PHD

Physical Rehabilitation of Military Amputees

Ladlow, Peter

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PHYSICAL REHABILITATION OF MILITARY AMPUTEES

PETER LADLOW

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ABSTRACT

The numbers of UK military personnel severely injured during combat was most recently at its highest during and after the conflicts in Iraq and Afghanistan. These conflicts created a relatively large population of young servicemen and women with exceptional long-term healthcare and rehabilitation needs not seen or treated in the UK or globally before. Despite having a combat casualty care pathway being described as 'exemplary' by the Care Quality Commission, the UK Defence Medical Services (DMS) are yet to provide evidence detailing the impact of their care pathway on the health and well-being of their severely injured patients with traumatic lower-limb amputation(s) (LLA). The first aim of this thesis (chapter 4) was to perform a retrospective analysis of functional and mental health outcomes in military personnel with traumatic LLA measured at their last in-patient admission to Defence Medical Rehabilitation Centre (DMRC) Headley Court. This found clinical outcomes indicative of full-preparedness for community integration. Previous research has indicated that individuals with traumatic LLA are at increased risk of a physical inactivity and a range of secondary health conditions, including obesity and compromised cardiometabolic health. Strategies to mitigate this risk have become a priority for DMS. The primary research question of this thesis (chapter 6 and 7) was to determine within and between group differences in physical activity (PA), physical function, body composition and components of cardiometabolic health in military personnel with unilateral and bilateral LLA. A 20 week longitudinal study was performed, consisting of two 4 week in-patient admissions of rehabilitation and two 6 week active recovery blocks at home. These measures were then compared against an age-matched normative control group employed within active roles within the UK Ministry of Defence. Before measuring PA during rehabilitation and at home it was important to first ensure the methods used were accurate and valid for the population of interest. Wearing an Actigraph GT3X+ triaxial accelerometer on the hip of the shortest residual limb and using a predictive model incorporating heart rate offers the most accurate prediction of PA energy expenditure (PAEE) in those with traumatic LLA (85%, 87% and 83% of the variance in PAEE for the unilateral, bilateral and control group, respectively). Chapter 6 and 7 found military personnel with unilateral LLA have a similar capacity for PA, and demonstrated comparable body composition and components of cardiometabolic health as active normative controls. Despite demonstrating levels of function and psychosocial health indicative of full-preparedness back into society, estimated daily PAEE in bilateral LLA was significantly reduced during habitual living in their home environments. The bilateral LLA group demonstrate unfavourable body composition (particularly the distribution of adipose tissue around the abdomen). This was also accompanied by an increased risk of cardiovascular disease and compromised metabolic health, with 63% being classified with metabolic syndrome. To support and manage the long-term health and well-being of military personnel with bilateral LLA, future research should aim to investigate strategies that promote regular engagement in PAEE and/or structured exercise whilst at their home environment.

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LIST OF PUBLICATIONS

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Ladlow, P., Nightingale, T.E., McGuigan, M.P., Bennett, A.N. Phillip, R., Bilzon, J.L.J. (2019) Predicting ambulatory energy expenditure in lower limb amputees using multi-sensor methods. *PLoS ONE* 14(1): e0209249

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Ladlow, P., Nightingale, T.E., McGuigan, M.P., Bennett, A.N. Phillip, R., Bilzon, J.L.J. Impact of anatomical placement of an accelerometer on prediction of physical activity energy expenditure in lower-limb amputees. *International Conference on Ambulatory Monitoring of Physical Activity and Movement (ICAMPAM)* (2015) Limerick, Ireland

Ladlow, P., Nightingale, T.E., McGuigan, M.P., Bennett, A.N. Phillip, R., Bilzon, J.L.J. Predicting ambulatory energy expenditure in lower limb amputees using multi-sensor Methods. At: *American College of Sport Medicine Annual Meeting*, (2015) Boston, USA

Ladlow P. Energy expenditure and clinical outcomes in trauma rehabilitation. At: *Defence Medical Services: Medical Innovation Conference: Prolonged Care* (2018), Birmingham, UK.

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LIST OF ABBREVIATIONS

ADL	Activities of Daily Living
AHR	Actiheart
AMPQ	Amputee Mobility Predictor Questionnaire
AMP-Pro	Amputee Mobility Predictor with Prosthetics
ANOVA	Analysis of Variance
BMI	Body Mass Index
BP	Blood Pressure
CO_2	Carbon Dioxide
CRP	C-Reactive Protein
DEXA	Dual- Energy X-ray Absorptiometry
DMRC	Defence Medical Rehabilitation Centre
DMS	Defence Medical Services
ECG	Electrocardiogram
EE	Energy Expenditure
ERI	Exercise Rehabilitation Instructor
GAD	Genral Anxiety Disorder
GSW	Gunshot Wound
HDL	High Density Lipoprotein
HOMA-IR	Homeostatic Model Assessment – Insulin Resistance
HOMA-β	Homeostatic Model Assessment – β Cell Function
HR	Heart Rate
iAUC	Incremental Area Under Curve
IED	Improvised Explosive Device
IDT	Interdisciplinary Team
ISI	Insulin Sensitivity Index
ISS	Injury Severity Score
LDL	Low Density Lipoprotein
LLA	Lower Limb Amputation
LoA	Limits of Agreement
MAE	Mean Absolute Error
MET	Metabolic Equivalent
MFCL	Medicare Functional Classification System
MOD	Ministry of Defence
MWD	Minute Walk Distance

MWT	Minute Walk Test
NCO	Non-Commissioned Officer
NEFA	Non-Esterified Fatty Acids
NHS	National Health Service
NISS	New Injury Severity Score
OGTT	Oral Glucose Tolerance Test
PA	Physical Activity
PAC	Physical Activity Count
PAEE	Physical Activity Energy Expenditure
PHQ	Patient Health Questionnaire
PTSD	Post-Traumatic Stress Disorder
QoL	Quality of Life
RER	Respiratory Exchange Ratio
RM	Repetition Maximum
RMR	Resting Metabolic Rate
RPE	Rate of Perceived Exertion
RTA	Road Traffic Accident
SD	Standard Deviation
SEE	Standard Error of the Estimate
SIGAM	Special Interest Group in Amputee Medicine
TC:HDL	Total Cholesterol : High Density Lipoprotein Ratio
VAT	Visceral Adipose Tissue
$\dot{V}CO_2$	Carbon Dioxide Production
$\dot{V}O^2$	Oxygen Consumption

CHAPTER 1. INTRODUCTION

More than two thousand years ago Plato wrote:

> *"Lack of activity destroys the good condition of every human being, while movement and methodical physical exercise save it and preserve it."*

Despite Plato writing this sentence back in 350 BC, the modern notion of exercise as a medical treatment is thought to have originated with R Tait McKenzie (Berryman, 1995). Working as a medic during the First World War; McKenzie perceived exercise as a technique to rehabilitate people with disabling injuries. McKenzie noted that some wounded soldiers can return to the battlefield, and those wounded soldiers who were permanently disabled required physical rehabilitation to help cope with their disabilities (Moore, 2004). UK military physicians have been delivering intensive rehabilitation to their injured military personnel ever since and this approach still exists today across UK Defence Medical Services (DMS).

