
Pink Noise = 1 ('Optimal self-organization' ['O'] represented as 0.75 – 1.30);
Brown Noise = 1.5 ('Impaired self-organization' ['I'] represent as > 1.30);
**Significantly greater than running*
†Significantly greater than men
‡Significant simple main effect of load for SL and SS (complexity increasing with load)

For stride length (SL) complexity, there was no significant interaction between load and locomotion ($F_{2,38}$=0.50, p=.61, η^2_p=.03). There also was no significant main effect of load ($F_{2,38}$=3.12, p=.06, η^2_p=.14). However, there was a significant main effect of locomotion ($F_{1,19}$=18.93, p<.001, η^2_p=.50), with forced-marching (0.82±0.12) having greater complexity than running (0.73±0.10).

For stride speed (SS) complexity, there was no significant interaction between load and locomotion ($F_{2,46}$=1.33, p=.28, η^2_p=.07). There was a significant main effect of load ($F_{2,38}$=7.13, p=.002, η^2_p=.27), with +45%BW (0.40±0.10) being greater (p=.004) than +55%BW (0.34±0.05). There was also a significant main effect of locomotion ($F_{1,19}$=17.63, p<.001, η^2_p=.48), with forced-marching (0.41±0.08) being greater than running (0.34±0.08).

4.4.2 Sex Comparison DFA

Refer to Table 10 for all mean and standard deviations of spatiotemporal related alpha coefficients (α) stratified by sex (final two columns). When conducting a secondary analysis to examine the effects of sex on spatiotemporal complexity (**Specific Aim 3.2**), for stride time (ST) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,44}$=3.03, p=.06, η^2_p=.12). There was no significant two-way interaction between locomotion and sex ($F_{1,22}$=3.31, p=.08, η^2_p=.13). However, there was a two-way interaction between load and sex ($F_{2,44}$=5.64, p=.007, η^2_p=.20). There was no significant simple main effect of load for males

($F_{2,21}$=1.39, p=.27, η^2_p=.12) or females ($F_{2,21}$=3.18, p=.06, η^2_p=.23). There was a significant simple main effect for sex at +45%BW ($F_{1,22}$=4.52, p=.05, η^2_p=.17) and +55%BW ($F_{1,22}$=15.20, p<.001, η^2_p=.41); with women (+45%BW=0.85±.08, +55%BW=0.89±0.11) having greater complexity than men (+45%BW=0.79±.11, +55%BW=0.76±0.10). There was no significant difference between sexes at the BW condition ($F_{1,22}$=0.32, p=.56, η^2_p=.01).

For stride length (SL) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=1.51, p=.24, η^2_p=.08). Similar to the stride time results, there was no significant two-way interaction between locomotion and sex ($F_{1,18}$=3.47, p=.08, η^2_p=.16). There was a two-way interaction between load and sex ($F_{2,44}$=3.43, p=.04, η^2_p=.16). There was a significant simple main effect of load for females ($F_{2,17}$=5.22, p=.02, η^2_p=.38); with BW (0.75±0.11) having significantly (p=.04, p=.02) less complexity than +45%BW (0.82±0.12) and +55%BW (0.86±0.11). There was no significant simple main effect of load for men ($F_{2,17}$=0.43, p=.87, η^2_p=.02). There was only a significant simple main effect of sex at +55%BW ($F_{1,18}$=8.48, p=.009, η^2_p=.32) with men (0.74±0.10) having less complexity than women (0.86±0.11). For BW and +45%BW there was no significant simple main effect of sex ($F_{1,18}$=0.04, p=.85, η^2_p=.002, $F_{1,18}$=3.53, p=.08, η^2_p=.16).

For stride speed (SS) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=0.21, p=.81, η^2_p=.01). There was no significant two-way interaction between locomotion and sex ($F_{1,18}$=0.20, p=.66, η^2_p=.01) or load and sex ($F_{2,36}$=0.36, p=.70, η^2_p=.02). There was no main effect of sex ($F_{1,18}$=0.002, p=.97, η^2_p=.00).

4.4.3 Group Completion Long-Range Correlation

When conducting a tertiary analysis stratifying the sample by individuals who completed the full ten-minute trial (completers) versus those who did not (non-completers) [**Specific Aim 3.3**], for stride time (ST) there was no significant three-way interaction between load, locomotion, and group ($F_{2,44}$=0.65, p=.53, η^2_p=.03). There was no significant two-way interaction between locomotion and group ($F_{1,22}$=0.002, p=.97, η^2_p=.00) or load and group ($F_{2,44}$=1.15, p=.33, η^2_p=.05). Lastly, there was no main effect of group either ($F_{1,22}$=0.00, p=.99, η^2_p=.00).

For stride length (SL) there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}$=2.33, p=.11, η^2_p=.12). There was no significant two-way interaction between locomotion and group ($F_{1,18}$=0.33, p=.57, η^2_p=.02) or load and group ($F_{2,36}$=0.73, p=.49, η^2_p=.04). Lastly, there was no main effect of group either ($F_{1,18}$=0.54, p=.47, η^2_p=.03).

Lastly, for stride speed (SS) there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}$=0.73, p=.49, η^2_p=.02). There was no significant two-way interaction between locomotion and group ($F_{1,18}$=0.28, p=.61, η^2_p=.02) or load and group ($F_{2,36}$=2.64, p=.09, η^2_p=.13). Lastly, there was no main effect of group either ($F_{1,18}$=0.89, p=.36, η^2_p=.05).

4.4.4 Spatiotemporal Higuchi Fractal Dimension

Refer to Table 11 for all mean and standard deviations of spatiotemporal related Higuchi method fractal dimension (**Specific Aim 3.1**). For stride time (ST) fractal dimension there was a significant interaction between load and locomotion ($F_{2,46}$=3.95, p=.03, η^2_p=.15). There was a significant simple main effect of load for forced-marching only ($F_{2,22}$=5.92, p=.01, η^2_p=.35); with BW (1.82±0.07) having greater dimension than +55%BW (1.77±0.10). Additionally, there was a significant simple main effect of locomotion at +45%BW ($F_{1,23}$=24.11, p<.001, η^2_p=.51) and +55%BW ($F_{1,23}$=14.67, p<.001, η^2_p=.39); with running having greater fractal dimension than forced-marching for both load conditions.

Table 11 Spatiotemporal Parameter Fractal Dimension

Load	**Locomotion**	**Variable**	**Total†**	**Men (n=11) ‡**	**Women (n=9)**
BW (No additional load)	Running	SL	1.84±0.11	1.87±0.12	1.80±0.09
		ST	1.85±0.07	1.86±0.07	1.84±0.07
		SS	1.83±0.11	1.85±0.13	1.80±0.08
	Forced-Marching	SL	1.81±0.13	1.85±0.15	1.77±0.11
		ST	1.82±0.07	1.85±0.07	1.78±0.06
		SS	1.82±0.10	1.84±0.12	1.79±0.08
+45%BW	Running	SL	1.85±0.12	1.88±0.11	1.81±0.12
		ST	1.86±0.10	1.87±0.13	1.85±0.06
		SS	1.84±0.11*	1.86±0.11	1.82±0.09
	Forced-Marching	SL	1.74±0.07	1.74±0.07	1.75±0.08
		ST	1.76±0.09	1.78±0.08	1.74±0.11
		SS	1.75±0.09	1.75±0.05	1.75±0.12
+55%BW	Running	SL	1.85±0.06	1.86±0.07	1.84±0.04
		ST	1.86±0.09	1.87±0.12	1.85±0.05
		SS	1.86±0.06*	1.86±0.07	1.86±0.05
	Forced-Marching	SL	1.81±0.11	1.84±0.11	1.78±0.10
		ST	1.77±0.10	1.79±0.09	1.75±0.11
		SS	1.82±0.09	1.84±0.08	1.79±0.11

mean ± standard deviation
SL=Stride Length; ST=Stride Time; SS=Stride Speed
BW=Body weight (No additional load); +45%BW=Plus an additional 45% of BW; +55%BW=Plus an additional 55% of BW;
FD=Fractal Dimension; FD=1.5 equivalent to 'brown noise'; FD=1.8 equivalent to 'pink noise'; FD≥2.0 equivalent to 'white noise' (stochasticity)
**Running significantly greater than forced-marching*
†Main effect of locomotion for SL and ST
‡Men significantly greater than women for SL and SS

For stride length (SL) there was no significant two-way interaction between load and locomotion ($F_{2,38}$=2.12, p=.12, η^2_p=.11). There was no significant main effect of load ($F_{2,38}$=1.28, p=.29, η^2_p=.06). However, there was a significant main effect of locomotion ($F_{1,19}$=14.38, p=.001, η^2_p=.43), with running (1.86±0.10) having greater fractal dimension than forced-marching (1.78±0.10).

For stride speed (SS) there was a significant interaction between load and locomotion ($F_{2,38}$=3.86, p=.03, η^2_p=.17). There were no significant simple main effects of load for running

($F_{2,18}$=1.30, p=.30, η^2_p=.13) or forced-marching ($F_{2,18}$=2.74, p=.09, η^2_p=.23). There were significant simple main effects of locomotion at +45%BW ($F_{1,19}$=14.75, p=.001, η^2_p=.44) and +55%BW ($F_{1,19}$=4.91, p=.04, η^2_p=.21); with running having greater dimension than forced-marching for both load conditions.

4.4.5 Sex Comparison Higuchi Fractal Dimension

Refer to Table 11 for all mean and standard deviations of spatiotemporal related Higuchi method fractal dimension stratified by sex (last two columns) [**Specific Aim 3.2**]. For stride time (ST) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,44}$=0.94, p=.40, η^2_p=.04). There was no significant two-way interaction for load and sex ($F_{2,44}$=0.86, p=.43, η^2_p=.04) or locomotion and sex ($F_{1,22}$=0.16, p=.70, η^2_p=.01). Lastly, there was no significant main effect of sex ($F_{1,22}$=0.25, p=.62, η^2_p=.01).

For stride length (SL) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=1.07, p=.35, η^2_p=.06). There was no significant two-way interaction for load and sex ($F_{2,36}$=0.38, p=.69, η^2_p=.06) or locomotion and sex ($F_{1,18}$=0.05, p=.83, η^2_p=.05). Lastly, there was a significant main effect of sex ($F_{1,18}$=4.28, p=.05, η^2_p=.19), with men (1.84±0.11) exhibiting greater fractal dimension than women (1.79±0.09).

For stride speed (SS) there was no significant three-way interaction between load, locomotion, and sex ($F_{2,36}$=1.07, p=.35, η^2_p=.06). There was no significant two-way interaction for load and sex ($F_{2,36}$=0.14, p=.87, η^2_p=.01) or locomotion and sex ($F_{1,18}$=0.00, p=.99, η^2_p=.00). Lastly, there was no significant main effect of sex ($F_{1,18}$=1.99, p=.18, η^2_p=.10).

4.4.6 Completion Group Comparison Higuchi Fractal Dimension

The tertiary analysis assessed the difference between completion groups (completers versus non-completers) [**Specific Aim 3.3**], for stride time (ST) there was no significant three-way interaction between load, locomotion, and group ($F_{2,44}$=0.27, p=.77, η^2_p=.01). There was no significant two-way interaction between load and group ($F_{2,44}$=0.49, p=.61, η^2_p=.02). However, there was a significant two-way interaction between locomotion and group ($F_{1,22}$=7.41, p=.01, η^2_p=.25). There was a significant simple main effect of locomotion for completers ($F_{1,22}$=11.93, p=.002, η^2_p=.35) and non-completers ($F_{1,22}$=26.38, p<.001, η^2_p=.55); with running having greater fractal dimension than forced-marching for both groups. There was a significant simple main effect of group for forced-marching only ($F_{1,22}$=7.18, p=.01, η^2_p=.25), with completers (1.81±0.08) having greater fractal dimension than non-completers (1.72±0.09).

For stride length (SL) there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}$=1.49, p=.24, η^2_p=.08). There was no significant two-way interaction between load and group ($F_{2,36}$=1.86, p=.17, η^2_p=.09) or locomotion and group ($F_{1,18}$=1.14, p=.30, η^2_p=.06). There was no significant main effect of group ($F_{1,18}$=1.86, p=.19, η^2_p=.09).

For stride speed (SS) there was no significant three-way interaction between load, locomotion, and group ($F_{2,36}$=0.07, p=.93, η^2_p=.00). There was no significant two-way interaction between locomotion and group ($F_{1,18}$=1.17, p=.29, η^2_p=.06). There was a significant two-way interaction between load and group ($F_{2,36}$=4.45, p=.02, η^2_p=.20). There was a simple main effect of load for non-completers only ($F_{2,17}$=4.83, p=.02, η^2_p=.36), with +45%BW (1.71±0.10) being less (p=.05, p=.02) than BW (1.84±0.12) and +55%BW (1.85±0.09). There was a simple main effect of completion group at +45%BW only ($F_{1,18}$=10.44, p=.01, η^2_p=.37), with completers (1.82±0.08) being greater than non-completers (1.71±0.10).

5.0 DISCUSSION

Motor control of bipedal ambulation is a complex multi-dimensional process and delimiting the examination to a couple factors will only provide a narrow interpretation. While computational paradigms delimit the dimensionality of the observed task (to simplify mathematical calculations), they account for only one particular aspect of motor control. Thus, multiple computational paradigms (goal equivalent manifold, detrended fluctuation analysis and Higuchi's fractal dimension) can be used simultaneously (albeit cautiously) to potentially illuminate a more comprehensive representation of the individual's motor control (or state of the locomotor system). However, considering the number of factors assessed (and presented here within) each outcome should first be examined independently. Moreover, given the dynamical nature of the present experimental protocol, descriptive information and their (or lack of) statistically significant differences should be discussed due to their potential ramifications on the interpretations of locomotor system function. Therefore, the following results will first present the descriptive findings (i.e., subject characteristics and RPE) to contextualize the experimental conditions. Then each outcome will be discussed independently starting with interpretations of results for the entire sample followed by the influences of between group factors (sex and completion ability as they were not adequately powered). All potential and competing explanations will be presented to develop the foundational knowledge necessary to be considered for the global interpretation of all outcomes together (i.e., Chapter 6).

5.1 Subject Characteristics

As discussed in section 2.3.1, actions and their subsequent affordances are derived from three distinct components: i) system constraints, ii) task constraints and iii) environmental constraints[51, 53, 114, 176-180]. System constraints represent the characteristics specific to the individual such as their strength/endurance capabilities, range of motion and even anthropometric characteristics (i.e., height, weight, and body composition). Although, analysis of subject characteristics does not represent a specific aim of the present investigation, they need to be described and discussed, nevertheless. To delineate effects/causes of locomotor system *function* between individuals, it must first be discerned if significant differences in system constraints exist (especially for between group factors of sex and completion ability).

Men were observed to be significantly greater in stature (height [m] and weight [kg]) compared to the women in this sample, congruent with previous sex related investigations of load carriage[8]. Additionally, in support that all participants were healthy, recreationally fit individuals, both sexes had healthy range BMI (20-25) with no significant differences between the two, indicating participants had healthy body composition. Although relative load magnitudes were used in this investigation, in contrast to previous research that utilized absolute loads[8]; men had significantly greater absolute loads compared to the women in this sample (due to their significantly greater body weight).

Despite men being taller on average than women, their trial velocities were comparable to the women even during loaded conditions (refer to Table 1). Theoretically, longer lower extremities should lead to a higher gait transition velocity resulting in higher trial velocities[286-288] (which were set to 10% above their gait transition velocity in the present investigation). However, given men had significantly greater absolute loads/weight, and each gait transition velocity was determined for each load condition separately, it is possible the additional mass counteracted the effects of longer limbs[289]. Kinetic outcomes such as peak force and loading rate have been shown to influence gait transition velocity over mechanical variables like limb length[290]. Likewise, the gait transition velocity may be initiated by the increased perceived effort of the tibialis anterior, rectus femoris and hamstrings[291]. Greater activation of the aforementioned muscles yields an afferent interpretation of greater force requirement[291]. Thus, due to larger masses (natural mass and added masses) the gait transition velocities of the men in the present study were lower to potentially meet a reduced kinetic cost function most likely triggered by increased lower extremity muscle activation (from increased force requirement to translate with additional load carriage).