The numbers of UK military personnel severely injured during combat was recently at its highest during and after the conflicts in Iraq and Afghanistan. Between 2003 and 2012 the number of UK military servicemen wounded in action was 2,184 (Penn-Barwell et al, 2015). In Afghanistan, 265 servicemen sustained 416 amputations (Edwards et al, 2015). Although deeply traumatic to each casualty, their family and serving colleagues, each military conflict often brings great advances in battle care emergency medicine that enables military personnel to survive injuries that would have once proved fatal (Gulland, 2008). The conflicts in Iraq and Afghanistan proved no different. Subsequently, this created a relatively large population of young servicemen and women with exceptional long-term healthcare and rehabilitation needs not seen or treated in the UK or globally since the previous two World Wars.

In the general population, physical inactivity is now considered a larger contributor to all-cause mortality than obesity (Ekelund et al, 2015). Reduced physical activity (PA) has been implicated in impaired metabolic function (Thyfault & Krogh-Madsen, 2011), with as little as three days of reduced PA demonstrating a negative effect on the body's ability to regulate plasma glucose concentrations (Mikus et al, 2012). This is a highly significant finding considering the amount of time severely injured military personnel are confined to a bed or wheelchair during the early phases of rehabilitation post-injury. Strategies to increase physical function, whilst reducing the development of secondary health disorders in their disabled military personnel became a very important issue for DMS. One of the primary concerns relates to the premise that disability is known to negatively impact PA behaviour, as described by the Conceptual Model of Disability-Associated Low Energy Expenditure Deconditioning Syndrome (DALEEDS) (Rimmer et al, 2012); (figure 1.1). Civilians with lower limb amputation (LLA) have demonstrated decreased PA levels compared to healthy adults (Bussmann et al, 2004; Bussmann et al, 2008; Halsne et al, 2013; Tudor-Locke et al, 2011) and the majority of individuals with amputations are found to live sedentary lifestyles (Pepin et al, 2018).

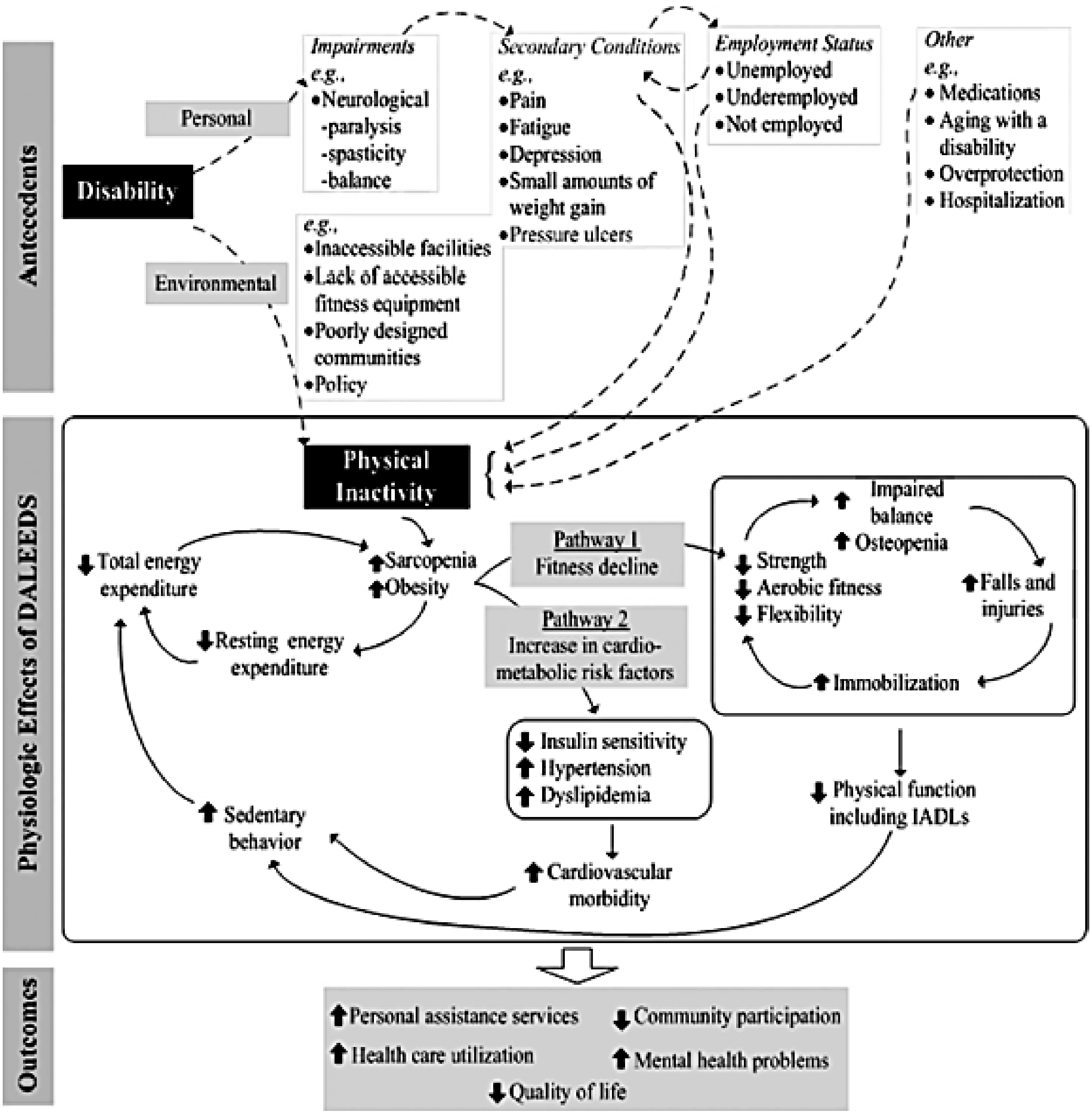


Figure 1.1: Conceptual Model of Disability-Associated Low Energy Expenditure Deconditioning Syndrome (DALEEDS). Taken from, Rimmer et al. (2012).

In Rimmer's DALEEDS model (figure 1.1), both personal and environmental factors may lead to increases or decreases in PA participation in people with disabilities. Physical inactivity leads to a decline in skeletal muscle strength and muscle mass and an increase in adipose tissue (i.e. obesity) leading to a dysregulated metabolism. Rimmer proposes two pathways that lead to these unfavourable physiological changes: pathway 1, fitness decline; and pathway 2, increase in cardiometabolic risk factors. In Pathway 1, the cumulative effect of deconditioning leads to reduced physical function expressed as a lower ability to complete various activities of daily living (ADL: e.g., transfers, pushing a wheelchair, walking with a mobility aid) and a further increase in sedentary behaviour (e.g., sitting time). In pathway 2, muscle loss and obesity increase cardiometabolic risk factors such as reduced insulin sensitivity, hypertension, dyslipidemia, and increased inflammatory biomarkers (Rimmer et al, 2012). Both of these pathways and their association with individuals following traumatic LLA are explored in greater detail in the next chapter. However, in both pathways 1 and 2, the end result is reduced total energy expenditure, which creates a continuous cycle of health risk leading to metabolic and physiological disturbances that cause further advancements in physical decline over time. This physical decline may lead to greater reliance on family members or personal assistance services to assist with ADL; treatment for cardiovascular morbidity, injury from falling, or secondary health conditions increasing health care cost; mental health problems including anxiety, depression and social isolation may also increase (Rimmer et al, 2012). The cumulative effect of these outcomes is likely to reduce overall quality of life (QoL). Therefore, the promotion of PA and participation in structured exercise is considered to be a vital component in the rehabilitation and reintegration of individuals with LLA back into society (Pepper & Willick, 2009).