While differences were observed between sexes, there were a lack of statistically significant differences between completion groups (completers vs. non-completers), except for trial velocity during the +45%BW condition. However, it is very likely this finding committed a type I error as the groups were very uneven in size. Moreover, two of the female participants did not even attempt the +45%BW load conditions (one stated they could no longer execute gait tasks with load, the other was exempt at the discretion of the study team). Therefore, for the +45%BW trial velocity comparison the degrees of freedom were further reduced resulting in a biased comparison. Levene's test for equality of variances was not violated, however the group comparison was mismatched in size (18 completers, 6 non-completers) which may have resulted in a potential type I error.

Importantly, the lack of significance indicates no difference between completion groups in terms of basic characteristics (age, weight, height, trial velocities and trial load magnitudes). Therefore, it can be surmised that basic characteristics regarding physical stature are not the primary contributing factor to inability to execute prolonged motor tasks. Moreover, when basic characteristics are equal, but motor behavior is divergent in ability (in terms of ability to complete the predetermined duration as observed in the present investigation) another measure is required to characterize the dichotomy of the groups (completers vs non-completers).

In athletic[292-294] and military[295-298] settings, basic characteristics such as age (non-modifiable) and anthropometrics (modifiable), such as body composition, are the factors most heavily relied on to identify MSI risk. Although it should be noted that athletic populations have adopted a more progressive approach to injury risk identification relying on (a more robust set of) performance metrics compared to military populations[299-301]. While factors of age and body composition have demonstrated some prognostic utility, only extreme outliers appear to be identified for greater risk (i.e., obese or ≥40 years old). Furthermore, slips, trips and falls still largely contribute to observed MSI[302] implicating deficits/changes in motor control (which is not addressed with assessment of age and body composition). This further warrants the need for further examination of the locomotor system function to determine a valid (and more sensitive) measure of prognostication for successful task execution and ultimately pre-emptively identify MSI risk in military populations.

Lastly, the limitation of the current sample should be addressed. In the present investigation, the observed sample has similar physical characteristics with recruit-aged populations, especially in terms of men being generally larger than women[8]. However, while women represent a smaller percentage of the military (women account for about 17% of all active duty personnel[303]) the present investigation oversampled to achieve equal group sizes for sex. One limitation of the current sample, however, is the lack of low end physically capable individuals (in terms of strength and conditioning). In order to ensure the greatest probability of trial completion, inclusion criteria were narrowed including the need to be able to run on a treadmill for ten minutes at six miles per hour. Despite, this limitation, the absence of significant differences between completion groups in terms of system constraints is important factor to consider as it implicates that differences in locomotor system function are attributed to alternative factors (to be discussed in the coming chapters).

5.2 RPE

Similar to subject characteristics, the analysis of RPE was not a primary aim of the present investigation. However, ten-minutes of prolonged ambulation (especially with load carriage) is a dynamical task, meaning that as time evolves the state of the locomotor system changes. Fatigue is one aspect that changes over the course of the experimental trial that warrants attention to understand how specific variables of the experimental protocols alters fatigue. The information presented in the following section (5.2) is to clarify the fatigue response (as some were statistically significant but not necessarily clinically meaningful) to each independent variable to provide context for interpretations of the outcomes regarding the specific aims of the present investigation.

Load played an important role on RPE in the individual, with the heaviest load condition of +55%BW (16±3 RPE) yielding significantly ($p<.001$) greater overall RPE values than BW (no additional load condition) [10±2]. Despite relative locomotion velocities only 10% above gait transition velocity for each respective load condition, the addition of 45% and 55% of BW yielded RPE values that correspond with maximal effort activities (RPE = 18-20), whilst the unloaded condition corresponded with minimal effort (refer to Figure 2). Additionally, a significant main effect of locomotion was observed but only accounted for 22% of the observed variation. The small effect size, coupled with how RPE is scored (whole integers 6-20) indicates there wasn't a clinically meaningful difference between locomotion patterns. Although RPE during running was statistically significantly greater, the mean overall RPE was 13.67 compared to 13.1 for forced-marching (for the purposes of clarity, RPE values from the analysis were rounded to the nearest whole number). Therefore, there was no clinically meaningful difference between locomotion patterns in terms of RPE.

Likewise, change in RPE (the difference of RPE from minute 0 and minute 10) demonstrated a significant effect for load (p<.001) and not locomotion (p=.09) [refer to Figure 3]. On average BW resulted in only a two-step change in RPE, indicating little change in perceived exertion rate; while both loaded conditions (+45%BW and +55%BW) both yielded a mean 6-step change in RPE (even considering some participants did not complete a full ten minutes of loaded ambulation). Thus, as the steady-state (constant velocity) gait task progressed, an individual's perception of their exertion increased greatly with the addition of greater load carriage magnitudes, consistent with previous research[85, 86, 152]. Therefore, it is reasonable to conclude that load is the primary contributing factor in differences of overall/change RPE and locomotion pattern has limited/varying impact (which may be more subject specific). Furthermore, locomotion at relative velocities of 10% above gait transition velocity require a greater metabolic cost when load carriage is present.

Sex played no role in ratings of perceived exertion evidenced by the lack of significant differences between men and women (for overall and change in RPE). In relative terms between sexes men and women experienced the same perceived effort. Despite some research indicating that women's physiology is better suited for local muscular endurance compared to men [304-308] (which may be taxed greater with an increase in mechanical demands of the lower extremities[20, 84, 98, 102, 104]), in the present study women *perceived* effort the same as men (women could have been more metabolically efficient but this would have to be confirmed with respiratory gas analysis which was not performed in the present investigation). Thus, this lack of difference between sexes can most likely be attributed to the loads being determined relative to body mass. Given that women were significantly smaller in stature in this sample than the men, had load magnitudes been absolute it is very likely that women would have reported higher RPEs than men.

Additionally, sex was not the only between group effect analyzed as ability to complete the trial was examined as well. Not surprisingly, individuals who were unable to complete the full ten-minute trial (non-completers) reported ~2 steps higher on the 6-20 RPE scale than those who did during the loaded conditions. Indeed, every participant completed the entire ten-minute trial unloaded for both locomotion patterns. A variety of reasons were given for inability to complete the full ten-minute trial. Some individuals reported an inability to maintain velocity with the load. Other individuals reported being out of breath and felt unable to continue much longer. It can be conservatively concluded that both mechanical and metabolic deficiencies played a role in the inability to execute the gait task successfully because it was too difficult relative to *their specific* system and task constraints. Therefore, RPE accomplished its required goal of accurately capturing an individuals perceived exertion (tasks they failed were harder and they reported higher RPEs). In all cases where participants failed to complete the ten-minute trial, it should be known that participants were weary of falling so they requested to stop premature of actual task failure.

5.3 GEM

Gait is a cyclical (executing the same task repeatedly [i.e., stepping]) and variable motor task[309] that is seemingly mundane due to the commonplace of its everyday use[310]. However, gait requires the complex integration of a many elements that can function independently and in concert to yield the emergent property of stepping[311-313]. Moreover, the locomotor system not only has to control/organize elements within the system to yield the appropriate step (i.e., correct foot orientation, placement, force, etc.) it must account for environmental constraints (i.e., terrain, weather, gravity, etc) that interact with the task and system[114, 177, 218, 219, 243]. External task constraints can be imposed on the locomotor system further perturbing it. In military populations a common task constraint imposed on the warfighter that perturbs their locomotor system is load carriage[314]. Additionally, warfighters are often required to utilize a walking gait pattern at velocities beyond their gait transition velocity (forced-marching) while carrying load.

Mean biomechanical outputs routinely reported, have uncovered important characteristics of average gait behavior but have failed to encapsulate driving factors of the locomotor system that controls gait[74]. The variability of movement is believed to provide greater insight to the limits/thresholds and regulation strategies of the locomotor system capacity[59]. The GEM framework provides an opportunity to correlate geometric (structure of variance) and dynamical (temporality) methods of variability[59]. Utilizing load carriage and imposed locomotion patterns (forced-marching) as task constraint perturbations, the GEM framework can elucidate variability, control and regulation strategies of gait that military recruits (novice to load carriage) may use when first introduced to load carriage tasks. Prior to the examination of the GEM derived outcomes (relative variability, subspace variability and subspace regulation which represent **Specific Aims 1** and **2**) spatiotemporal parameters will be discussed to provide further context.

5.3.1 Spatiotemporal Parameters

Utilizing a forced-marching locomotion pattern (walking at a velocity above gait transition velocity) exhibited ~22% longer strides on average compared to running regardless of load magnitude. Longer strides being performed during forced-marching compared to running is likely a compensation to achieve the trial velocity (task goal) as forced-marching eliminates a flight phase[183, 315, 316]. However, as load magnitude increased mean stride length became ~7% shorter for forced-marching. The latter finding supports previous research that has demonstrated that load carriage (especially load magnitudes of 30% of body weight and greater) decrease stride length[20, 89, 105, 107, 109, 159-161]. Furthermore, the present investigation extends this finding to greater load magnitudes (+55%BW) and to women (whom have largely been underrepresented in the load carriage literature[8, 93, 109, 161, 317]). Likewise, mean stride time was ~23% longer for forced-marching compared to running (at the same velocity) and remained unchanged with load magnitude regardless of locomotion pattern. Similar to mean stride length, stride speed decreased with load for forced-marching only.

The reason for the observed mean spatiotemporal differences is likely mechanical in nature. During bipedal ambulatory tasks, force is transmitted up through the kinetic chain (sometimes referred to as impact force or shock) [318-324]. However, not all force is transmitted through the entire structure of an individual as some of the force is attenuated through various structures of the human body (i.e., ligaments, cartilage, muscle and even bone)[318-324]. Considering Newton's third law, when now introducing load carriage to the system (i.e., individual), the additional mass will contribute to a greater force applied into the ground. Not surprisingly, the vGRFs observed in load carriage perturbed gait tasks, increase significantly[102, 103, 163]. Therefore, a greater impact force will be transmitted up the kinetic chain, potentially increasing musculoskeletal injury risk[325]. Given this notion, it is possible the locomotor system recognizes this as it would be indicated from increased mechanoreceptor feedback (particularly from the golgi tendon organs and muscle spindles)[326]. As a countermeasure, the locomotor system possibly modulates stride length as load increases (organizing around a kinetic cost function) to be shorter to ensure that the lower extremities are more optimally aligned to absorb shock (impact force). Thus, the appropriate structures (such as the eccentric action of muscle) are capable of ameliorating force rather than compromising passive elements (i.e, ligaments, cartilage, or bone)[324].

Modulation of stride length because of load magnitude may not only be performed for a kinetic cost benefit but a stability one as well (specifically in terms of postural stability). Indeed, much of motor control research has reduced the dimensionality of motor tasks in an effort to improve computational simplicity. However, it is widely regarded that even simple motor tasks (such as stepping) are highly dimensional[327, 328], hence it is more likely that motor output choices are made by a combination of cost functions (weighted differently depending on a variety of other factors). Specifically for load carriage, postural stability reduces with increases in load magnitude especially in the medial-lateral axis[212, 213]. Thus, a shorter stride may be performed as load increases to reduce the anterior-posterior length of the base of support, which consequently would extend the medial-lateral base of support improving stability along that axis (which is already compromised by load). Likewise, shorter steps would further reduce COM excursions so that the center of gravity trajectories would be easier to maintain within/near the base of support. Lastly, the postural stability cost function would explain why this change in mean stride length was observed only in forced-marching as that locomotion pattern exhibits much longer strides compared to running to achieve the necessary trial velocity (task goal).

Men in general had greater mean stride lengths, and stride times. This finding is expected given that the men in this sample were significantly taller than women (Table 1), providing further support for the notion that, longer limbs will result in longer strides[329]. Furthermore, increased stride times can be explained by the lack of significant differences in trial velocity, even during loaded conditions, between men and women (1.65±0.21 and 1.63±0.22 m/s respectively) Tables 1). Therefore, men take longer strides but perform them more slowly (hence longer stride time) to achieve a velocity relative to what the women executed (who performed shorter quicker strides to achieve the same velocity). For theoretical reasoning as to why men and women executed the gait transition around similar velocities despite differences in anthropometry refer back to chapter 5.1 paragraph 2.

Importantly, there were no differences in mean spatiotemporal parameters between individuals who could complete the full ten-minutes and those who could not, suggesting that on *average* both groups executed strides similarly. Moreover, this further indicates that there is not an optimal mean spatiotemporal parameter value that correlates with prolonged performance of loaded gait tasks. The latter finding further demonstrates the limitations of mean parameter assessment in terms of task execution outcome. When considering the equifinality of motor behavior, an optimally functioning locomotor system will utilize a family of motor solutions to execute a specific task that yield similar performance outcomes but provide degeneracy [327, 328]. The prominent benefit of degeneracy is it affords the locomotor system the ability to maintain performance in the presence of perturbation/constraint (stability) or utilize a unique solution to overcome a perturbation (flexibility)[251, 327, 328, 330-332]. Ultimately, the mean spatiotemporal parameter represents the average of observed values but fails to i) describe range of behavior and ii) describe how those range of behaviors evolve over time. Therefore, the mean behavior is not necessarily a determinant of task execution outcome (especially when executed for a prolonged duration) and that the temporal correlation and variability must also be considered when trying to determine completion group differences in dynamics of motor control.

Therefore, to comprehensively examine motor behavior of gait, spatiotemporal parameter variance was also assessed to determine if load magnitude and locomotion pattern effected the *amount* of variability observed. Stride length variance remained unaffected by load and locomotion denoting the *range* of observed values remained the same across conditions. This particular finding is supported by previous research[74, 77] and is to be expected when an individual utilizes a goal manifold relevant strategy. Conceptualizing that the locomotor system constructs a multidimensional unitless space (termed state-space), the task goal represents a trajectory or specific location in that space (i.e., the goal manifold). Task space encapsulates the solution space specific to task outcome (task relevant) whereas the null space represents the solution space that does not impact task outcome (task irrelevant)[333]. When leveraging a goal manifold relevant strategy, the locomotor system modulates variability in the task space domains (i.e., around the goal manifold) and not global variability (i.e., increasing variability of all parameters universally)[74, 77, 334-337]. In the present investigation the parameter coupling (stride length*stride time combination [stride speed]) constructs the task space (task relevant) and null space (task irrelevant) elements of state-space[59, 74, 76, 77] (which is why stride length variance is not expected to change but rather stride speed variance). Thus, load and locomotion pattern only affected the mean behavior of stride length, and not it's variability over the course of a prolonged gait task.

In contrast with previous findings[74, 76, 77] where no changes were observed in stride length or stride time variance across varying conditions (only changes in stride speed variance (coupling parameter) were observed), increases in load magnitude increased stride time variance. Moreover, stride speed variance was significantly greater for forced-marching compared to running. It should be noted, in the aforementioned investigations[74, 76, 77] comparisons were made at different velocities with no additional perturbation such as load magnitude or imposed locomotion pattern. Furthermore, the second investigation by Dingwell et al (2017)[77] performed unperturbed walking tasks but simply compared young and elderly healthy adults. Therefore, stride speed variance wasn't necessarily the only *order* parameter that was controlled/regulated when executing the various gait tasks as perturbations of load magnitude and imposed locomotion pattern were introduced into the system.