To date, the DMS have not published data on rehabilitation outcomes of their military personnel with traumatic LLA. Nor has it investigated any of the known secondary health risks associated with LLA or how these may compare with a physically active peer group. UK military personnel with traumatic LLA are often young men who prior to their injuries were likely to be very active with many determined to demonstrate levels of physical function that allow them to identify with their pre-injured selves. They are facilitated to achieve these goals with support from clinicians delivering the complex trauma rehabilitation care pathway that provides unique intensive interdisciplinary (IDT) rehabilitation alongside the most advanced prosthetic provision available in the UK (see chapter 3 for a description of DMS combat casualty care pathway). However, the effectiveness of this approach at reducing the risk factors for secondary health problems has not been evaluated.

The UK's Healthcare Commission (2009) and Care Quality Commission (2012) have described the military trauma care provided at the UK Defence Medical Rehabilitation Centre (DMRC) and across the DMS as 'exemplary'. The DMS model of rehabilitation is highly regarded by these commissions, however little is known or documented about the process of rehabilitation or its evaluation. Despite 'exemplary' care, anecdotal evidence suggests some shortcomings still remain in this model of rehabilitative care. For example, unfavourable changes in body composition (abdominal fat) in many UK military personnel with LLA have been observed by clinical staff and it is unknown what effect this may have on short to long-term health outcomes. Within the UK military, and the National Health Service (NHS), there is a medical and economic interest in ensuring that the chosen complex trauma rehabilitation pathway is the most effective and efficiently administered one available to ensure the best possible functional, medical and psychosocial health outcomes. An evaluation of this care pathway would be a useful step in this process.

The overall aim of this thesis is to evaluate the effect of the UK DMS Complex Trauma Rehabilitation Care Pathway on the health and well-being of serving UK military personnel with traumatic unilateral and bilateral LLA at DMRC Headley Court. The thesis will begin with a literature review exploring the impact of traumatic LLA on secondary health conditions, physical function and body composition. The initial research question is to determine the functional and mental health outcomes of individuals with LLA at the end of the complex trauma rehabilitation pathway and comment on whether these outcomes are compatible with independent living or community integration. The impact of structured exercise rehabilitation at DMRC and free-living PA at home on the health and wellbeing of individuals with unilateral and bilateral LLA against age-matched uninjured controls will then be investigated. The necessary starting point for this assessment is to develop population specific models capable of accurately estimating physical activity energy expenditure (PAEE). These methods, incorporating validated wearable technology, will then be utilised in a longitudinal observational study to ascertain the impact of intensive in-patient rehabilitation and recovery at home on functional health, body composition and cardiometabolic health outcomes in individuals with traumatic LLA (see figure 1.2).

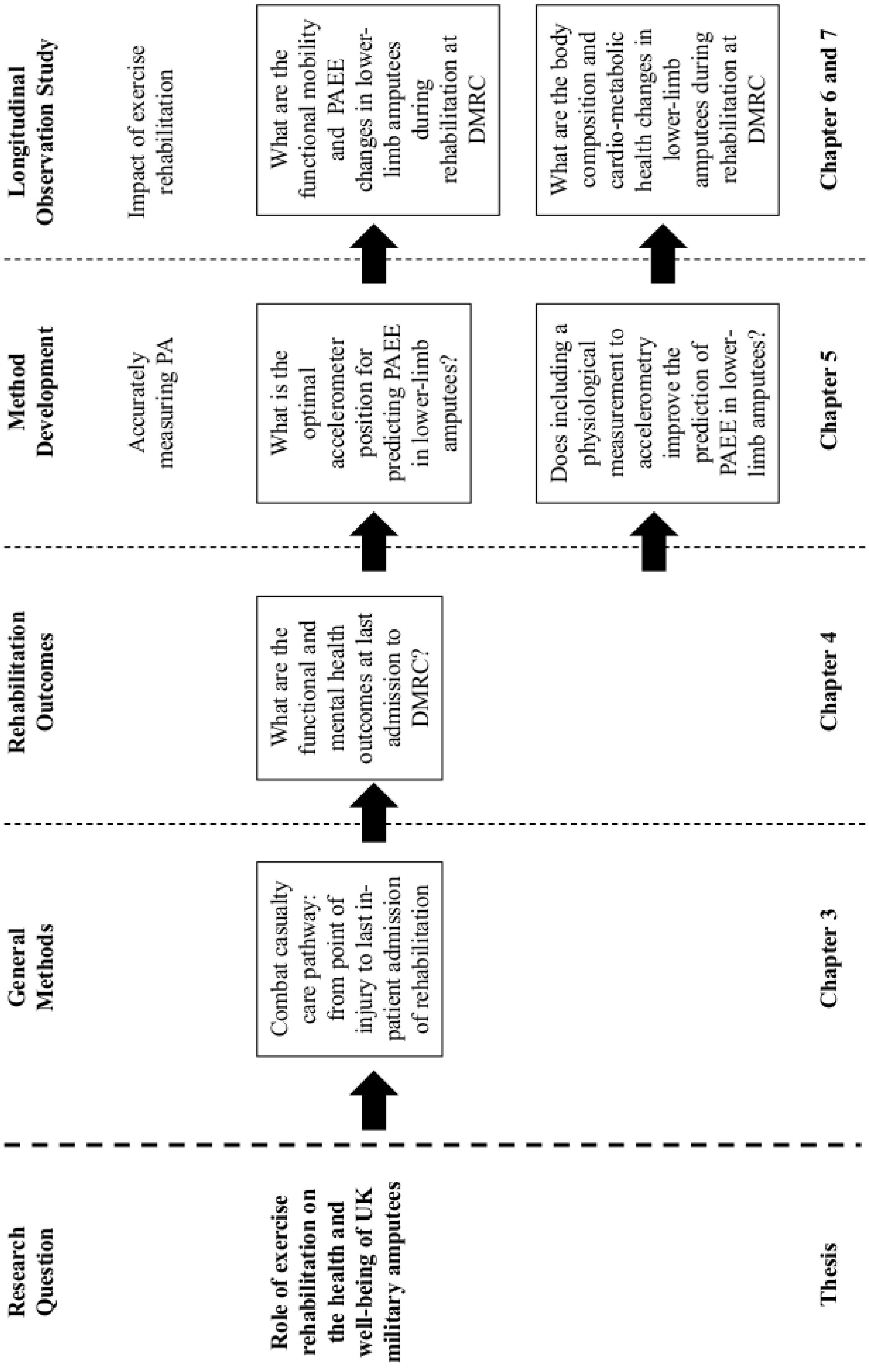


Figure 1.2: Schematic of the research questions and layout of thesis.

CHAPTER 2. REVIEW OF THE LITERATURE

2.1. DEFINING LOWER-LIMB AMPUTATION

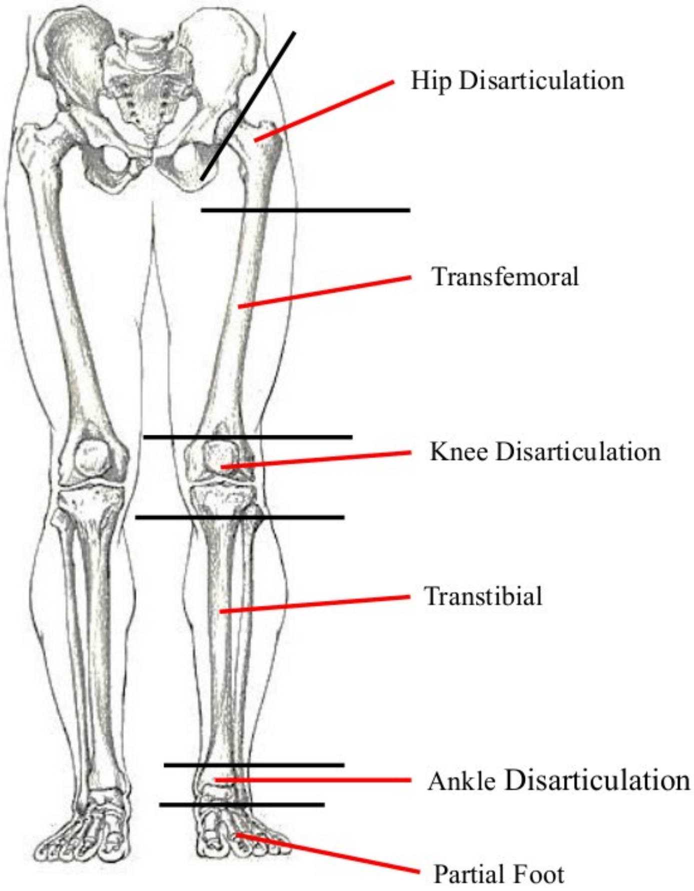


Figure 2.1: Lower-limb amputation level nomenclature

Amputation is the removal of part or the whole of a limb. When defining LLA it is important to remain mindful that the level of amputation may have considerable influence on future levels of functional capacity and health (discussed in greater detail in section 2.4 of this chapter). The terms ‘transfemoral’, ‘knee disarticulation’, and ‘transtibial’ amputation are used interchangeably in the available literature with ‘above knee’, ‘through knee’ and ‘below knee’ amputation, respectively.

2.2. EPIDEMIOLOGY OF TRAUMATIC AMPUTATION

2.2.1. Incidence and Prevalence

In the year 2005, 1.6 million people (or one in 190) were living in the United States with a loss to an extremity, mainly due to vascular aetiology. It is projected that the number of Americans living with an amputation will increase 2-fold by the year 2050 to 3.6 million (Ziegler-Graham et al, 2008). In the United States, an average of 133,235 amputation-related hospital discharges occur each year, with the vast majority (82%) linked to vascular-related conditions (Adams et al, 1999). In the UK, an estimated 5,500 people require a LLA each year, and of these, 53% are transtibial amputation and 39% transfemoral amputation (NHS, 2009). Similar to the US, three quarters of all referrals for LLA are due to dysvascularity, 8% due to infection and 7% of amputations are due to trauma (NHS, 2009).

Traumatic amputation has long been a devastating consequence of war. The prevalent mechanism of injury leading to combat amputations is powerful explosive weapons (Melcer et al, 2010) that create extensive tissue trauma and disruption of vascular and neurological networks (Keklikci et al, 2010). As a consequence of such high forces, velocities and temperatures at the time of injury most combat injured personnel have also sustained other associated injuries in addition to amputation(s) (Smurr et al, 2008). At the end of World War I there were at least 29,400 British servicemen with LLA and 11,600 with limb loss to the upper extremity. By the end of 1938 there were only 3,400 surviving amputees in receipt of artificial limbs in the UK (Ministry of Pensions, 1939). At the end of World War II the UK military had an additional 9,000 amputees (Stewart, 2008). During the most recent conflicts in Iraq and Afghanistan (Operation Iraqi Freedom and Operation Enduring Freedom), 52,022 US Military servicemen were wounded in action with 1,645 requiring major limb amputation(s) (Fischer, 2013). Between 2003 and 2012 the number of UK Military servicemen wounded in action during these conflicts were 2,184 (Penn-Barwell et al, 2015), in Afghanistan, 265 individuals sustained 416 amputations (Edwards et al, 2015).

Webster et al. (2018) determined the mortality rates for each traumatic LLA level and associated injuries sustained in both Iraq and Afghanistan between January 2003 and the end of UK operations in August 2014. Of the 977 casualties, there were 298 fatalities (30.5%) and 679 (69.5%) survivors (Webster et al, 2018). Injury characteristics associated with an increased mortality rate include a more proximal amputation level, pelvic fracture, and abdominal injury. Those sustaining bilateral transfemoral amputation had a mortality rate of nearly 4.5 times those with a unilateral transtibial amputation and those sustaining a traumatic LLA with a pelvic fracture had a mortality rate nearly three times greater (60.8%) than those without a pelvic fracture (22.9%) (Webster et al, 2018). Although deeply traumatic to each casualty, their family and serving colleagues, each military conflict often brings great medical advances in battle care emergency medicine that enables military personnel to survive injuries that would have once proved fatal (Gulland, 2008). UK military personnel are now surviving injuries during conflict they would not previously survived; therefore we need to consider the risk associated with chronic disease in this unique population.

2.2.2. Chronic Disease Related Mortality

All-cause mortality as well as the risk of mortality due to cardiovascular disease in patients with traumatic amputation has been more widely studied in military populations. The largest study investigating the impact of traumatic amputation on risk of mortality was a 30 year follow-up study investigating US male soldiers wounded during World War II. Hrubec and Ryder (1979) investigated 3,887 military personnel with proximal limb amputees (transfemoral or transhumeral – above elbow), 3,890 injured with disfigurement (but without amputation), and 2,917 were distal limb amputees (loss of limb to part of the hand or foot). After 15 years, the relative risk of cardiovascular disease related mortality was 1.58 and 3.5 times greater in individuals with unilateral trans-femoral amputation and bilateral trans-femoral amputation respectively when compared to veterans who had limb salvage procedures (Hrubec & Ryder, 1979).

Subsequent studies have also demonstrated that traumatic trans-femoral amputations are associated with an increased cardiovascular morbidity or mortality in the long-term. Modan et al. (1998) compared the mortality rates of male individuals with traumatic unilateral LLA (n = 201) from the Israeli army against a cohort sample representing the general population (n = 1,832) and found that mortality rates were significantly higher (21.9% vs 12.1%) in military personnel with amputation compared to uninjured controls. Mortality due to cardiovascular disease was 2-fold greater in individuals with LLA compared to controls ($p < 0.001$) (Modan et al, 1998). Although Modan et al. (1998) did not perform additional analysis to address the specific causes of mortality within cardiovascular disease; they did find a trend towards an increase in the risk of myocardial infarction among the amputation group. Whilst these results are informative, its relevance to today’s lower-limb amputee could be questioned. Assistive technologies, such as prosthesis design and technology, have advanced considerably since these studies were published allowing greater scope for improvements in physical function and increases in PA. Therefore, the associated risk of mortality for the modern day lower-limb amputee may have changed since these older studies were published in the 70’s and 90’s. However, many secondary health conditions associated with traumatic LLA remain and these conditions must be considered and appropriately managed to optimise the long-term health of this population.

2.3. PREVALENCE OF SECONDARY HEALTH CONDITIONS ASSOCIATED WITH TRAUMATIC LOWER-LIMB AMPUTATION

Whilst mortality is an important factor and metric, it is arguably more important to know and understand the prevalence of those living with chronic diseases due to the known financial and well-being implications. In this section, the following chronic diseases are discussed; cardiovascular disease, diabetes mellitus, obesity, osteoporosis, low back pain, chronic pain and mental health disorders.

2.3.1. Cardiovascular Disease

Using self-report questionnaires and interview techniques to determine the long-term QoL and health of 247 Vietnam veterans with combat related limb-loss, Foote et al. (2015) reported cardiovascular disease in 18.2% of the cohort. Shahriar et al. (2009) performed a 22 year cross-sectional study using 327 Iranian males with war-related traumatic bilateral LLA. The prevalence of coronary artery disease was similar to the general Iranian population; however, 95% presented with at least one modifiable cardiovascular disease risk factor, which is significantly greater than population norms. The prevalence of risk factors included: hyperglycemia (13.1%), systolic hypertension (18.9%), diastolic hypertension (25.6%), abdominal obesity (82.5%), high total cholesterol (36.7%), reduced high density lipoprotein (HDL) cholesterol (25.9%), elevated low density lipoprotein (LDL) cholesterol (24.7%), high triglycerides (32.1%), and smoking (31.8%) (Shahriar et al, 2009). The authors conclude that the majority of individuals with trauma related bilateral LLA are at an increased risk of cardiovascular disease in the near future.