Additionally, the cost function dimensionality likely increased in the present investigation compared to previous investigations[74, 76, 77]. As stated previously, participants in the previous investigations ambulated at preferred speeds with no additional load carriage, within which the locomotor system is presumed only to account for a metabolic cost function (to drive optimal self-organization). Indeed, for walking specifically, individuals organize locomotor function around a metabolic cost function choosing velocities that reduce metabolic cost for given distances[338-340] by utilizing a step frequency that minimizes energy use[339, 341-345]. Furthermore, individual's strong preferences towards specific step frequencies suggest spatiotemporal parameters are an important component to the locomotor system's adaptation[343, 344]. It has been demonstrated that individual's preferred step frequencies correlate with a reduction of 4-8% energy cost[346]. How the locomotor system identifies the optimal movement strategy that adheres to the metabolic cost function remains unclear[346] but its suggested that this process occurs both over the lifetime of an individual[347] as their locomotor system adapts body shape, muscle action, and neural circuity networks[348-350], and continuously, evolving and adapting in response to more immediate perturbations[350-354]. It is important to note, that while experimental data may demonstrate adaptation to reduce metabolic cost, additional cost functions may serve as the determinant of locomotor system organization such as stability, accuracy, or force minimization, with their weighting varying depending on a multitude of factors [355-358]. The present investigation diverges from previous work[74, 76, 77] by increasing the perturbations/constraints imposed on the locomotor system. Not only does the addition of load alter the energy cost per step (metabolic cost function), but it can also impose potentially deleterious mechanical forces (kinetic cost function)[8, 32, 93, 97, 325, 359-361] and even disrupt balance (postural stability cost function)[99, 107, 108, 211-213, 362, 363]. Compounding the computational complexity, the individual must also maintain a specific velocity (i.e., trial velocity, which may differ from *their* preferred locomotion velocity) to avoid drifting off the back of the treadmill. Therefore, the locomotor system may explore changes in additional spatiotemporal variance (beyond the coupling parameter [stride speed]) in an attempt to address multiple cost functions simultaneously.

The novelty of the task should be acknowledged but clearly defined to avoid confusion concerning the term ‘novel’. Participants in the present investigation were all *novices* to load carriage (i.e., had never served in the military or extensively executed load carriage related activities such as backpacking). However, the experimental tasks are not necessarily ‘novel’, in that they were not completely unfamiliar movement patterns. Indeed all subjects were recruit-aged (18-35 years), healthy (free of musculoskeletal injury/disorders), relatively fit (as determined by the inclusion criteria of engaging in moderate physical activity at least three times a week and being able to run on a treadmill for 10 minutes at 6 mph), and fully ambulatory indicating that they've been walking and running for at least ~17 years (considering infants begin walking on average around 12 months old[364, 365]).

The control and experimental gait tasks in the present investigation may not have been 'novel' motor behaviors for the participants but the experience with how to adapt to the perturbations/constraints were. As such, participants may be unsure how to select/weight the various cost functions (i.e., metabolic, kinetic, postural stability) simultaneously to optimally organize the locomotor system to utilize the most appropriate family of motor solutions. Further, how gait optimization (while adhering to various cost functions) is achieved tends to be somewhat subject specific as some individuals exploit natural gait variability to identify new optima, while others need directed/enforced gait changes to identify new optima[366]. It is unclear whether the observed changes in variance were effective or ineffective state-space exploration of the various local landscapes (each cost function inhabiting a different landscape that represents an expense gradient)[366]. Regardless, the specific perturbation of load magnitude and forced-marching likely increased task dimensionality effecting variance of multiple spatiotemporal parameters incongruent with previous work[74, 76, 77].

Sex comparisons of spatiotemporal parameter variance yielded no statistically significant differences, indicating sex is not a moderator of spatiotemporal parameter variance despite differences in mean behavior. An absence of difference between sexes demonstrates the paradoxical nature of motor control and why the analysis must be comprehensive and nuanced. Men and women executed strides differently from one another in terms of mean behavior, which is believed to be attributed to the anthropometric differences between the sexes (i.e., height). Yet despite having different system constraints, men and women utilized similar *ranges* of motor behavior around their mean (or preferred operating point for the specific task). However, from a performance perspective (especially sport) much emphasis is placed on improving system constraints (i.e., force production, power, size, etc.) through means of strength and conditioning[118]. The present findings suggest alternatively that differences in system constraints and mean behavior may not be indicative of task performance and that alternative factors contribute to an individual's locomotor system being able to optimally self-organize (and thus converge on a true optimum).

While sex failed to influence spatiotemporal parameter variance, the ability to complete the ten-minute trial did (completers versus non-completers). Specifically, for stride length variance, non-completers exhibited more variance than the completers during forced-marching. It is unclear what contributes specifically to this finding but a few plausible explanations prevail that should be discussed: i) the individual (non-completer) refrains from exploratory behavior and tries to maintain a preferred spatiotemporal parameter behavior (which is suboptimal for the current task, ii) motor variability of the non-completer executed on a stochastic basis inhibited the rate of adaptation, and/or iii) load overrode their (non-completers) true natural gait transition and experimentally derived trial velocities were too high.

Firstly, when discussing exploratory behavior in regard to motor variability an important distinction between adaptation and learning must be made which primarily centers on the timescale of the activity[367]. Adaptation occurs quickly in a relatively short time scale (i.e., within a single 10-minute trial) whereas learning occurs over much larger time scales (i.e., days, months and years)[367]. While both incorporate variability which serves as a form of state-space exploration to identify optimal motor solutions[368], adaptation is attempting to reduce afferent (sensory information) prediction errors[369] to improve the immediate performance outcome of the task goal[367]. Likewise, behaviors acquired through adaptation are typically lost following the removal of a perturbation[370]. Therefore, a hallmark of de novo learning is the retention of the desired skill[367]. The design of the present investigation (a single trial performed for each condition) predicates that *adaptation* is the likely observed exploratory behavior and not learning (which would require multiple trials/sessions of the same condition to confirm).

When executing common tasks (i.e., walking), preferred behaviors (or attractor state) can become deeply entrenched with resistance to identifying new optima (attractor states) even with the introduction of perturbation[346, 366]. Selinger et al (2017) examined how the locomotor system responded to resistive torque modulating exoskeletons (i.e., the perturbation) during gait tasks and observed some individuals maintained their preferred step frequency despite the energetic penalties[346]. However, when r*equired* to perform a step frequency exploratory phase (utilizing varying sinusoidal metronome tempos that enforced larger variation in step frequency), individuals adopted a step frequency matching the energetic optima (based on the exoskeleton perturbations) rather than their preferred frequency[346]. Thus, in the presence of new/unfamiliar perturbations/task constraints some individuals maintain (or revert to) their preferred motor behavior even if it is suboptimal[346, 366]. In the present investigation the non-completers may have been individuals resistant to transition to new optima (staying in their preferred behavior) while executing loaded forced-marching. Consequently, their preferred behavioral parameters were unable to meet the demands of the task (i.e., maintain velocity) resulting in them having to run or ‘gallop’ towards the front of the treadmill at times to keep from drifting off the back. The interspersal execution of a varying motor pattern would then contribute to the observed greater variance in stride length.

Alternatively, the observed increase in stride length variability of non-completers may be due to a stochastic state-space exploratory behavior (i.e., driven by random processes over time)[371]. By increasing motor variability there may be an increase in solution exploration, however it can adversely impact learned solution retention[333]. Motor variability represents adaptation in the locomotor system, to identify new task solutions and break free from suboptimal solutions[114, 372, 373]. But executing a task with greater variability may inhibit the ability to effectively consolidate a family of solutions for a specific skill (i.e., forced-marching with load carriage), resulting in more task performance errors (and more variability in the task space [task relevant] specifically)[334]. A potential explanation for this behavior is that calculated adaptation (successive trials are similar or correlated[374, 375]) is inhibited because variability disrupts the system from utilizing similar solutions trial to trial to identify an optimal task solution space[334]. Therefore, non-completers are randomly executing variations that do not enhance the search for the optimal attractor state (family of motor solutions)[334]. Likewise, the completers are fine-tuning their motor behavior as time evolves to adapt to the task resulting in the lesser stride length variance.

Lastly, non-completers may have executed more stride length variance compared to completers because of their gait transition velocity. A variety of factors can influence gait transition velocity such as limb length, peak force and loading rate[286-288, 290, 291]. In the current investigation, gait transition velocity was determined as the moment a participant transitioned from walking to running and calculated as the mean of three ramped-protocol trials (accelerating at 0.05 m/s^2). Furthermore, this protocol was repeated for each load condition separately with the trial velocity set at 10% above that specific load condition's gait transition velocity. Considering that all participants (n=26) completed the BW conditions, their gait transition velocity was most likely accurate with their *true* natural gait transition velocity for BW. However, the additional load may have deterred individuals from transitioning at a natural point because the locomotor system was delaying the transition based on a potential cost function (i.e., kinetic, such as loading rate [refer back to chapter 5.1]).

Additionally, non-completers potentially had discordance between their representation of system capabilities and their ability to (re)calibrate perceptions with appropriate actions[51, 53, 165, 168-172]). Multisensory information is continuously obtained, and recurrent processing updates an individual's representation of themselves and the environment (i.e., their perception). Based on those internal representations, the locomotor system determines a set of possible actions available to them (affordances)[51-53]. However, if perceptions are inaccurate then impractical/unattainable affordances may be available to the individual[51-53]. With the addition of load carriage, which was substantial in the present investigation (≥45% of BW), new sensory information is introduced into the locomotor system (even when just standing). Non-completers may have been unable to accurately calibrate their affordances based on the new sensory information, leading them to transition at velocities near or above their unloaded gait transition velocity. Hence, non-completers were executing loaded trials at a velocity way beyond their capabilities.

It should be noted findings (or lack thereof) should be interpreted with caution when considering between subject factors of sex and ability to complete the entire ten-minute trial (completers vs non-completers). In the present study, these were secondary and tertiary exploratory analysis that were not adequately powered as it would have required a sample n>100 (based on a priori determination). Moreover, the ability to complete group sizes were unequal with completers having 15 participants and the non-completers having 5 participants. However, analysis did not violate assumptions (Mauchly's test of sphericity, Box's M, Levene's test of equality of variance) and ANOVAs being robust against non-normality (in terms of type I error)[376] indicating the findings are still valid, but additional investigations are warranted to confirm findings.

5.3.2 Relative Variability

When examining relative variability (to address **Specific Aim 1.1**), the findings of the current investigation were in partial support of previous load carriage research by Krajewski et. al. (2020)[109], where relative variability was significantly impacted by load magnitude and locomotion pattern independently. In contrast to the aforementioned investigation[109], as the load magnitude increased, relative variability increased by ~20% from BW to +55%BW load conditions (also failing to support our hypothesis1.1a). Similar to previous findings (and supporting hypothesis1.1b), forced-marching, demonstrated ~25% more relative variability than running regardless of load condition[109]. Moreover, relative variability values observed during the natural locomotion/unweighted trial (running at BW) [refer to Table 7.] were consistent with those observed in healthy younger and elderly adult populations ambulating (running and walking) at preferred speeds[74, 76, 77].

A relative variability value greater than 1 is considered ideal as this outcome is calculated as the standard deviation of tangential ('good') variability divided by the standard deviation of perpendicular ('bad') variability[74, 76, 77]. Perpendicular ('bad') variability (i.e., the task space) encapsulates the solution space specific to task outcome (task relevant) whereas tangential ('good') variability (i.e., the null space) represents the solution space that does not impact task outcome (task irrelevant)[333]. In more laymen terms specific to the present investigation, a movement solution variation (parameter coupling of stride length and stride time) in the task space (perpendicular to goal manifold) would result in a velocity other than the trial velocity. Conversely, movement solution variations in the null space (tangential to goal manifold) will result in a velocity that matches the trial velocity.

Task skill and variability has been considered inversely related, however greater variability of task execution can result in reduced outcome variability (i.e., less deviations from the trial velocity in the present investigation)[377]. The caveat to the previous statement being that greater variability is observed in the null space specifically and variability reduction is limited to certain aspects (i.e., task space) [or relative variability >1] during redundant/cyclical tasks (i.e., walking)[334-337]. Thus, variability (in particular null space variability) serves a multitude of purposes including task exploration (adaptation/learning/skill acquisition) and flexibility/adaptability to perturbation (i.e., degeneracy)[377]. Most likely however, there is an optimal amount of variability (in general and in terms of the ratio between null space and task space variability [i.e., relative variability]) as too little or too much could be detrimental to performance[378].

Much larger relative variability values (like those observed in the present study, see Table 7) may be suboptimal. It was recently proposed by Krajewski et. al. (2020)[109] that larger relative variability values (>2.0) may indicate a motor variability geometric representation at the edge of an individual’s action boundary as the individual must rely on a larger number of stride parameter combinations (family of motor solutions) to successfully execute the task[223]. While all movement solutions in the null space may achieve the task goal (trial velocity) they are not equal in terms of various cost functions[59]. For example, some coordinative stride patterns will achieve a specific trial velocity but come with a penalty of higher energy cost. Likewise, a different variation may achieve the trial velocity but align the lower extremity segments in an orientation that puts them at risk of suffering a musculoskeletal injury (i.e., kinetic cost penalty), but even these explanations oversimplify the dynamics of null space[355-358].

Even within the null space landscape there are various attractor states (with different families of solutions that achieve the task goal) formed by the different cost gradients[334, 346, 366, 379, 380]. Each cost function (i.e., metabolic, kinetic, stability, etc) constructs a new dimension and the gradient of the landscape (imagine a heatmap overlayed a multidimensional surface plot) within that dimension represents the range of cost (i.e., the metabolic cost per stride of using one stride pattern versus another)[346]. Thus, utilization of too large an area of the null space may be suboptimal as individuals are: i) inducing large cost penalties, ii) ineffectively weighting the multitude of cost functions, iii) ineffectively executing local searches to identify a true optimal. Therefore, the findings of greater relative variability may be attributed to a multitude of factors that need to be discussed further such as: i) combating effects of fatigue (or more rightly, cost function competition), ii) state-space exploration, iii) noise and/or iv) use of alternative locomotion patterns within a single trial.

Indeed, significantly greater RPE values were observed during the loaded conditions (consistent with other investigations[84-86, 152]) which support the theory that higher relative variability values indicate an individual acting at the limit of their action boundary (in terms of bioenergetics). To delay the effects of fatigue and maintain successful task execution (ambulate at the prescribed treadmill velocity) individuals are utilizing a greater variety of movement solutions to reduce local muscular fatigue[227, 317]. Thus, mechanical work can be distributed across a larger number of muscle fibers, ameliorating the compounding fatigue effects of using a localized subset of fibers[227, 317]. Specifically, during loaded forced-marching (imposed locomotion), mechanical work is shifted proximally to the hip compared to loaded running (natural locomotion), reducing the contributions of the optimally arranged musculature around the knee[317]. Not surprisingly in the present investigation, forced-marching at +55%BW exhibited the largest relative variability values. Therefore, the large relative variability value may reflect the locomotor system organizing based on a metabolic cost function[74] by attempting to reduce local muscular fatigue, especially for the hip musculature[317] utilizing a wider array of motor solutions.

Further contributing to the notion that greater relative variability is an attempt at fatigue mitigation; the current sample were novice load carriers. Although, the sample was relatively fit, performance adaptations (physiological and mechanical) reflect the stimuli they experience. Load carriage imposes a constant burden on the individual by increasing mechanical and physiological work[8, 20, 26, 84-87, 92, 94, 96-98, 104, 151, 359, 381]. Anecdotally, many participants indicated breathing required more effort during loaded conditions, likely due to the muscles responsible for respiration never having to work while being physically restrained (the load itself is strapped securely to the individual's torso). Thus, this sample had yet to adapt *their* system constraints (i.e., local muscular endurance, force production, fatigue resistance, etc) to this specific task constraint (i.e., addition of load) [reflected in the large observed relative variability values].

Due to the samples' unfamiliarity and insufficient adaptation of system constraints to the specific task (i.e., load carriage and forced-marching), individuals potentially gravitated towards preferred attractor states attempting to correlate *preferred* or *experienced* parameter couplings with known energy cost (even if these attractor states came at greater metabolic penalty)[339, 341-345]. Likewise, the sample had little to no experience with the effects load carriage and forced-marching has on other cost functions (i.e., kinetic or postural stability). As such, the locomotor system cannot accurately generate cost gradients for each cost function landscape (i.e., their perception is altered). Therefore, the metabolic cost function was potentially weighted more greatly (due to life experience), in effort to reduce the 'dimensionality' of the motor task and improve the accuracy of their perception. Even despite the evidence within support, velocity maintenance (i.e., the goal manifold) was still likely the primary cost function and metabolic cost function secondarily weighted. However, it is likely given the magnitude of the perturbations in the present study, that cost functions would need to be weighted more deliberately and recalibrated as the task prolonged.

In addition to combating fatigue (adhering strictly to a metabolic cost function), the observed larger relative variability values may indicate state-space exploratory behavior. During the early stages of a novel or unfamiliar task execution (the present investigation would categorize as 'unfamiliar'), variability is considered task solution space exploration[367]. This 'exploration' can be thought of as an experimentation with various movement solutions to discover an optimal movement pattern[367]. Utilizing error feedback (derived from afferent sensory information) the locomotor system leverages motor variability to optimize task performance[334, 367]. However, due to failed experimental replication, rate of learning may be independent of variability observed initially[333, 367, 382-384]. Indeed, variability can be attributed to a variety of sources including sensory uncertainty, noise in motor planning, execution and muscle noise (not just as task solution space exploration [i.e., learning])[367, 382] . Initial stages of learning a novel task exhibit more random and larger magnitude excursions in motor variability as a means to intentionally explore the task solution space (especially when that null space is unknown)[367]. Importantly, these deviations aren't always Gaussian in nature, but can exhibit some structure (hence the need for temporal variability analysis)[367, 385, 386]. During the 'exploratory' phase of variability, viable task solutions are 'saved' (i.e., short-term memory) and used to construct a representation of the solution task space[367]. At the later stages of learning/exploration there is a transition to smaller scale searches of the task solution space (i.e., more refined experimentation)[367]. Some changes in execution strategy are intentional for biomechanical reasons (i.e., compensation for injury) or to interrupt psychological monotony[367].

Further, exploitation of motor variability by the locomotor system can possibly be achieved through local searches in a more expansive range of perceptual-motor workspace (such as the cost landscape) which can resolve cost estimates and identify optimal dimensions[334, 366, 379, 380]. The latter point may be reflected in the larger relative variability as the individual explores a greater range of motor solutions that achieve the task goal (null space or tangential variability) with the introduction of load carriage and imposed locomotion perturbations (which this sample was unfamiliar with).

While state-space exploratory behavior may explain observed variability and seem beneficial to the adaptation of new perturbations/constraints (i.e., load carriage and imposed locomotion patterns) it can also impede adaptation as well. An important component of motor behavior optimization is the need to identify optimal strategies quickly[366]. Given the vast number of degrees of freedom of the locomotor system, not only in terms of joint actions but motor units and neural circuitry as well; the system must efficiently determine the objective function of a task[70, 366, 387]. To complicate matters, there is no guarantee that the locomotor system will identify the optimal strategy[366]. Moreover, if the locomotor system continues state-space exploratory behavior, even if it has self-organized into the optimum strategy, then that would make it suboptimal[388]. Therefore, the locomotor system must not only identify the optimal motor strategy (specific family of solutions within the null space) but also determine when (or not to) explore for new strategies or leverage previously utilized strategies[334, 379, 389].

In the present investigation, the large relative variability may indicate too large of excursions through state-space (using too wide of an array of movement solutions). Because the sample is novice with the task, they first have little experience to draw from and direct their searches to potentially optimal attractor states (family of solutions). Secondly, the lack of experience inhibits the locomotor system organization due to the competition (or inappropriate weighting) of various cost functions. Optimal exploration of state-space utilizes small meshes and large excursions adhering to a power law distribution (fractal dynamic)[390, 391] (belying the importance of temporal organization of variability which will be discussed in a later chapter). In an effort to counteract the hinderances brought on by inexperience, the locomotor system potentially executes large searches all over state-space in the attempt to *stumble* onto the optimal attractor state (indicated by relative variability significantly increasing with increases in load magnitude and use of the imposed locomotion pattern [forced-marching]).

Noise may also contribute to observed relative variability, which is inherent to all biological systems and can take on many forms such as sensory (afferent) noise, motor planning noise or even muscle noise[367, 382]. For example, sensory information regarding shoulder pain from nociceptors due to load carriage would be considered noise when it comes to executing a stride to step over a root as it might distract from important information regarding proprioceptive spatial sense of the stepping limb. Thus, noise can disrupt information transmission/translation thereby diminishing the effectiveness of the signal (i.e., important information is lost that is necessary to the perception-action loop of motor behavior and the signal is no longer veridical)[367].

Although, noise has been commonly regarded as a nuisance to the locomotor system, some noise has exhibited structure (i.e., 1/f noise) that is both informative and beneficial[367].

Considering forced-marching is a walking locomotion pattern executed above the gait transition velocity, it requires greater muscle activation[229] and mechanical work[317] to be performed, increasing physiological noise[228]. Increased system noise from forced-marching execution and burden of load carriage (i.e., increased muscle activation and pain generating more noise) may explain why greater (>2) relative variability values were observed during forced-marching at the +55%BW load condition.

Lastly, in some instances the larger observed relative variability values can be attributed to utilizing multiple locomotion patterns, especially for loaded forced-marching. Some participants, in order to keep from drifting off the back of the treadmill would 'run' or 'gallop' for a couple strides to displace their center of mass towards the front of the treadmill (refer to Figure 5 section E). It was observed they would slowly begin drifting back (unable to maintain the necessary velocity) and would execute the aforementioned locomotion patterns (run or 'gallop') to regain their position. In some cases, participants then informed study personnel they could no longer perform and wished to terminate the trial. With the use of different locomotion strategies such as running or galloping, the stride parameter results in very different stride length and stride time combinations compared to the mean behavior (as observed in the present study and others as well[76, 77, 109, 161]). The result being a wider range of stride variations to accomplish the task goal (maintaining trial velocity) which can't necessarily be attributed to a specific locomotion pattern. However, this is important because these specific findings indicate the lack of feasibility of forced-marching to ambulate at higher velocities for prolonged periods with load (i.e, individuals need more than one locomotion pattern to execute the task).

Interestingly, when considering the effects of time, there were no significant differences between the relative variability during the first 30% of the trial versus the final 30% of the trial for any condition. These results are consistent with previous findings of relative variability during reaching tasks where fatigue did not change deviations tangential to the goal manifold and only slightly increased deviations perpendicular[227]. This lack of statistically significant differences may indicate that during prolonged gait tasks, it is not the relative variability, but other factors which are modulated during the execution of a task. Moreover, the absence of any significant findings when examining the mean variance of one portion of an entire trial and comparing it to another portion, further demonstrates the importance of examining the temporally relevant (i.e., interval to interval fluctuations) component of movement behavior when evaluating a repetitive task, as a means to quantify regulation strategy and qualitatively explain source of observed variability.

A primary limitation of previous load carriage investigations have been the absence of female sex representation or direct sex comparison[8, 106]. In the present investigation, sex appeared to play a complex role in relative variability (**Specific Aim 1.2**). Interestingly, when stratified by completion ability, no significant differences of note were observed *between* groups in regard to relative variability (**Specific Aim 1.3**, failing to support hypothesis1.3a). When assessing the entire trial there were no observed differences between sexes (failing to support hypothesis 1.2a). However, during the first 30% of the trial women had greater relative variability than men at BW independent of locomotion pattern, contrary to our hypothesis. Likewise, relative variability increased with load for women (by contrast for men relative variability remained unchanged across load conditions). By the final 30% of the trial, any differences between sexes were no longer apparent, as evidenced by the lack of statistically significant findings. Thus, at first women utilized a greater range of stride time and stride length combinations (greater variation tangential to the goal manifold) to accomplish task execution.

Importantly, women demonstrated a significant difference between running and forced-marching whereas men had no significant differences between locomotion patterns (in support of hypothesis 1.2b). This may be due to the potential physiological differences between men and women, notably less fat-free mass and muscle power observed in women[110], with women having to rely on varied coordinative patterns to effectively execute the task and attempt to delay the effects of fatigue. Furthermore, forced-marching has been shown to shift mechanical work proximally to the hip in women[317], a joint that is commonly, with respect to the anatomical structure, shaped differently from men [392-397].

In particular, the hip joint morphology for women is different than men most notably in geometry (shape): women typically have a greater q-angle altering alignment of the lower extremity[392, 393], and smaller femoral head which can contribute to more mechanical stress[394, 397], along with acetabular differences such as roof angle that results in dysplasia (more dysplasia observed in women), different dimensional ratios of the pelvis[395] contributing to hip joint difference. Thus, anatomical differences contribute to altered frontal and transverse plane mechanics of the hip[398]. Indeed, women exhibit less frontal dynamic joint stiffness with increased hip adduction and internal rotation compared to men[398]. Furthermore, women demonstrate greater late swing phase hip adduction compared to men[399]. Importantly, late swing phase hip adduction is predictive of stance phase peak hip adduction[399]. Moreover, women exhibit greater peak hip adduction (3.5º more) and internal rotation (3.4 º more) compared to men during running tasks[400].

Given the anatomical and physiological differences between men and women[110, 392-397], how those influence the source of variability remains unclear. Due to the difference in anatomical structure women may have to execute state-space exploratory behavior to identify the optimal attractor state which is evidenced by women exhibiting greater relative variability during the first 30% of the trial compared to men. Therefore, significant differences in relative variability responses to load magnitude were observed between men and women potentially because women have an anatomical disadvantage at the hip compared to men, which are further exacerbated by the execution of forced-marching. Thus, compensatory or maladaptive movement patterns about the hip (result of greater movement variability) in conjunction with increased mechanical work potentially contributing to the greater prevalence of hip musculoskeletal injuries observed in women compared to men in the military [14, 23].

Exploratory behavior implies (subconscious) intentional increase in variability, but the source may be completely unintentional. An investigation by Patil et. al (2019)[192] demonstrated that the pelvis geometry of the individual can change the amount of observed variability. Moreover, additional external factors such as the treadmill dimensions can further influence the amount of observed motor variability[257]. Thus, to definitively conclude that a particular source of variability is being observed, all sources of variation need to be discriminated, therefore long-term longitudinal studies with eloquently arranged independent variables are required to elucidate motor variability[367].

As stated in the beginning of this section of the chapter (and not discussed immediately because many theoretical foundations needed to be presented first), the findings of the present investigation were divergent from those in a similar investigation[109]. Specifically, the previous investigation observed a decrease in relative variability with an increase in load[109] whereas the present observed an increase in relative variability as load increased. These conflicting results Could be due to two factors: i) trials in the present investigation were considerably longer (10 minutes vs 1.5 minutes), and ii) The previous investigation walked for 30 seconds at a velocity

10% below their gait transition velocity then transition to 10% above gait transition velocity and remained at that velocity for 1 minute (only the final portion was analyzed) whereas the present ramped up to the trial velocity and then the 10 minute trial began. It is likely that the 30 second period below gait transition velocity could have entrenched the individual in an attractor state optimized for a true walking condition (as they were unexperienced with load carriage). Thus, once immediately thrust into the new locomotion velocity, the individual was reluctant to engage in state-space exploratory behavior (reflected in lower relative variability as load magnitude increased).

5.3.3 GEM Subspace Variance

To further elucidate findings of relative variability (**Specific Aim 1.1**), the variability in the tangential (null space) and perpendicular (task space) directions were examined as well. Tangential variability (‘good’ or goal-irrelevant variability) was always greater than perpendicular variability (‘bad’ or goal-relevant variability) as to be expected with healthy fully ambulatory populations[74, 77, 109] (and in support of hypothesis 1.1c). Independent of locomotion pattern tangential variability increased as load magnitude increased. Conversely (and in direct contrast of hypothesis 1.1d), perpendicular variability decreased as load magnitude increased (See section 5.3.2 for detailed explanations of this behavior).

The increase in tangential variability while perpendicular variability simultaneously decreased with load magnitude increases, helps explain why greater relative variability values were observed for heavier load conditions (relative variability calculated as tangential variability divided by perpendicular variability). As the perturbation of load became greater, individuals utilized more of the null space (i.e., more variations tangential to the goal manifold). Moreover, this behavior may be indicative of: i) cost function competition, ii) state-space exploratory behavior, and/or iii) utilizing multiple locomotion patterns during a single trial (refer to the previous section for more detail).

Likewise, forced-marching yielded greater tangential variability whereas running exhibited greater perpendicular variability (in contrast of hypothesis 1.1e). However, time only affected perpendicular variability (independent of load and locomotion), with the final 30% being greater than the first 30% of the trial. The latter results reinforce previous findings which have also demonstrated, that over a similar time period, tangential variability was unaffected while perpendicular variability slightly increased[227]. The observed changes in directional variability with more difficult or constrained tasks (i.e., loaded forced-marching) may indicate a strategy to reduce site specific fatigue (by utilizing more goal-irrelevant variations)[74, 227]. Furthermore, goal-relevant variation was restricted to reduce the likelihood of task failure (i.e, reducing the chance of too many variations that fail to maintain trial velocity)[109]. Lastly, the change may be a result of exploratory behavior due to the samples' task naivety. Regardless the definitive source of changes in variability remains unclear.

5.3.4 Regulation

When assessing the entire trial stride regulation (**Specific Aim 2.1**), load magnitude had no effect on tangential variability stride regulation (in support of Hypothesis 2.1a). Thus 'good' variability (task irrelevant/null space) was regulated the same regardless of the load condition. However, forced-marching (0.85±0.11) demonstrated significantly greater alpha coefficients than running (0.81±0.11). Given the small difference, while it is a statistically significant difference, it may not be a clinically, or behaviorally meaningful difference as both values are considered optimal. By contrast, perpendicular variability (‘bad’) was regulated much more strictly compared to tangential variability, consistent with previous findings regarding treadmill-based gait tasks[59, 74, 76, 77, 125, 253] (supporting hypothesis 2.1a). Moreover, as load increased stride regulation was controlled more tightly (i.e., strides were corrected immediately with opposite behavior) (in support of hypothesis 2.1b). Likewise, forced-marching yielded stricter stride regulation compared to running (supporting hypothesis 2.1c).

The regulation of repetitive movements results in constraining the task/goal relevant fluctuations (those that impact the achievement/accuracy of executing the task goal [i.e., perpendicular to the goal manifold]) while allowing redundancy in the task irrelevant (tangential) fluctuations (those that do not interfere/fail to execute the task goal)[351]. The feedback parameters (various streams of sensory information from recurrent processing) are tuned to regulate these task subspaces rather than suppress variability altogether[351]. Optimal feedback control postulates that the locomotor system attempts to determine the best regulation strategy for a given activity (aspect of the task that should be considered the goal at which to modulate behavior around)[351].

Not only is variability controlled/regulated in general but fluctuations are regulated interval to interval leaving (subconscious) decisions/actions to the last possible moment to leverage a greater magnitude of sensory information to inform the choice[351]. A ‘minimal intervention principle’ (MIP) theorizes that the locomotor system minimally intervenes with variability regulations, especially task/goal irrelevant fluctuations[351]. Therefore, a MIP driven locomotor system is only concerned with modulating deviations that directly affect the task goal reflected as alpha coefficients (from DFA) of 1 and 0.5 for null space (tangential) and task space (perpendicular) variability respectively[74, 351].

With forced-marching there was a greater focus on quickly correcting errant strides (anti-persistent behavior in the task space [perpendicular] variability). Interestingly, forced-marching elicited a minimal intervention strategy (as evidenced by greater statistical persistence of tangential variability). Conversely running allowed task relevant strides to persist more at least in terms of statistical significance. However, forced-marching alpha coefficients (0.58±0.11) may not have been clinically or behaviorally different from running alpha coefficients (0.63±0.10). While some would argue that forced-marching demonstrates more stochastic regulation of perpendicular coordinates (i.e., task space or goal relevant motor solutions) and running is actually persistent regulation, this interpretation is completely dependent on which investigation you reference. Indeed, this finding highlights one of the limitations of assessing temporal variance with DFA, as it is still not clear from the extant literature, what the *true* cutoff values are to indicate change in behavior.