Stewart et al. (2015) performed a retrospective analysis of 3,846 US injured military personnel wounded between 2002 and 2011 during conflicts in Iraq and Afghanistan. Using International Classification of Diseases - 9th edition administrative codes, the authors aim was to determine the impact of injury severity on subsequent development of four conditions; hypertension, coronary artery disease, diabetes mellitus and chronic kidney disease. Each 5-point increment in injury severity score (ISS) was associated with a 6%, 13%, 13%, and 15% increase in developing hypertension, coronary artery disease, diabetes mellitus, and chronic kidney disease, respectively (Stewart et al, 2015). The estimated incidence rates of hypertension and coronary artery diseases for the most severely injured patients (ISS > 25) were 2.5 to 4-fold higher than published rates for the overall US military population, respectively (Crum-Cianflone et al, 2014; Granado et al, 2009). It is quite compelling that these outcomes were observed after a relatively short period of 1 to 3 years post-injury. This may have profound implications for both the individual wounded during combat and for healthcare departments responsible for funding long-term care of US military veterans.

2.3.2. Diabetes Mellitus

Hyperinsulinemia has been reported in older individuals with traumatic unilateral and bilateral LLA (Peles et al, 1995; Rose et al, 1987). Using self-reported questionnaires and interview techniques, Foote et al. (2015) reported diabetes mellitus in 23% of Vietnam veterans (n = 247) approximately 40 years following injury. In the same retrospective analysis mentioned earlier, Stewart et al. (2015) estimated incidence of developing diabetes mellitus for the most severely injured patients (ISS > 25) to be 2.6 fold higher than published rates for the overall US military population (Boyko et al, 2010; Stewart et al, 2015).

2.3.3. Obesity

Individuals who undergo LLA are at increased risk for weight gain and obesity as a result of positive energy balance and comorbid conditions (Bouldin et al, 2016; Eckard et al, 2015; Gailey et al, 2008; Kurdibaylo, 1996; Shahriar et al, 2009). Cross-sectional studies show that obesity is more common in those with an amputation than those without an amputation (Kurdibaylo, 1996; Rose et al, 1987). In 327 Iranian men with combat-related traumatic bilateral LLA, the most commonly reported modifiable cardiovascular disease risk factor at 22 years follow-up was abdominal obesity (82.5%) (Shahriar et al, 2009). Anthropometric markers taken from adults during the first year after amputation demonstrate an increase in body fat mass directly related to the level of amputation (Kurdibaylo, 1996). The largest increase in obesity progression was reported in individuals with bilateral transfemoral or transfemoral/transtibial amputation (64%), with both groups averaging 25.9% body fat. The frequency of obesity in individuals with unilateral transtibial amputation and transfemoral amputation was 38% and 48%, respectively. Unfortunately, this study did not recruit individuals without amputation(s) to act as a control group comparison.

2.3.4. Osteoarthritis

Post-traumatic degenerative arthritis from injuries to joints on non-amputated limbs takes years to develop and is more common in individuals with traumatic LLA and contralateral limb involvement than in individuals without amputation (Reiber et al, 2010). Most studies that have assessed the future development of arthritis following amputation are in older military veterans and they reveal a prevalence of 54% to 71% (Dougherty et al, 2014; Foote et al, 2015; Gailey et al, 2010; Kulkarni et al, 1998; Reiber et al, 2010). Kulkarni et al. (1998) found a 3-fold increased risk for osteoarthritis in individuals with transfemoral amputation compared with transtibial amputation. Reiber et al. (2010) compared individuals with unilateral LLA from Vietnam (n = 178) to those injured during the recent conflicts in Iraq and Afghanistan (n = 172). Arthritis in the contralateral limb was reported in 72% and 30% of Vietnam and Iraq/Afghanistan servicemen, respectively. This finding by Reiber et al. (2010) is clinically significant as the mean time since amputation in those participants from Iraq/Afghanistan was 3 ± 1 years. Therefore, traumatic limb loss may promote early onset of osteoarthritis. Early amputation (< 90 days post-injury) is also reported to have a lower prevalence for osteoarthritis (10%) compared to 21% of late amputation (≥ 90 days post-injury) (Melcer et al, 2017).

Individuals with traumatic LLA are more than twice as likely to develop knee pain in their intact limb as uninjured adults (Norvell et al, 2005); the risk increases the more proximal the level of amputation (Ebrahimzadeh & Fattahi, 2009; Ebrahimzadeh & Hariri, 2009; Norvell et al, 2005). The prevalence of contralateral knee joint pain in elderly veterans (mean 63 ± 12 years) with traumatic unilateral transfemoral and unilateral transtibial LLA was 50% and 36%, respectively, compared to 20% in aged matched controls (Norvell et al, 2005). These authors believe the high prevalence of contralateral joint pain arises from a combination of gait abnormalities and increased physiological loads on intact joints.

2.3.5. Osteopenia / Osteoporosis

Due to the limited ability to weight bear through the lower-limb(s) and participate in lower-limb based exercise for weeks or months prior to prosthetic training, individuals with traumatic LLA are likely to be predisposed to osteopenia and osteoporosis (Kulkarni et al, 1998; Melcer et al, 2017; Rush et al, 1994; Smeltzer et al, 2005; Tanaka et al, 2001). Rush et al. (1994) and Kulkarni et al. (1998) found a greater incidence of femoral neck osteopenia on the residual limb among individuals with unilateral transfemoral amputation. More recently Melcer et al. (2017) reported a greater prevalence of osteoporosis following one year of limb-loss in individuals with early amputation (< 90 days post-injury) compared to late amputation (> 90days: 16% and 8%, respectively) (Melcer et al, 2017). Melcer et al. (2017) did however note a substantial decline in the prevalence of osteoporosis after the first year suggesting the condition improves after initial prosthetic fitting, weight-bearing activities, and daily ambulation. A reduction in bone mineral density, along with the increased likelihood of falling during prosthetic training, may increase the risk for bone fractures (Kulkarni et al, 1998; Rush et al, 1994).

2.3.6. Low Back Pain

For veterans with LLA back pain is associated with lower scores on the subscales for bodily pain, vitality, social functioning and mental health (Rahimi et al, 2012). Furthermore, back pain is associated with poor physical health-related QoL (Taghipour et al, 2009). Back pain occurs in 52% to 81% of individuals with traumatic LLA (Ebrahimzadeh & Fattahi, 2009; Ebrahimzadeh & Hariri, 2009; Ehde et al, 2001; Ephraim et al, 2005; Smith et al, 1999), considerably higher than the 15% to 25% reported in the general population (Andersson, 1998). When comparing individuals who sustained major traumatic LLA during the Vietnam War and Iraq/Afghanistan conflicts, the prevalence of reported chronic back pain was 36% and 42%, respectively (Reiber et al, 2010). Back pain may be more common following transfemoral than transtibial amputation (Ebrahimzadeh & Fattahi, 2009; Ebrahimzadeh & Hariri, 2009; Smith et al, 1999). The increased susceptibility is thought to be the result of the myofascial changes following amputation and the altered gait pattern developed to accommodate the prosthesis (Kulkarni et al, 2005; Smith et al, 1999).