Ultimately when assessing the entire trial, healthy, fit recruit-aged individuals (which this sample represents), adhere to a goal manifold regulation strategy[74]. Stride combinations that achieved the task goal of trial velocity (goal-irrelevant) were not immediately corrected and allowed to persist, thus were loosely/weakly controlled. Regardless of gait velocity, load magnitude and locomotion pattern, individuals utilized a regulation strategy adhering to the goal manifold, meaning they adjusted their steps based on maintaining velocity and not another component such as a specific position on the treadmill[74]. Moreover, the GEM regulation strategy primarily limited goal-relevant (perpendicular) errors and slightly over-corrects these deviations (evidenced by the alpha coefficients ≤.5 [refer to Table 9]), but loosely/weakly controls goal-irrelevant (tangential) variations (indicated by the alpha coefficients ~1 [Table 9])[74]. Thus, the locomotor system exploits movement redundancy (i.e., variations that are goal-irrelevant) allowing persistence and selectively regulates variability, most notably goal relevant or variations that don't achieve the task goal (trial velocity)[74]. Therefore, examining the temporal correlation of stride parameters (i.e., SL, ST, etc) independently is insufficient for describing the dynamics of control[74]. Moreover, observing coupling of stride parameters (i.e., variance ratios) can yield misleading interpretations when considering *control*[74].

Unlike relative variability which remained mostly unaffected by time, stride regulation did significantly alter over time. For tangential and perpendicular variability, regulation became more strict as the trial progressed, consistent with previous research[227]. An investigation by Gates et al (2008)[227], demonstrated during a reaching task, as individuals fatigued, their regulation strategies altered slightly resulting in quicker corrections of errors. Therefore, individuals modulate movement variability in response to fatigue but preserve the goal relevant aspects of task execution[227]. Similarly in the present investigation, during the final 30% of the trial, individuals utilized a stricter stride regulation control, correcting strides immediately rather than letting that variation persist for several strides. Loads ≥45% of BW significantly increase mechanical work, especially when utilizing a forced-marching locomotion pattern[317], thus potentially increasing muscle activation which leads to greater physiological noise[228]. The greater mechanical demands compounded by the associated increased magnitude and onset of fatigue from prolonged gait, may further increase physiological noise, which in turn yields stricter stride-to-stride regulation[76]. Moreover, as fatigue begins to accumulate, individuals' control strategy slowly moves away from goal manifold regulation (however not completely) towards prioritization of regulating variability, with the locomotor system interpreting variability as a constraint that must be overcome, and thus resulting in quick corrections (in this model anti-persistence, alpha~.5, would be observed for variations perpendicular *and* tangential to the goal manifold)[74]. This stricter control is important as it requires greater ‘attention’ or workspace capacity devotion to regulating how the stride was performed exactly. As a result, perception of other stimuli may be inhibited.

Lastly, completion group had no effect on regulation strategy (**Specific Aim 2.3**, failing to support hypothesis 2.3a), whereas sex (**Specific Aim 2.2**) had minimal influence on stride regulation and significant differences were only observed for the entire trial of tangential variability (in contrast to hypothesis 2.2a). Firstly, at no point was a minimum intervention principle observed for either sex (tangential alpha coefficient of 1.5 and perpendicular alpha coefficient of .5) which would have meant that *only* goal-relevant (perpendicular) variations were controlled and goal-irrelevant (tangential) variations were completely uncontrolled[74]. As load magnitude increased for women, their alpha coefficients increased (less strict control). Likewise, women, had looser/weaker control compared to men at the +55%BW condition. Therefore, the regulation strategy exhibited in women most closely mimicked an over-correction model (during +55%BW), where deviations perpendicular to goal manifold are *over-corrected* in the successive stride as indicated by an alpha coefficient <.5[74] (women had an alpha coefficient of 0.43±.08 compared to men having .51±.13 [refer to Table 9 forced-marching Perpendicular α]). Men by contrast (during +55%BW) utilized a regulation strategy that more closely mimicked a goal-manifold strategy operating around a preferred operating point (i.e., stride length and time variations similar to mean behavior) which is characterized by alpha coefficients of 1 and slightly above .5 for tangential and perpendicular respectively[74].

A preferred operating point strategy may ignore/reduce the weight of the task goal function (i.e., trial velocity) in favor of acting at/around their *preferred* mean spatiotemporal behavior for the given task, even if it comes with penalty (such as not meeting the trial velocity)[59, 74]. The implication for men is that this control/regulation strategy is suboptimal as motor behavior is not regulated about a truly optimal attractor state (family of solutions within the null space). If the locomotor system imposes greater control (in both the goal relevant [perpendicular] *and* irrelevant [tangential] domains) then it will generate a larger corrective control signal[351]. Likewise, noise and effort are both control dependent and thus greater control could result in an increase for both signals (more data to transcode, interpret and filter)[351]. An optimal controller allows greater variability (redundancy) in the task irrelevant domain (null space) and reduces the variability observed in the task relevant domain[351]. By contrast, the suboptimal controller yields equal variance in both domains[351]. Thus, the optimal controller uses the redundant dimension as a 'noise buffer'[351]. Although these are simply theoretical computational representations of motor control, they should be considered nonetheless when observing the results.

5.4 Complexity

Variability in motor behavior exemplified by fluctuations in stride dynamics, such as spatiotemporal characteristics (i.e., stride time, stride length, stride speed, etc) has been utilized to successfully differentiate between healthy and neurologically impaired populations[81, 119, 252, 401, 402]. Importantly, this aforementioned research has demonstrated that variability is not just random system noise that simply needs to be reduced but can indeed have structure[81, 119, 252, 371, 401, 402]. Thus, the scientific interest in motor variability has re-emerged for its potential to elucidate the state of the locomotor system[72, 225]. Consequently, some motor variability research has transitioned from clinical populations to discriminating healthy populations as a means to potentially determine potential markers of musculoskeletal risk[109, 362].

Standard deviations are commonly used to describe variability of data but they only represent a magnitude of difference with respect to 'mean' behavioral representations, and do not explain how fluctuations are impacted by the previous intervals[74, 234]. Therefore, to make better use of the variability construct and determine its efficacy with respect to the current study aims, the temporal correlation and fractal dynamics of spatiotemporal parameters, detrended fluctuation analysis (DFA) and Higuchi's box-counting method were all performed to derive scaling exponents (α) and fractal dimension (FD) respectively on the stride time, stride length and stride speed data series. DFA was used to assess the long-range correlation (predictability) of the time-series whereas fractal dimension was used to measure roughness (complexity). Load magnitude, locomotion, sex and ability to complete the task all had varying effects on α and FD. As stated earlier, each outcome will first be discussed independently examining the entire sample and then examining the between group factors of sex and completion ability.

5.4.1 Long-Range Correlations

Firstly to address **Specific Aim 3.1**, there were no significant effects of load or locomotion for stride time or stride speed on the long-range correlation(s) (scaling exponents derived from DFA) (failing to support hypothesis 3.1a). Moreover, the majority of observed α were in the pink noise (optimal) range of 0.75-1.0 suggesting optimal long-range correlation (see Table 10). From a mathematical perspective, long-memory (long-range correlation) in a time series is characterized by a covariance of two intervals separated by n samples that cannot be summed over n (summation of the auto-correlation function is not finite)[403]. Thus, long-range correlation indicates extent to which the auto-correlation function of the time series decays slowly and persists over a large range of consecutive intervals[403], among many time scales, and not just the short-range (i.e.

previous interval)[276]. This self-affine property, commonly, albeit incorrectly, referred to as self-similarity[404], drives scale-invariant property of fractals, as the absolute scale is not the determining factor of the geometric shape, but the ratio of scales[273]. With stride time and stride speed time series exhibiting fractal behavior (α=0.75-1.0), the scale independent behavior suggests that a perturbation in one sub-system (element of a complex system) is less likely to permeate/affect the emergent property (stride) of the global system (locomotor system) negatively[62-64, 181]. This observation of optimal long-range correlation is not surprising given the participants were all healthy, physically fit, young adults.

From a control/stride regulation perspective the findings of the present study indicate that fluctuations of stride length and stride speed are free to persist, implying looser/weaker control of these parameters[59, 74, 76, 77]. Indeed, α=0.75-1.0 is considered an optimal level of persistence as it closely mimics a minimal intervention principle (refer to section 5.3.4 for detailed explanation of this principle), where variability is leveraged to accomplish the task goal rather than treated as an obstacle that must be overcome (prior understanding of motor control posited that the focus of the locomotor system was to reduce variability to zero)[59, 74]. However, the 'computational' effort required for the locomotor system to eliminate *all* variability in the system would be enormous given the degrees of freedom (and therefore impossible)[59, 74]. But allowing some degree of variability to persist (evidenced by α=0.75-1.0) is less computationally expensive and affords the locomotor system computational workspace to control parameters that may yield potentially deleterious consequences on the system[59, 74].

Previous computational modeling of gait, has illustrated that speed regulation enhanced the global stability of an individual's gait, compared to position regulation[310], which may potentially explain why individuals prioritize speed regulation stride-to-stride (reflected in optimal

α=0.75-1.0 for stride speed). The global stability refers to the size and shape of the basin of attraction in state-space, meaning if unstable/shallow, perturbations can push the locomotor system into another attractor state (potentially suboptimal family of solutions for a given task)[310]. By contrast, position regulation (reflected as $\alpha \geq 1.30$ for stride speed) improves local disturbance (decay of perturbation) rejection better than speed regulation, but the global stability is reduced as a consequence (stability of the attractor state)[310]. Therefore, task-level (i.e., maintaining speed, heading direction, etc.) potentially enhances global stability by improving perturbation rejection[310]. With participants exhibiting α=0.75-1.0 for stride speed time series, they may have been managing the task-level in an effort to improve global stability and reduce the likelihood of a task failure (i.e., slip, trip, fall from an object/obstacle [perturbation]).

The findings, with respect to stride time and stride length long-range correlation (**Specific Aim 3.1**), observed in the current investigation, support both the theoretical assumptions and results previously observed from comparing healthy versus clinical populations[81, 119, 252, 401, 402]. However, in regard to the effects of load magnitude and locomotion pattern, these results are in direct contrast to previous findings (and hypothesis 3.1a), which observed a loss of long-range correlation as load increased and specifically for forced-marching[109]. Although differences in results were observed between studies, the aforementioned study[109] didn’t have an adequate number of strides (<512) to confidently assess the long-range correlation of gait spatiotemporal parameters potentially yielding false positives (as stated in their limitations). Therefore, the findings of the present study can be stated with more certainty, given that stride counts were well above the 512 strides required (1265±295 strides across all trials for the present study) [113].

Unlike stride time and stride speed, stride length was significantly impacted by locomotion, with forced-marching demonstrating greater (~11%) long-range correlation than running, counter to our hypothesis 3.1a (refer to Table 10). It is unclear as to why forced-marching (considered an unnatural imposed locomotion) yielded greater long-range correlation (closer to the ideal α=1.0) but may be due to the motor driven treadmill. The locomotion task in the present study required individuals to ambulate at a constant velocity with altered conditions (load magnitude and locomotion pattern) separately for ten-minutes at a time. Due to the belt moving under its own power, individuals potentially did not have to generate the necessary equivalent propulsive forces to translate anteriorly, compared to overground ambulation. Furthermore, since forced-marching is a walking gait pattern, there is a reduction in vertical (superior-inferior) center of mass displacement over the course of a stride[405]. Conversely, running would yield greater vertical center of mass displacement[405] which can be more fatiguing with load carriage as the additional load requires greater mechanical work[92, 97, 103, 317] to overcome its momentum (aligned with the vector of gravity). Likewise, in managing the load's momentum, the degrees of freedom must be frozen or reduced for certain parts of the body[406], requiring more control. Thus, forced-marching may have a lesser task dimensionality (less variables to be managed to successfully execute the task) compared to running in this specific scientific setting.

When comparing sexes (**Specific Aim 3.2**), statistical analysis revealed interesting results of long-range correlation. Women demonstrated an increase in long-range correlation of stride time/stride length time series as load increased and greater long-range correlation than men for both loaded conditions (+45%BW and +55%BW) [refer to Table 10]. This finding failed to support our hypothesis 3.2a, which suggested there would be no statistically significant differences in alpha coefficients between sexes. However, the observed differences may be a consequence of the physiological differences between sexes. Women, although smaller in stature (observed in the present study as well) with less muscle mass compared to men [304, 305], typically have greater mitochondrial density in skeletal muscle[306], capillary density[304, 307] and enhanced lipid binding transport proteins[308]. As such, women are more reliant on fat oxidation energy pathways[306] yielding greater local muscular endurance. Importantly, the inclusion of load carriage increases the metabolic/physiological demands of locomotion[26, 84-87, 100, 101] taxing the aerobic pathways greater than anaerobic pathways. Nevertheless, women are potentially able to leverage their physiological advantages through better fatigue maintenance via robust oxidative pathway architecture[306, 407] compared to men. With an increase in local muscular fatigue for men[407], specifically in the lower extremities, the fatigue disrupts cooperation between sub-systems resulting in less predictable output of the emergent behavior (i.e., α trending towards 0.5 or completely stochastic behavior).

In a military setting absolute loads are used (typically between 20-60kg depending on mission) regardless of an individual's size, and women are generally smaller in stature than men. In the present investigation relative loads were used to understand the degree to which load magnitude affected the locomotor system and compare between sexes controlling for differences in size. The women in the present investigation had significantly lighter loads compared to the men (see Table 1), with their +55%BW (~34kg) failing to achieve the greatest loads observed in military settings (60kg). Therefore, different outcomes could be observed for women (i.e., potentially $\alpha \leq 0.5$) when they are required the maximal/absolute military relevant loads.

Lastly, when assessing the ability of an individual to complete the full ten-minute trial by comparing completers versus non-completers (**Specific Aim 3.3**) there were no significant differences for any of the spatiotemporal characteristic (ST, SL, or SS) long-range correlation. It was hypothesized (hypothesis 3.3a) that an inability to complete the task would yield over-regularity (i.e., $\alpha \geq 1.30$ [brown noise]) with the spatiotemporal characteristics showing a distinct trend in their fluctuations overtime. Indeed, brown noise denotes that each successive increment is dependent on the previous[78] meaning a fluctuation in one direction is followed by a fluctuation in the same direction. Importantly, brown noise is characterized by a powerful low frequency that dominates the signal. Hence, an error in one sub-system permeates across the global system altering the emergent property to exhibit over-regularity, for example low functioning heart capacity yields brown noise heart rate variability[122]. The explanation for the observation of the present investigation is likely due to multiple reasons: i) long-range correlation does relate to the ability to execute a task for a prolonged period of time, ii) spatiotemporal characteristics may not be the correct emergent properties to assess for task execution capability, and iii) no participants outright failed the task (i.e., did not fall over or drift off the back of the treadmill) but rather indicated to the study personnel they could not continue.

5.4.2 Fractal Dimension

When assessing fractal dimension (via Higuchi's box-counting method) [**Specific Aim 3.3**] effects were observed for all three spatiotemporal parameters (stride time, stride length and stride speed). For all spatiotemporal parameters, running exhibited greater fractal dimension than forced-marching, especially during the loaded conditions (+45%BW, +55%BW) [see Table 11]. This result is the opposite observed for alpha coefficients derived from DFA and failed to support hypothesis 3.1b. Fractal dimension is a measure of 'roughness' (higher values indicating more roughness) [representing a local property] or short-term irregularity, while alpha coefficient (from DFA) is a measure of long-range correlation (representing a global characteristic)[277]. Many previous models have defined DFA and FD as having a linear relationship, as it is theorized that the state of the local property is reflected in the global characteristic; however caution should be used with this interpretation as the two values can be independent of one another[277]. Indeed, in an investigation of forward versus backward walking, fractal dimension was altered independently of the alpha coefficients[408].

Whilst forced-marching and running locomotion patterns had fractal dimensions (~1.77 and ~1.86 respectively) near values related to pink noise (FD=1.8) or optimal complexity, forced-marching trended towards brown noise (FD=1.5)[280]. Given that fractal dimension represents the short-term irregularity (amount of variance of immediate fluctuations retained in a smaller observation window)[280], a larger fractal dimension may indicate more complexity where a participant has greater capacity to alter their stride from one interval to the next (adaptability/flexibility)[408]. Too much irregularity (fractal dimension ≥2) may represent instability as each successive stride has a large, random fluctuation (i.e., drunk individual walking), a behavior ill-suited for handling perturbation[408]. Likewise, too little variation would suggest that the participant lacks flexibility (in terms of lack of array of movement solutions) [i.e., robotic movement] making them less adaptable to perturbation[408]. Thus, the reduction in local variance (lower fractal dimension) may indicate minor instability to certain perturbations, but also yield more predictable behavior (greater long-range correlation [larger alpha coefficient values]) and explain the observed divergence in ‘complexity’ measures for forced-marching.

When examining the effects of sex (**Specific Aim 3.2**), failing to support hypothesis 3.2b, women exhibited a lower fractal dimension compared to men for stride length and stride speed time series (there were no effects of sex for stride time, in direct contrast with long-range correlation findings) [see table 10 and 11]. Once again, these results demonstrate divergent behavior from the long-range correlation findings reported earlier. where women had greater values than men. Although enhanced local muscular endurance capabilities in women relative to men[407] may have attributed to improved long-range correlation, the relative lack of strength/power[409, 410] and altered hip anatomy[392-397] may have acted to influence and reduce the fractal dimension observed. Relatively, women exhibit lower muscle force production capabilities compared to men[409] and certain stride patterns may require a greater mechanical power output to perform. Thus, in conjunction with the alterations due to hip anatomy, women potentially have a reduced set of stride patterns (constrained motor solutions), available to utilize to execute the task. Importantly, this reduced movement solution capacity potentially acts to decrease the local adaptability, making the women more susceptible to task failure from additional perturbation (such as maintaining velocity and avoiding debris).

Lastly, when comparing completion group (completers versus non-completers), completers exhibited greater fractal dimension of stride time compared to non-completers (there were no effects of group for stride length or stride speed). This finding supports the previously mentioned notion that fractal dimension represents stride adaptability[408]. When healthy individuals walk backwards their long-range correlation is unaffected, but their fractal dimension is reduced[408]. Individuals are at a greater risk of falling when moving backwards compared to forwards because they have less movement options (reduced degrees of freedom / not analogous to their everyday walking) to overcome a perturbation (less adaptability)[411]. Therefore, in contrast to long-range correlation, fractal dimension may present a better marker for prolonged task execution capability. Despite fractal dimension assessing the short-term irregularity (roughness) of a time series[276-278] the lack of adaptability in local variance foreshadows the locomotor system’s inability to later rely on other variations to continue successfully executing the task[408].

5.5 Limitations

Prior to the global interpretation of the results (all outcomes considered together in Chapter 6), the limitations of the present investigation must be mentioned. The primary limitation of this investigation was small sample size for sex comparisons. An a priori power analysis determined that 180 participants (90 men and 90 women) would have to be recruited to examine sex differences. Due to available funds and pragmatic considerations this was deemed unobtainable. Thus, power calculations were generated based on the effects of load and locomotion to ensure adequate power. As a result, sex was determined a priori to be a secondary exploratory analysis. Moreover, the effect of completion group was also inadequately powered (hence the tertiary exploratory analysis designation). Therefore, all results regarding the between group factors of sex and trial completion should be interpreted with caution. Future, research should focus on obtaining larger sample sizes to verify the findings observed in this investigation regarding the aforementioned between subject factors.

Additionally, there are some limitations regarding the ecological validity of the study. Firstly, in some respects the investigated sample represents a military recruit population (i.e., age, stature, relative fitness and lack of load carriage experience). However, not represented in our sample is the lower end of the fitness spectrum (by design of the inclusion/exclusion criteria). Indeed 17.4% of the military is classified as obese[412] whereas none of the participants in the current sample were classified as obese (by BMI standards). A fitter sample was determined to be the more practical starting point of assessing temporal variability with load carriage. Many analysis techniques require large numbers of consecutive data points to return valid results (i.e., DFA needs at least 512)[113, 408]. Fitter individuals were more likely to successfully execute the full ten-minute trial (or at minimum enough time to be included in the analysis). Therefore, results of the present study can only be generalized to men and women on the healthier/fitter end of the spectrum.

Thirdly, concerning ecological validity is the use of a motorized treadmill. The motorized treadmill presents a few potential issues. Firstly, because the belt is driven by a motor and not the individual it can alter biomechanical characteristics[413]. Specifically, individuals will not have to generate as much propulsive force since the belt translates under them, but instead just need to ensure that their feet match pace with the belt to keep from drifting back[413]. Although these effects may be negligible, they alter motor behavior nonetheless and potentially reduce the dimensionality of the task (which in turn can result in differences in control/regulation strategies). Secondly, the motor driven belt can influence rhythmicity of gait acting as a sort of metronome for the user, which in turn may influence temporal outcomes[258, 414, 415]. Lastly, the dimensions of the treadmill itself are confined, restraining individuals' range of movement in terms of medial-lateral and anterior-posterior drift. Thus, the treadmill can alter the amount of motor variability[257], complicating the interpretation of results since there is an additional external source exerting a degree of control on the behavior.

In terms of military relevance, the laboratory setting is very controlled compared to those ground close combat personnel operate in. In essence (and this applies to most motor control research) the controlled setting reduces the dimensionality of the task, making the motor control problem 'simpler' to solve and as such the motor behavior may be different than what would be observed in a natural setting. Specifically, military load carriage (even recreational load carriage such as backpacking) is commonly performed over varying or mountainous terrain which has ever changing surface orientations, restitutions, and random obstacles to account for. Further, in theaters of war, military personnel will also need situational aware to identify potential combatant risks (i.e., human or IED). Lastly, individuals in position of command (i.e., sergeant) will need to account for all of these and make decisions for the entire unit. The present investigation can really only be generalized to training road marches (which will still have varying terrains from potholes). Therefore, this research should be replicated on a self-driven treadmill or with wearable sensors in a stimuli rich environment to confirm that findings are observed in more ‘natural’ settings.

6.0 Conclusion

While the results of the present investigation may have failed to support the stated experimental hypothesis, they do however support findings presented by previous research[74, 76, 77, 79-81, 246, 252, 401, 402, 408, 414]. Furthermore, the findings observed do present a few important implications to be considered for military relevant populations/settings. Importantly, healthy, recreationally fit recruit-aged (18-35 years) men and women adopt a goal manifold relevant control strategy during *treadmill-based* gait tasks[74, 76, 77] (**Specific Aims 1.1 and 2.1**). Despite the introduction of novel/unfamiliar perturbations and constraints of load carriage and imposed locomotion patterns (forced-marching), individuals exhibit optimal temporal arrangement of spatiotemporal parameter (stride time and stride length), coupling parameter (stride speed) and subspace parameter (tangential and perpendicular coordinates) time series (**Specific Aim 3.1**). Combined, these results indicate individuals within these population parameters (See Table 1 for participant characteristics of the current sample) possess degenerate locomotor systems (especially compared to clinical populations)[81, 125, 252, 401, 416]. In the laboratory setting (controlled and dimensionally constrained) the current sample broadly exhibited optimal self-organization of their locomotor system[225, 242, 247]. Careful examination of the confluence of findings however reveals important discriminations to be considered further, especially when generalizing to a more dimensionally rich natural settings.

Firstly, to comprehensively interpret the assemblage of the dynamic outcomes in the present study, the concept of complex systems must be revisited. Complex systems (like the locomotor system) incorporate many elements that operate on different timescales, evolve over time (dynamical) and exhibit nonlinear (chaotic) behavior [meaning the predicted outcome of a signal cannot be derived from a simple summation function][56, 59, 114, 176-181]. This chaotic behavior has been demonstrated eloquently with simple systems (have a limited number of elements, especially compared to the human locomotor system) that exhibit complex behavior (stochastic and deterministic)[65]. Due to the numerous elements and their vast degrees of freedom within a complex system, every aspect cannot be unilaterally controlled (the computational requirement alone for this complete control would likely bring about system failure)[311, 351]. Therefore, a complex system must construct a multidimensional state-space from: i) synergistic movement solutions (family of solutions)[224, 337], multisensory information[164-168], iii) task/goal manifolds[59] and iv) cost function expense gradients[346, 355-358, 417]. Within the state-space construct, based on the aforementioned aspects, various attractor states are formed that provide the locomotor system ranges of behaviors to utilize. These attractor states then present the locomotor system with the opportunity leverage variability with minimal control effort[311, 351]. However, not all attractor states are optimal, nor is state-space accurately/appropriately constructed to achieve optimal task performance outcomes[51, 53, 165, 168-172].

A complex system that exhibits ‘complexity’ is critically self-organized, meaning independent subsystems (elements) become correlated acting in concert to produce an emergent behavior[63]. Complex signals (i.e., output of the emergent property, such as stride length time series) exhibit fractal characteristics from a temporal perspective where an interval in the present is influenced by an interval in the remote past[234, 273, 418]. Thus, it demonstrates long-range dependence or long-term memory[78]. The 1/f or pink noise structure (α=1.0) of a signal exhibits an evenly distributed power across the entire spectrum and not a specific frequency[78]. Therefore, fluctuations at one time scale are weakly correlated at other scales, suggesting local perturbations are less likely to disrupt the entire system due to the scale independence of all elements (sub-systems) within the global system[62, 78]. Furthermore, a system with complexity leverages motor variability, in the form of utilizing families of motor solutions (attractor states) for a motor task problem, to provide stability and flexibility to perturbation/constraints (i.e., degeneracy)[251, 327, 328, 331, 332, 419, 420]. Likewise, complexity will manifest in the accurately calibrated and constructed state-space for a given task due to all the elements in the complex system self-organizing in a heterarchical manner.

In the present investigation when assessing from the dimensionally constrained perspective (i.e., laboratory setting relevant, which may explain the lack of support of our hypothesis), the current sample demonstrated complexity in the locomotor system (See Tables 10 and 11). The entire sample (including individuals that could not complete the full ten-minute trial) were able to leverage motor variability (**Specific Aim 1.1)** to maintain the trial velocity through greater use of the null space (i.e., more tangential variability), limiting task space area (i.e., less perpendicular variability) and quickly correcting errant/task goal deviant strides (i.e., α~.5) [**Specific Aim 2.1**]. Moreover, this regulation of the coupling parameter (stride length/stride time) of the task goal was not randomly executed as evidenced by the stride speed α~1.0 (thereby indicating there was a presence of an underlying deterministic process exhibiting long-range memory)[371] (**Specific Aim 3.1**). Thus, as a means to execute the specific task goal of velocity maintenance, the observance of fractal characteristics in conjunction with favorable relative variability the locomotor system organized demonstrating cooperation between independent sub-systems (elements)[252]. Furthermore, the reported high RPEs for the loaded conditions (even though velocity was only 10% above their gait transition velocity, therefore a minimal effort zone) further demonstrate that individuals optimized adaptation (structured null space exploratory behavior) to primarily meet the task goal function (treadmill velocity) and not a metabolic cost (i.e., metabolic efficiency). Had stochastic temporal structure (white noise) with large variability in *both* null space and task space domains (tangential and perpendicular variability respectively) been observed in this controlled setting, then it could be rationalized that experimental load magnitudes and imposed locomotion patterns were too significant to overcome even to meet a task goal function.

Dimensional constraints imposed by the experimental conditions in the present investigation appear to improve state-space exploratory behavior for the *benefit of achieving the task goal specifically*. The sample was comprised of novice load carriers and therefore a certain degree of adaptation would be needed to execute the task goal (note that walking and running is not a completely novel coordination pattern for this sample, so it is less likely that learning behavior was exhibited. See chapter 5.3.1 for more detail regarding this topic). Without past experience, individuals may rely solely on preferred stride frequencies (i.e., previously experienced behaviors) to achieve the task goal regardless of the increased metabolic cost[346]. However, combined with prior knowledge that the task will only prolong for ten minutes (versus potential hours and unknown lengths in military settings) individuals may unweight the importance of a metabolic cost function slightly. Individuals then explore a greater area of state-space (greater tangential [null space] variability) based on achieving the trial velocity even if those movement solutions can be metabolically/mechanically costly to the individual and inefficient in the identification of the energy optimum[366]. As such components of fatigue, kinetic consequence, pain, and postural stability may have been ignored (which would influence the state of the locomotor system more greatly during a much more prolonged execution of the task). Therefore, this sample may have prioritized task goal function to the extent that optimal motor variability (relative variability ≥1) and temporal structure was reflected in the results even during perturbed conditions (loaded and imposed locomotion conditions).

When generalizing to a ‘natural’ setting (non-laboratory), the same motor task (i.e., executing gait with load magnitude and an imposed locomotion pattern) becomes much more dimensionally rich[351]. As a result, what is observed in the controlled laboratory setting may diverge from behavior observed in the natural setting. Furthermore, organizing primarily around a task goal function of velocity may be feasible in the laboratory setting, especially considering the motor driven treadmill acts as a pacing mechanism to enhance state-space exploration, but not a natural setting. Thus, in the present investigation structured variance (i.e., pink noise or α~1.0 and FD~1.8) was observed in the null space and for coupling parameter (stride speed) time series indicating directed state-space exploration to adapt to the task (and its perturbations/constraints) [366]. In the natural setting (removal of treadmill), the individual will potentially be without an external influence to drive state-space exploratory behavior which may manifest in two ways: i) constrain motor variability to known *preferred* parameters (reducing stability and flexibility of the system to additional perturbations), and/or ii) exhibit stochastic motor variability (the temporal structure of the fluctuations resemble white noise).

Firstly, as a consequence of novelty or unfamiliarity with the task (which military recruits typically are with load carriage and the current sample reflected this aspect) individuals have no known past experience to draw from to optimize task performance[366, 417, 421]. As such, individuals then draw from experiences they do have and attempt to *force* these behaviors to work for the unfamiliar task, entrenching them in that attractor state (think trying to put the square peg in the round hole)[346, 366, 417, 421]. Importantly, individuals gravitate towards experience regarding *preferred* behavior that does not always coincide with the optimal attractor state of that task (some individuals are however, capable of exploiting natural variability)[346]. Essentially, individuals are constraining motor variability closely around *their* preferred operating point. However, by constraining the magnitude of motor variability (potentially reducing the number of motor solutions available to them in the moment) the system becomes less stable and flexible (i.e., loss of degeneracy). Therefore, with the introduction of new perturbations (i.e., a pothole) in additional to their current ones (i.e., load carriage and forced-marching) an individual may not possess the motor solution at their immediate disposal. This issue will be further exacerbated if the cost function driving locomotor system organization is a task goal function (i.e., the system in this scenario has weighted postural stability with little importance in favor of velocity maintenance and is entrenched in a suboptimal attractor state). Thus, in the present study reduction of motor task dimensionality, removal of unexpected perturbations (i.e., controlled laboratory setting) allowed the locomotor system to expand the magnitude of variability comfortably (i.e., greater observed relative variability).

Alternatively, individuals may maintain the same magnitude of motor variability in a natural setting, but the temporal structure of these fluctuations may exhibit stochasticity (i.e., white noise). Stochastic behavior is driven by random processes, meaning a series of motor behaviors are equally probable in occurring at the next interval (within the confines of the system at that present moment)[371]. Emergent property time series (i.e., spatiotemporal parameters) that have a stochastic structure are demonstrative of ineffective state-space exploratory behavior[367, 385, 386]. That is because their motor solutions are randomly actioned, successive trials are not similar or correlated inhibiting the identification of an optimal task solution[334, 374, 375]. Therefore, if an individual exhibits stochastic temporal structure in fluctuations they are potentially failing to leverage their motor variability to improve task performance through adaptation/learning[346]. In the present investigation, the motor driven treadmill may have acted as an external influence (i.e., a physical pacemaker) directing state-space exploratory behavior towards optimal attractor states as evidenced by the pink noise variance structures observed on all motor outcomes[258].