2.3.7. Chronic Pain (phantom and residual limb pain)

Chronic pain is highly prevalent, and a significant cause of disability following trauma-related LLA (Castillo et al, 2006; Ephraim et al, 2005; Hagberg & Branemark, 2001). Furthermore, pain may worsen the functional, vocational and psychiatric outcomes of individuals with amputation (Castillo et al, 2006). The prevalence of phantom limb pain, a form of neuropathic pain, is typically reported in 50% to 80% of individuals with traumatic LLA (Ebrahimzadeh & Rajabi, 2007; Ehde et al, 2000; Ephraim et al, 2005; Flor, 2002; Foote et al, 2015; Ketz, 2008; Reiber et al, 2010). For veterans with amputation, the duration of phantom pain is associated with worse physical functioning, bodily pain, mental health and a lower score on the physical component scale (Rahimi et al, 2012). The development of phantom limb pain is associated with the magnitude of pre-amputation and residual limb pain experienced, the duration of limb pain before amputation, and psychological factors such as emotional stress and anxiety (Perkins et al, 2012). When comparing phantom limb pain reported among veterans from Vietnam and those from Iraq and Afghanistan, the prevalence appears comparable between groups (72% and 76%, respectively) (Reiber et al, 2010).

There is almost a two-fold increase in the reporting of residual limb pain (remaining part of the amputated limb) following traumatic LLA compared to amputations resulting from non-traumatic pathologies (Ephraim et al, 2005). Whilst common immediately after amputation, residual limb pain was traditionally believed to resolve with surgical healing (Perkins et al, 2012). However, Reiber et al. (2010) reported an elevated number of servicemen/veterans who sustained major traumatic LLA during Iraq and Afghanistan report residual limb pain (63%) 3 years post injury compared to Vietnam veterans (48%) who have lived with their injury for 39 years (Reiber et al, 2010). It important to remain mindful that the severity of injury between these groups are unlikely to be comparable and as those injured in Iraq/Afghanistan who have only lived with their injury for 3 years are younger and more likely to pursue an active lifestyle thereby increasing the loading demands on their residual limb(s) potentially elevating the prevalence of residual limb pain in this group. Edhe et al. (2000) recorded residual limb pain intensity scores in individuals with amputation as 5.4 ± 2.7 (scored on a scale of 0 to 10), with 38% of participants rating their residual limb pain intensity as severe (Ehde et al, 2000).

2.3.8. Mental Health (Depression, Anxiety and PTSD)

An intense emotional response is common following traumatic amputation and forms part of the psychological adjustment. Psychosocial reactions to traumatic LLA begin with shock, denial, anxiety, distress, depression, acute grief, acknowledgement, hostility and frustration.; combined with a willingness to participate in rehabilitation activities, early acceptance, reorganization and reframing (Bradway et al, 1984). Therefore, psychological disorders can be common in individuals with traumatic LLA (particularly during the early stages). Difficulties associated with mental health, such as depression and post-traumatic stress disorder (PTSD) are described as among the biggest reasons for poor QoL among older veterans with amputation (Caddick et al, 2019; Ebrahimzadeh & Fattahi, 2009; Epstein et al, 2010; Foote et al, 2015; Howard et al, 2018).

PTSD is an anxiety disorder unique to trauma exposure and is characterized by symptoms of avoidance, re-experiencing and hyperarousal (Perkins et al, 2012). The development of PTSD has been linked to various negative outcomes that further diminish a patient's ability to cope with their physical disabilities. These include; substance abuse (Seal et al, 2011), hypertension (Burg et al, 2017; Cohen et al, 2009; Howard et al, 2018; Kibler et al, 2009), weight gain and obesity (Cohen et al, 2009; Kubzansky et al, 2014), coronary artery disease and mortality (Ahmadi et al, 2011), including suicide (Krysinska & Lester, 2010; Sundin et al, 2014).

In a review of papers reporting the prevalence of mental health comorbidities among older veterans with amputation, depression was reported between 10% and 28% and PTSD between 15% and 46% (Caddick et al, 2019). A systematic review also reported the prevalence and variable levels of anxiety (16% to 35.5%) and psychological distress (13% to 36%) among military personnel with a physical impairment (Stevelink et al, 2015). Levels of depression and anxiety appear more pronounced among individuals with LLA up to 2 years following amputation followed by a gradual decline to levels similar to the general population (Horgan & MacLachlan, 2004).

2.3.9 Summary

Whilst it is important to remain mindful of the many secondary health outcomes that individuals with traumatic LLA may be predisposed to following their injury, it is also important to be aware of how this literature may inform the population of interest (i.e. UK military personnel currently receiving rehabilitation at DMRC). Many of the previous studies used in this section are from the US or Iranian military. Whilst these groups are likely to have experienced their traumatic LLA whilst young adults, the rehabilitation care pathways (including prosthetic provision) and the subsequent national health care systems available to them are very different to the UK. For example, the traditional continuous 4 weeks residential rehabilitation model adopted by DMS is unique to the UK military (see chapter 3 for more details). Much of the evidence used to support long-term chronic conditions detailed here are from US servicemen injured during the Vietnam War where the mechanisms of injury differed. There have also been advances in emergency care medicine since the Vietnam War, with UK servicemen surviving injuries that would have proved fatal to US servicemen during this conflict, therefore presenting a different challenge to UK DMS.

The outcomes that demonstrate unfavourable changes in short-term cardiometabolic health are primarily based on retrospective administrative reports rather than prospective observations using objective methods of assessment. The majority of the studies mentioned used body mass index (BMI), skinfolds or waist:hip circumference ratios to determine levels of obesity. Whilst informative, only one study (Eckard et al, 2015) has used dual-energy X-ray absorption (DEXA), a more accurate method of determining total body composition.

Based on the available evidence, it is unclear what impact traumatic LLA has on many of these secondary health outcomes in injured servicemen who are still embedded within a rehabilitation care pathway. To my knowledge, no prospective longitudinal study has used objective methods of assessing cardiometabolic health in individuals with LLA injured during the Iraq/Afghanistan conflicts, where access to advanced prosthetic provision is commonly available. Also, none of these studies attempted to quantify PA, one of the primary modifiable risk factors in a lot of these chronic conditions. Accurate information on PA levels in individuals with LLA may help explain some of the high incidences of secondary health conditions reported in the literature so far. In order for UK DMS to better understand the risk of developing some of these secondary health conditions in their severely injured servicemen and women, further exploratory research embedded within their unique rehabilitation care pathway is warranted.

2.4. PHYSICAL FUNCTION OF INDIVIDUALS WITH LOWER-LIMB AMPUTATION

Among the primary goals of any rehabilitation programme for people with LLA is the aspiration to improve physical function. An understanding of the factors that influence function and the ability to perform ADL may inform rehabilitation programmes and facilitate social, community and vocational integration. However, first it will be useful to identify the criteria used to classify functional performance and identify reference values of physical function for individuals with LLA.

2.4.1. Classifying Levels of Physical Function and Reference Values for Individuals with Lower-Limb Amputation

Table 2.1 describes the Medicare Functional Classification Level (MFCL). Medicare established the K-Levels in 1995 to quantify need and the potential benefit of prosthetic devices for patients with LLA. The rating system is still commonly used today by insurance companies to determine eligibility for payment or reimbursement, but not traditionally by the UK DMS.