Although, the two previously mentioned potential consequences (constrained motor variability and stochastic temporal structure) can occur independently (i.e., constrained motor variability exhibiting long-range correlation and vice versa), they can be exhibited simultaneously as well. The occurrence of both phenomena is eloquently exemplified in the motor variability of patients with Parkinson's disease[401, 422, 423]. Due to neurological impairments, Parkinson's patients have disruptions in motor control exhibited as tremors in movement[401]. When fluctuations of motor behavior are quantified, the temporal correlation of fluctuations is stochastic (driven by random processes) *and* the magnitude of variability is constrained (believed to be an attempt at controlling movement better) compared to healthy populations[401, 422, 423]. Thus, in a natural setting with load carriage and locomotion pattern imposition, the dimensionality of the motor task may become so large individuals constrain the magnitude of motor variability and inadvertently organize the locomotor system so that elements no longer act in concert (i.e., stochastic temporal correlation).

Sex played an interesting role in the results of the present investigation and causality remains unclear (**Specific Aims 1.2, 2.2 and 3.2**). Men in this sample were significantly larger than women in terms of height and mass (see Table 1). Additionally, it has been demonstrated repeatedly than men and women differ physiologically, specifically in terms of body composition, bone density and muscle architecture[110, 304-308]. Importantly, these differences in system constraints lead to altered performance characteristics where in general men are stronger (greater force production) and more powerful (high velocity – high force muscle contractions)[110]. By contrast women typically have physiological architecture better suited for local muscular endurance[304, 306-308]. Moreover, women on average have significantly different hip joints with respect to anatomy, which leads to altered lower extremity walking mechanics[392-397]. The sexual dimorphism may have manifested in a few of the observed differences between sex, most notably relative variability, long-range correlation and fractal dimension.

Women in the present sample exhibited a simple main effect of increasing relative variability as load increased whereas relative variability remained unchanged across load conditions. Furthermore, during the first 30% of the trial women exhibited greater relative variability than men. Lastly, women exhibited less fractal dimension in spatiotemporal parameter time-series. Together, these results suggest that women have to rely on a larger array of motor solutions in the null space (tangential variability) and have less local or short-term adaptability to perturbation (evidenced by lower fractal dimension compared to men). These findings may be the direct result of the aforementioned physiological differences. Due to men having greater force/power generating capabilities it provides more action choices at their disposal. Moreover, the differences in hip anatomy that alter gait mechanics[392-397] may further limit the number of motor solutions available to women (at least in terms of appropriate motor solutions to achieve the task goal). This seemed to be particularly highlighted during loaded forced-marching where women had the lowest fractal dimension and highest relative variability (see Tables 7 and 11).

While it may be speculated that men have greater local adaptability due to the greater fractal dimension compared to women, when the conglomeration of results are examined, an additional competing rationale arises. Alternatively, the greater fractal dimension for men coupled with the lower alpha coefficient may indicate that men are executing an ineffective state-space search, with large excursions in the short term (i.e., varying coordination patterns) that aren't necessarily correlated or dependent on prior strides (evidenced by the lower alpha coefficient which indicates less long-range correlation)[367, 382]. Further, regulation of the tangential and perpendicular subspaces (α~1.0 and α~.63 respectively) indicated that men may be controlling behavior around a preferred operating point, regulating based on preferred mean spatiotemporal parameters rather than achieving the trial velocity (sexual dimorphism in system constraints allows men to 'overcompensate' and execute the task even with poor locomotor system arrangement). Therefore, for men (or at least the ones represented in this sample), the locomotor system may be driven by random processes and are struggling to identify the optimal attractor state[371]. Although fewer men were non-completers (3 men versus 5 women), it is possible that men were capable of completing more because of their system constraints (i.e., greater strength/power, longer limbs, etc.) but that does not necessarily mean they performed the task *better*.

Further supporting this alternative theory is that women exhibited fractal dimension (although close in magnitude to men) skewed more towards over-regularity. It is entirely possible that women (in the present sample) were executing more directed local searches to determine the optima of the spatiotemporal parameter[367, 382]. Because the local searches were more directed, women were able to draw knowledge gained from prior iterations (strides) to influence present and future motor behavior (hence the observed alpha coefficients close to 1 or 'pink' noise). This noise structure is driven by a balance between stochastic and deterministic underlying processes[371]. Perhaps these calculated local searches interspersed with large excursions is why women had greater relative variability. Through the enhanced adaptation process, women (in the present sample) identified more optimal attractor states to generate a larger family of solutions, thus constructing a more comprehensive null space as a means to *specifically* achieve the task goal (maintain velocity). However, it remains unclear why sex would alter state-space exploratory behavior and additional research is needed to elucidate the cause.

In addition to sex, another between group factor was examined; the ability to complete the entire ten-minute trial (**Specific Aims 1.3, 2.3, and 3.3**). The only observed significant difference between completers and non-completers was in stride length variance. All other outcomes (relative variability, subspace regulation, long-range correlation, and fractal dimension) failed to reveal any significant differences (and failed to support any of our hypothesis). However, when considering all the results, the difference between completers and non-completers may not have been in task execution/regulation but due to another component of the locomotor system. For each load condition a gait transition was determined by performing three separate ramped treadmill protocols (accelerating at 0.05 m/s^2). The velocity at which the individual transitioned from walking to jogging was recorded and the mean was calculated from the three trials. Experimental trials were executed at a velocity 10% above their mean gait transition velocity. It is plausible that non-completers mismatched their perceived action capabilities with the perceived effort of locomoting with load carriage resulting in an inappropriate affordance choice (transitioning at too high of a velocity)[51, 53, 165, 168-172]. This is evidenced by some individuals not transitioning to jogging until ~2.0 m/s (4.5mph) for *loaded* conditions (the addition of 45% or 55% of bodyweight), who essentially engaged in forced-marching on their own before finally transitioning. It should be noted that approach marches in the military setting where warfighters must use forced-marching can be executed up to 2.28 m/s[7]. To put in perspective, *unloaded* walking exponentially increases in metabolic cost at 2.0 m/s, thus most individuals naturally transition before that velocity for a cost benefit (reduction in energy expense per step)[424]. The result was a trial velocity that greatly exceeded the system's capability of ambulating at with the addition of load carriage for a prolonged period, especially if forced-marching (shortest completion times were observed during loaded forced-marching conditions). Therefore, the disruption was not in task execution or regulation (exhibited in similar outcomes with completers) but inaccuracy in the perception-action coupling (identifying transition velocities incorrectly)[51, 53, 165, 168-172]. Further supporting this notion is that most individuals only failed to complete trials for a single load condition, indicating that their locomotor system recalibrated their perception of the load carriage task constraints and their own system constraint capabilities.

Lastly, some motor control research has transitioned to military relevant populations to address the increase in MSI especially during load carriage activities[105, 109, 161, 362, 406, 425]. While the present study is cross-sectional in design there are a few implications for MSI that should be considered to inform future longitudinal MSI research (prospective cohort and randomized controlled trials). The most observed MSI include overuse injuries (osteoarthritis, stress fractures), strains and sprains[9-12]. For men, injuries predominantly occur at the knee whereas women experience a greater number of injuries at the hip[9, 14, 18, 19]. Aside from mechanical overuse classification of MSI, acute mechanical MSI from slips, trips and falls are the second most common classification of lower extremity MSI[9, 18, 19] (see chapter 1.1 for more detailed information regarding observed MSI in the military). The results from the present investigation may initially indicate low MSI risk based on the favorable outcomes (relative variability greater than 1, greater null space variability, pink noise structure for long-range correlation and fractal dimension)[66, 67]. However, the nuance of interpretation must be considered beyond the experimentally controlled environment.

As stated earlier, greater null space variability (tangential variability) was observed for all experimental conditions indicating stability and flexibility in motor solutions (i.e., degeneracy).

While this *might* reduce chances of slips, trips and fall, due to a more motor solution degeneracy; it is likely this range of motor variability was the result of adhering primarily to a task goal function (achieving trial velocity). As such, some of the motor solutions utilized are potentially coming with a large expense penalty for other cost functions[355-358]. Consider first a kinetic cost function, meaning using a motor solution that allows for large impact forces to be transmitted through the kinetic chain in order to achieve the trial velocity. In a short duration (ten minutes) ~1300 strides are completed (based on stride counts observed in the present study), perhaps only 5% of null space variability represents motor solutions (~65 strides) with the highest kinetic cost penalty (greatest impact force transmission). But load carriage tasks can persist for hours; during a 3-hour loaded march ~23,000 strides will be executed (shorter individuals will execute more compared to their taller counter parts), thus ~1200 strides of the highest kinetic penalty will be executed (assuming the percentage does not change with other factors that will influence the system like fatigue). If individuals (military recruits) have to focus on maintaining pace with the rest of the group adhering to a forced-marching locomotion pattern, their motor variability will likely reflect those observed in the present investigation which will include motor solutions across a large range of various cost gradients. Therefore, over time the accumulation of these interspersed large kinetic cost motor solutions could become problematic.

Recall chapter 2.1, where the elastic properties of tissue (i.e., its stiffness) is altered with consecutive repeated loadings[41, 47]. Further, with repeated loadings tissue incurs microdamage (i.e., microfractures in the bone)[47-50, 136, 141]. This can be exacerbated further if the forces are not dispersed through the supportive structure (through motor variability) and are instead relegated to site-specific loading[136, 137]. In the early stages of the task execution (i.e., first ten-minutes) utilizing motor solutions with a high kinetic cost penalty is tolerable because the elastic properties of the lower extremity tissues can withstand higher loading rates. However, at a later stage (two hours in for example) the threshold of tissue tolerance has reduced (due to changes in elastic properties and sustained microdamage[41, 47-50, 136, 141]) and that same motor solution (with a high kinetic cost) could now result in a tissue failure (such as a stress fracture). While favorable temporal correlation was observed, the significantly greater relative variabilities observed during loaded conditions potentially indicates that individuals generate a large null space (tangential variability) in order to achieve the task goal (velocity requirement) with large cost penalties in other cost function domains. Importantly, although kinetic cost is discussed here, other penalties for cost functions such as postural stability or situational awareness (enemy target and IED identification) may be incurred as well and would explain the high incidence of slips, trips and falls (utilizing motor solutions at the expense of postural stability that result in perturbations such as rocks leading to adverse events when burdened with load carriage). Therefore, utilizing a forced-marching locomotion pattern with load carriage may increase MSI risk due to prioritizing task goal achievement at the expense of properly weighting alternative cost functions (that have direct implications on MSI risk).

Many of the interpretations presented here while interesting remain speculation. Although several frameworks are compatible in terms of conceptual theories of motor control, they remain separate computational paradigms[74, 76, 77, 79-81, 246, 252, 401, 402, 408, 414] (one the difficulties in motor control research has been assimilating and comparing similarly designed investigations that often utilize varying computational methods of quantification). Given the complexity of motor behavior, a single computational framework will likely never fully encapsulate the behavior quantitatively. Alternatively, further work is needed to confidently parse sources of variability to establish the validity of the conjecture presented here[367]. Future investigations should delimit independent variables and utilize clever, eloquent longitudinal study designs to elaborate on the findings. However, in healthy, recreationally fit recruit-aged men and women, in a controlled, treadmill-based setting (dimensionally constrained) the locomotor system can adapt (evidence by long-range correlations and fractal dimension) to perturbations of load magnitude (up to +55%BW) and imposed locomotion patterns (**Specific Aim 3.1**). Likewise, with introduction of perturbation, the locomotor system can expand the null space (tangential variability), while constraining and tightly controlling the task space (reducing perpendicular variability and α~0.5 for perpendicular coordinate time series respectively) to achieve a task goal of maintaining a specific velocity (**Specific Aims 1.1** and **2.1** respectively).

Appendix A PAR-Q

PHYSICAL ACITIVY & MEDICAL HISTORY SCREENING FORM

Physical Activity:

1. Do you participate in weekly aerobic exercise?_________
    a. If yes, how many days per week?_________
    b. How many minutes per exercise session?_________
        i. Total minutes per week?_________
    c. What types of exercises?______________________

Medical History:

	Yes	No
1. History of heart problems, chest pain, or stroke?		
2. Have you ever been diagnosed with MI or Peripheral Vascular Disease?		
3. Increased blood pressure?		
4. Any chronic illness or condition?		
5. Difficulty with physical exercise?		
6. Advice from a physician not to exercise?		
7. Recent surgery? (Last 6 months)		
8. Pregnancy? (Now or within the last 3 months)		
9. History of breathing or lung problems?		
10. Muscle, joint, back disorder, or any previous injury still affecting you?		
11. Diabetes or thyroid conditions?		
12. Cigarette smoking habit?		
13. Increased blood cholesterol?		
14. History of heart problems in your immediate family?		
15. Do you have any implantable devices (i.e. pacemaker, defibrillator)		
16. Hernia or any condition that may be aggravated by lifting weights?		
17. Do you have any condition limiting your movement?		
18. Are you aware of being allergic to any drugs or insect bites?		
19. Do you have asthma?		
20. Do you have epilepsy, convulsions, or seizures of any kind?		
21. Do you follow any specific diet?		

Please explain in detail any “YES” answers:

Has any member of your family had any of those listed above?

Completed by:

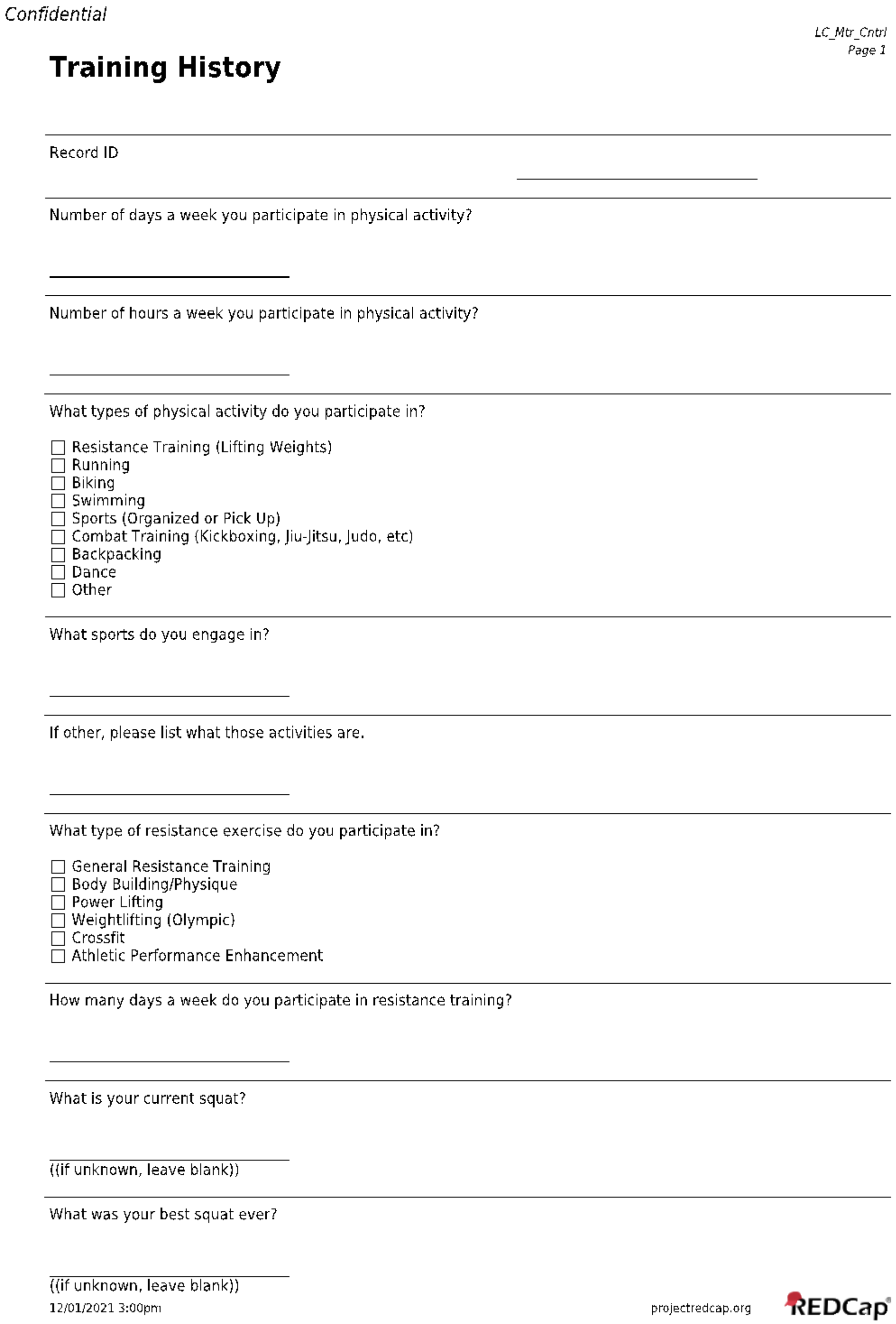
____/_____/_______

Signature of Study ParticipantDate

____

Printed Name

Appendix B Physical Activity History Questionnaires



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Training History

Record ID

____

Number of days a week you participate in physical activity?

____

Number of hours a week you participate in physical activity?

____

What types of physical activity do you participate in?

- ☐ Resistance Training (Lifting Weights)
- ☐ Running
- ☐ Biking
- ☐ Swimming
- ☐ Sports (Organized or Pick Up)
- ☐ Combat Training (Kickboxing, Jiu-Jitsu, Judo, etc)
- ☐ Backpacking
- ☐ Dance
- ☐ Other

What sports do you engage in?

____

If other, please list what those activities are.

____

What type of resistance exercise do you participate in?

- ☐ General Resistance Training
- ☐ Body Building/Physique
- ☐ Power Lifting
- ☐ Weightlifting (Olympic)
- ☐ Crossfit
- ☐ Athletic Performance Enhancement

How many days a week do you participate in resistance training?

____

What is your current squat?

____
((if unknown, leave blank))

What was your best squat ever?

____
((if unknown, leave blank))

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What is your current deadlift?

____
((if unknown, leave blank))

What is your current bench?

____
((if unknown, leave blank))

What is your current mile time?

____
((if unknown, leave blank))

What is your current 5k time?

____
((if unknown, leave blank))

What type of cardio exercise do you participate in?

☐ general/leisurely (comfortable pace)
☐ Mid range (5-10ks)
☐ Long range (half/full marathons, sprint/olympic triathlons)
☐ Ultra (50+ miles, ironman triathlons)
☐ High Intensity Interval Training (HIIT)

How many days a week do you participate in cardio exercise?

____

Comments:

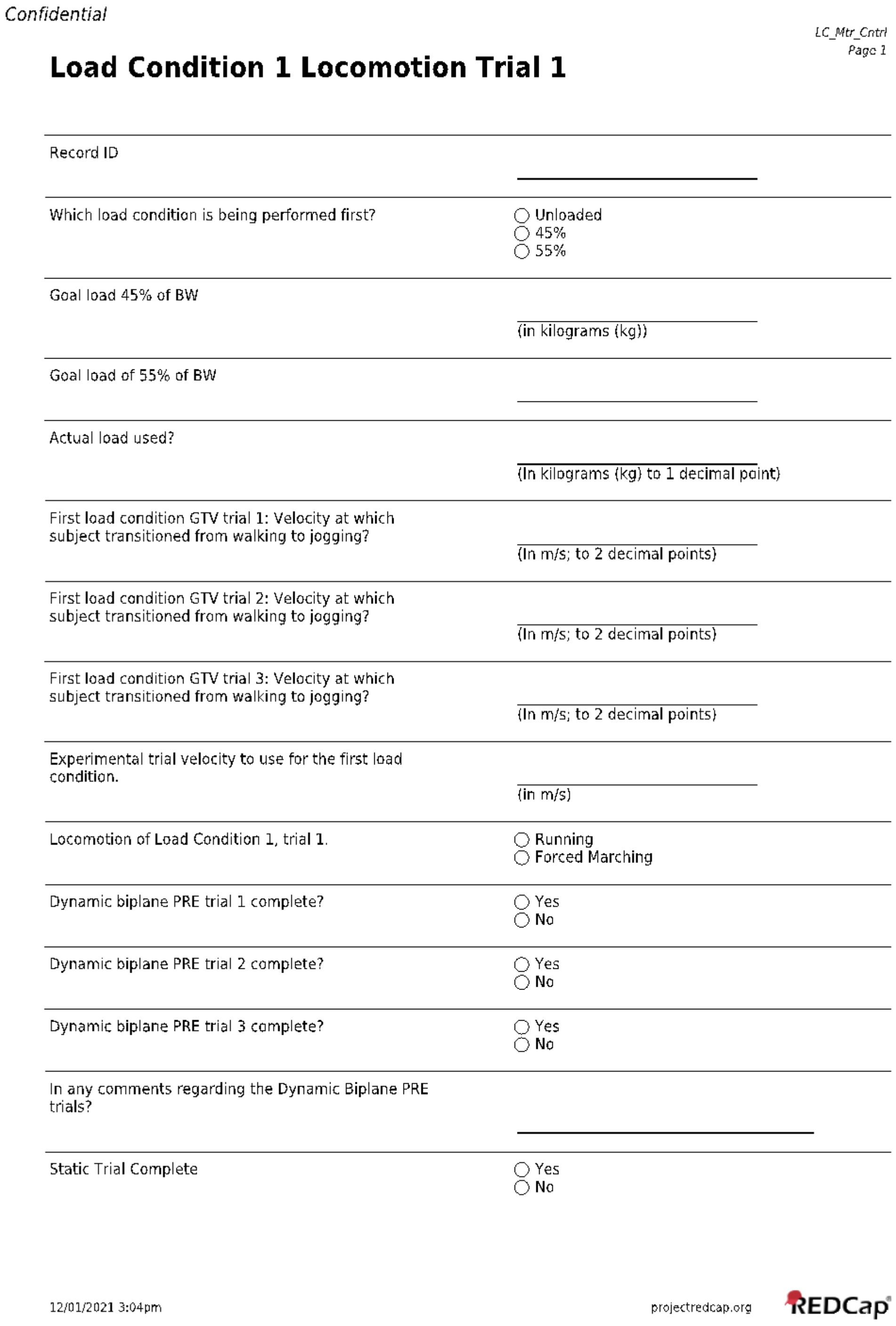
Have you ever executed any type of loaded marching, hiking or running?	○ Yes ○ No (i.e, weight vests, plate carriers, backpacks, rucksacks, sandbags, or carried a heavy object for a prolonged distance)
What type of loaded activities did you participate in? (Check all that apply)	☐ Mountaineering ☐ Loaded runs ☐ Adventure racing (i.e., Spartan races) ☐ Day hikes (single day trip loads) ☐ Backpacking (multi-day loads) ☐ Military setting

How often did you engage in the loaded activity?	________________________________ (i.e., once a week, once a month, once a year or only engaged in that activity a couple times, etc...)
Have you ever served in the military?	○ Yes ○ No
If yes, which branch?	○ Army ○ Airforce ○ Marines ○ Navy
Were you in a ground close combat position (i.e. infantry)	○ Yes ○ No
How many years did you serve?	________________________________ (Report in years)
Have you previously suffered a serious musculoskeletal lower extremity injury? -Required medical treatment (surgery, rehab, etc). -Activity was limited as a result.	○ Yes ○ No
If yes, what lower extremity injuries have you sustained? (please describe all that you have experienced).	________________________________
Have you previously suffered a serious musculoskeletal upper body injury? -Required medical treatment (surgery, rehab, etc). -Activity was limited as a result.	○ Yes ○ No
If yes, what upper body injuries have you sustained? (please describe all that you have experienced).	________________________________

Pre-exercise Survey

Record ID	
Date	
How was the quality of your sleep last night?	Worst Sleep Best Sleep *(Place a mark on the scale above)*
How many hours of sleep did you get last night?	
How many times did you wake up during your sleep last night?	
How many bottles/glasses of water did you drink today?	
Have you eaten in the last 3 hours?	○ Yes ○ No
Did you take any supplements today (Pre-workout, BCAAs, Creatine, etc)	○ Yes ○ No
What did you take?	
How sore are you?	Extremely Sore No Soreness *(Place a mark on the scale above)*
Do you feel like exercising today?	Don't want to train Can't wait to train *(Place a mark on the scale above)*
Today I feel I would physically perform ____________ than usual.	Worse Same Better *(Place a mark on the scale above)*
Comments:	

Appendix C Data Collection Form



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Load Condition 1 Locomotion Trial 1

Record ID	
Which load condition is being performed first?	○ Unloaded ○ 45% ○ 55%
Goal load 45% of BW	(in kilograms (kg))
Goal load of 55% of BW	
Actual load used?	(In kilograms (kg) to 1 decimal point)
First load condition GTV trial 1: Velocity at which subject transitioned from walking to jogging?	(In m/s; to 2 decimal points)
First load condition GTV trial 2: Velocity at which subject transitioned from walking to jogging?	(In m/s; to 2 decimal points)
First load condition GTV trial 3: Velocity at which subject transitioned from walking to jogging?	(In m/s; to 2 decimal points)
Experimental trial velocity to use for the first load condition.	(in m/s)
Locomotion of Load Condition 1, trial 1.	○ Running ○ Forced Marching
Dynamic biplane PRE trial 1 complete?	○ Yes ○ No
Dynamic biplane PRE trial 2 complete?	○ Yes ○ No
Dynamic biplane PRE trial 3 complete?	○ Yes ○ No
In any comments regarding the Dynamic Biplane PRE trials?	
Static Trial Complete	○ Yes ○ No

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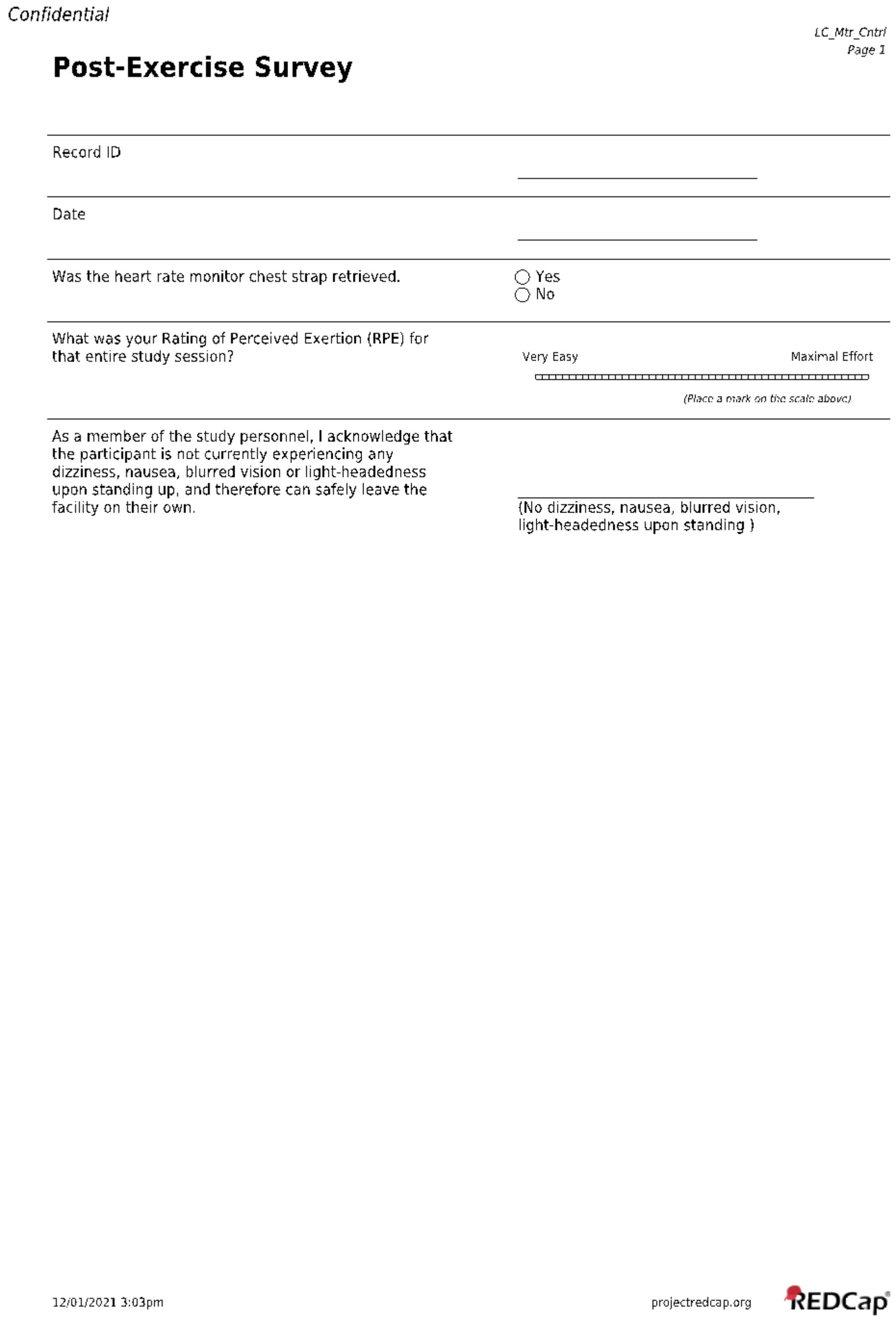
Functional Calibration Trial Complete	○ Yes ○ No
RPE Effort Minute 0 (Scale of 6-20, with 6 representing at rest and 20 being maximal effort)	______________________________
Heart Rate (HR) at minute 0	______________________________ (in beats per minute)
RPE Effort Minute 2 (Scale of 6-20, with 6 representing at rest and 20 being maximal effort)	______________________________
Heart Rate (HR) at minute 2	______________________________ (in beats per minute)
RPE Effort Minute 4 (Scale of 6-20, with 6 representing at rest and 20 being maximal effort)	______________________________
Heart Rate (HR) at minute 4	______________________________ (in beats per minute)
RPE Effort Minute 6 (Scale of 6-20, with 6 representing at rest and 20 being maximal effort)	______________________________
Heart Rate (HR) at minute 6	______________________________ (in beats per minute)
RPE Effort Minute 8 (Scale of 6-20, with 6 representing at rest and 20 being maximal effort)	______________________________
Heart Rate (HR) at minute 8	______________________________ (in beats per minute)
RPE Effort Minute 10 (Scale of 6-20, with 6 representing at rest and 20 being maximal effort)	______________________________
Heart Rate (HR) at minute 10	______________________________ (in beats per minute)
Did the subject complete all 10 minutes of the locomotion trial?	○ Yes ○ No
If no, how much time did they complete?	______________________________

Overall RPE rating for that trial? (6-20 scale)	____________________
Any comments regarding the execution of this trial? (i.e., did a marker fall off, did the subject keep shifting a certain way, did they complain of pain or discomfort, etc.)	________________________
Dynamic biplane POST trial 1 complete?	○ Yes ○ No
Dynamic biplane POST trial 2 complete?	○ Yes ○ No
Dynamic biplane POST trial 3 complete?	○ Yes ○ No
In any comments regarding the Dynamic Biplane POST trials?	________________________

Appendix D Post-Testing Questionnaire



Confidential

LC_Mtr_Cntrl
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Post-Exercise Survey

Record ID	__________________________
Date	__________________________
Was the heart rate monitor chest strap retrieved.	○ Yes ○ No
What was your Rating of Perceived Exertion (RPE) for that entire study session?	Very Easy — Maximal Effort *(Place a mark on the scale above)*
As a member of the study personnel, I acknowledge that the participant is not currently experiencing any dizziness, nausea, blurred vision or light-headedness upon standing up, and therefore can safely leave the facility on their own.	__________________________ (No dizziness, nausea, blurred vision, light-headedness upon standing)

12/01/2021 3:03pm

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