Table 2.1: Medicare Functional Classification Level (MFCL) for individuals with lower-limb amputation

MFCL Classification	Definition
K-Level 0	Does not have the ability or potential to ambulate or transfer safely with or without assistance, and a prosthesis does not enhance quality of life or mobility.
K-Level 1	Has the ability or potential to use a prosthesis for transfers or ambulation in level surfaces at a fixed cadence. Typical of the limited and unlimited household ambulator.
K-Level 2	Has the ability or potential for ambulation with the ability to transverse low-level environmental barriers such as curbs, stairs, or uneven surfaces. Typical of the limited community ambulator.
K-Level 3	Has the ability or potential for ambulation with variable cadence. Typical of the community ambulator who has the ability to transverse most environmental barriers and may have vocational, therapeutic, or exercise activity that demands prosthetic use beyond simple locomotion.
K-Level 4	Has the ability or potential for prosthetic ambulation that exceeds basic ambulation skills, exhibiting high impact, stress, or energy levels. Typical of the prosthetic demands of the child, active adult, or athlete.

Two of the most commonly used methods of assessing physical function in individuals with LLA include the six-minute walk test (6-MWT) and the Amputee Mobility Predictor Questionnaire (AMPQ). Physical function reference values for different aetiologies of LLA are demonstrated in table 2.2. Based on the mean reference values provided, level of function increases linearly with every increase in K-level, military personnel demonstrate greater levels of function than civilians, trauma-based mechanisms of injury achieve greater levels of function than vascular-based mechanisms and transtibial LLA achieve superior levels of function than transfemoral LLA. Reference values for distances walked in six minutes by the general population (aged 20 to 50 years) range between 459 and 738 metres (Chetta et al, 2006) and provide a useful reference distance for community integration.

Table 2.2: Reference values for the six-minute walk test and the Amputee Mobility Predictor Questionnaire (AMPQ) for a variety of lower-limb amputee populations. Data are presented as mean ± SD and (range)

Amputation Group	6-MWD (metres)	AMPQ score
Unilateral Amputation		
Civilian mixed aetiology [a]		
K-Level 0 to 1	50 ± 30 (4 to 96)	25 ± 7 (14 to 33
K-Level 2	190 ± 111 (16 to 480)	35 ± 6 (19 to 46)
K-Level 3	299 ± 102 (48 to 475)	41 ± 4 (26 to 46)
K-Level 4	419 ± 86 (264 to 624)	45 ± 2 (38 to 47)
Peripheral Vascular Disease [b]	410 ± 66 (298 to 463)	37 ± 2 (35 to 39)
US military (Trauma) [c]		
Transtibial	661 ± 87 (433 to 858)	46 ± 1 (42 to 47)
Transfemoral	542 ± 67 (442 to 686)	43 ± (41 to 46)
Bilateral Amputation		
US military (Trauma) [d]		
Transfemoral	452 ± 141 (264 to 645)	35 ± 3 (32 to 39)

Abbreviations: 6-MWD = six-minute walk distance, AMPQ = Amputee Mobility Predictor Questionnaire. References values used include: [a] = (Gailey et al, 2002), [b] = (Gailey et al, 2012), [c] = (Gailey et al, 2013b), and [d] = (Raya et al, 2013).

Reference values like the ones demonstrated in table 2.2 are useful for clinicians and patients when it comes to goal setting. However, in an effort to attain these reference values of physical function, it is important to be aware of factors that affect functional capacity and inactivity in LLA.

2.4.2. Factors Effecting Functional Capacity and Inactivity

Sedentary lifestyles and reduced physical function contribute to the increased morbidity and mortality observed in LLA. Self-reported QoL measures consistently show significantly worse physical health outcomes in traumatic amputees compared with population norms (Dougherty, 2001; 2003; Hagberg & Branemark, 2001; Hoogendoorn & van der Werken, 2001; MacKenzie et al, 2004; Pezzin et al, 2000). Poor long-term physical function scores have been demonstrated after traumatic amputation (Akula et al, 2011; Bosse et al, 2002; Doukas et al, 2013; MacKenzie et al, 2005). At 7 years post-injury, only one-third (34.5%) of civilians with LLA had a functional status (measured using three components of the Sickness Impact Profile questionnaire; (1) walking (2) mobility and (3) body care and movement) typical of the general population of similar age and gender (MacKenzie et al, 2005).

The ability of individuals with LLA to perform common functional tasks associated with ADL like standing from and sitting in a chair (Highsmith et al, 2011), negotiating uneven terrain (Lamoth et al, 2010), ascending and descending stairs (Jones et al, 2006; Schmalz et al, 2007) and hills (Vrieling et al, 2008) have all been reported to be significantly impaired compared to non-amputation controls. Older research exploring gait in individuals with traumatic unilateral transtibial and transfemoral amputation in both military and civilian populations have demonstrated higher EE compared to individuals without amputation, resulting in greater physical stress (expressed as heart rate or oxygen consumption) at specific speeds of walking (Bell et al, 2014; Czerniecki, 1996; Detrembleur et al, 2005; Genin et al, 2008; Ward & Meyers, 1995). Increased EE could be explained by postural asymmetry induced by compensatory strategies caused by the lack of ankle musculature in the lower-limb that now require other larger proximal muscles of the amputated limb to be recruited to power movement of the centre of mass (Genin et al, 2008). However, more recent analysis using UK military personnel with unilateral amputation, demonstrate comparable gait (temporal spatial parameters) and metabolic profiles to active male controls at self-selected walking speeds are possible (Jarvis et al, 2017). These findings were attributed to greater access to advance prosthetic provision and prolonged intensive rehabilitation not freely available to civilians or previous generations of injured military personnel. How these favourable temporal spatial gait parameters and metabolic profiles in serving UK military personnel with traumatic LLA transfer to habitual PA levels is not yet fully understood.

Daily step counts below 5,000 have been classified as ‘sedentary’ in adult populations (Tudor-Locke et al, 2011). Previous research investigating PA levels in individuals with LLA report mean daily step counts ranging between 1,540 and 4,217 steps per day (Halsne et al, 2013; Klute et al, 2006; Parker et al, 2010; Stepien et al, 2007). A recent study performed at DMRC Headley Court (UK) investigated the daily step count of individuals with bilateral LLA during rehabilitation and whilst at home (Sherman et al, 2019). Mean daily step count significantly reduced (39%) from 2,258 ± 192 during in-patient rehabilitation to 1,387 ± 363 at home ($p < 0.01$) and were indicative of a sedentary activity levels. In Vietnam War veterans with unilateral amputation, 1% do not walk, 63% walk in households or the community, and 20% engage in low or high-impact activity. When compared to military personnel with amputation from Iraq/Afghanistan, 1% do not walk, 42% walk in households and communities, and 52% engage in low or high-impact activities (Reiber et al, 2010). There remains a paucity of evidence on the implications of various amounts of PA on the health and well-being of adults with LLA. No recommended guidelines exist for the optimal level of PA (i.e. number of steps per day or quantity of PAEE) necessary to optimize health benefits for individuals with LLA. However, before measuring PA in individuals with traumatic LLA, determining factors that influence prosthetic wear time to enable ambulatory PA is important.

2.4.2.1. Determinants of Prosthetic Wear Time and Ambulatory Physical Activity

A lower-limb prosthesis is commonly provided to replace an absent limb to facilitate ambulation and functional independence. Prosthetic devices can improve functional ability, enhance mobility and safety, facilitate higher levels of activity, and can also reduce the risk of secondary comorbidities and problems resulting from overuse of intact limbs among veterans with amputation(s) (Gailey et al, 2008; Reiber et al, 2010). Prosthetic usage has been shown to vary by type of amputation. For instance, a series of long-term follow-up studies of Vietnam veterans conducted by Dougherty revealed that 87.5% of individuals with unilateral transfemoral amputation were current prosthetic users (average of 13.5 hours per day) compared with just 22% of people with bilateral transfemoral amputation (average of 7.7 hours per day), thereby highlighting the significant additional impact of multiple compared to unilateral LLA (Dougherty, 1999; 2001; 2003). Among individuals with bilateral LLA, 33% of Vietnam veterans (compared with just 6% of Iraq/Afghanistan veterans) could no longer walk wearing a prosthesis (Dougherty et al, 2010). Differences in functional status and PA levels between UK military personnel with traumatic unilateral and bilateral LLA are not yet known (this will be explored in greater detail in chapter 4 and chapter 6, respectively). However, access to a comfortably fitted prosthesis is likely to play a significant role.

Complications associated with prosthetic socket fitting may negatively impact participation in structured exercise and/or habitual PA thereby limiting the ability to perform ADL and fully integrating into society. Poorly fitting prosthesis, symptomatic neuromas and stump infections have all been reported as reasons for increases in residual limb pain. Both bone pathology (bone spurs and sharp bone ends, heterotrophic ossification and stress fractures) and soft tissue pathology (excess soft tissue, failure of muscle reconstruction to protect bone ends, symptomatic scar tissue and wound breakdown) have also been implicated (Perkins et al, 2012). Chronic skin problems in the residual limb can often cause serious limitations in mobility and QoL (Demet et al, 2003; Pezzin et al, 2004). Skin exposed to weight-bearing areas of the socket is not always resilient to the pressure and friction caused by the socket during ambulation (Van de Meent et al, 2013). Approximately one third of individuals with transfemoral amputation report skin damage associated with prosthetic socket fitting (Hagberg & Branemark, 2001; Lyon et al, 2000; Meulenbelt et al, 2009). The occurrence of complications associated with skin health of the residual limb can vary from 18% to 75% (Schoppen et al, 2001b; Sinha et al, 2011; 2014). The most prevalent problem assessed in US military personnel with LLA was sweating inside the socket, with 67% of Vietnam and 62% of Iraq/Afghanistan veterans identifying sweating as a problem (Reiber et al, 2010). Increases in injury severity (i.e. more proximal amputations) are associated with an increased likelihood of any infection, osteomyelitis, and non-healing wounds (Melcer et al, 2017). The level and number of LLA also has an impact on functional performance.

2.4.2.2. Determinants of Functional Performance with Different Levels / Number of Lower-Limb Amputation(s)

Poorer scores in the 6-min walk test (6-MWT) (Lin et al, 2014) are found to correlate with lower levels of PA. In US servicemen a significant contrast in ambulatory function (209 metres) was demonstrated between individuals with unilateral transtibial amputation (661 ± 87 m) and bilateral transfemoral amputation (452 ± 141m) during the 6-MWT (Linberg et al, 2013). Suggesting individuals with bilateral transfemoral amputation find ambulation more challenging and therefore may be at increased risk of physical inactivity.

Gaunaurd et al. (2013) predicted high level mobility measured using the Comprehensive High-Level Activity Mobility Predictor assessment in US servicemen with LLA. The most common factors that significantly contributed to higher levels of mobility include; the ability to ascend/descend stairs, sitting down from standing, reduced waist circumference, number of intact knee joints, greater time spent with an amputation and reduced injury severity (Gaunaurd et al, 2013). In order to perform maximally on the Comprehensive High-Level Activity Mobility Predictor assessment, it is important to perform quick, alternating eccentric and concentric contractions of the lower-limb musculature to generate maximum power in multiple planes. A rehabilitation intervention aimed at improving lower-limb strength, power and dynamic balance could lead to improved high-level mobility and subsequently facilitate higher levels of habitual PA and long-term positive cardiometabolic health (see section on muscle atrophy and strength later in this chapter: section 2.6.2). Another benefit from increased levels of physical function includes current and future employment prospects.

2.4.2.3. Employment Status

Employed individuals have greater prosthetic use than those who are unemployed (Schoppen et al, 2001b; Whyte & Carroll, 2002). The ability to return to work following a traumatic LLA is dependent on numerous factors including age, preinjury vocational ability, level of amputation, residual limb health, associated injuries, social support and the national disability systems (Perkins et al, 2012). Assessments of physical function and pain at 3 months post-injury are significant predictors of later return to work (MacKenzie et al, 2006). For individuals who do return to work it is rarely to their previous job role. Most often, the individual returns to a less physically demanding role or they require job modifications (Hebert & Ashworth, 2006; Schoppen et al, 2001a; Whyte & Carroll, 2002). Among service members, amputation resulted in only 11% to 13% continuing on activity duty (Belisle et al, 2013; Hurley et al, 2015) with approximately 85% who continued on active duty returning with a job modification to a less physically demanding role (Hurley et al, 2015).

2.4.3. Summary

Improvements in physical function are a key determinant of any successful rehabilitation programme. The level of function achieved by individuals with LLA appears to be affected by aetiology, level (transtibial or transfemoral) and number of (unilateral or bilateral) LLA. Caution must be applied when interpreting findings using civilians with LLA compared to military due to the different mechanisms of injury (predominantly blast during combat) and access to rehabilitation and prosthetic services is likely more uniform in the military. In addition, the underlying physical and social determinants of health-related QoL may differ between veteran amputees compared to civilians (Christensen et al, 2016). Soldiers are typically younger men, exposed to superior pre-injury physical conditioning, higher levels of self-efficacy, and a robust support network, all of which correlate with better outcomes (Doukas et al, 2013). Access to the more advanced prosthetic design and technology has also facilitated additional improvements in physical function, making direct comparisons between older and modern day research also more challenging. It is widely acknowledged that the DMS care pathway differs considerably from most civilian care pathways (discussed in greater detail in chapter 3 section 3.6) making direct comparisons to civilian based research problematic. However, it remains unclear what effect the current UK DMS combat casualty care pathway has on the functional status of military personnel with traumatic LLA.

One component associated with PA and function which is integral to any personalised rehabilitation program is cardiorespiratory fitness. The next section discusses the role of cardiorespiratory fitness in individuals with LLA and its role in optimising health outcomes.

2.5. CARDIORESPIRATORY FITNESS

The assessment of cardiorespiratory fitness represents the synergistic functioning of multiple organ systems to effectively transport oxygen from the air to the mitochondria of the working skeletal muscle to produce the necessary energy to meet the demands of activity as well as effectively remove the resultant metabolic by products that impair the ability of the muscle to sustain activity when accumulated in excess (Lavie et al, 2019). Common measures of cardiorespiratory fitness include the peak attainable rate of oxygen uptake ($\dot{V}O_2$ peak) or peak workload achieved during an incremental test to volitional exhaustion. A strong relationship between low levels of cardiorespiratory fitness and functional limitations has previously been demonstrated in uninjured men (n = 3,495) and women (n = 1,175) over the age of 40 (Huang et al, 1998). In able-bodied populations, high levels of cardiorespiratory fitness are associated with reduced prevalence of several cardiometabolic risk factors including hypertension, hyperlipidemia, inflammation, insulin resistance and lower incident rates of metabolic syndrome and type 2 diabetes mellitus (Booth et al, 2017). Indeed, physical inactivity leads to a decrease in cardiorespiratory fitness, increasing the risk of numerous chronic diseases/conditions (Bauman et al, 2016; Knaeps et al, 2016; Lavie et al, 2015b; Olsen et al, 2008; Warburton & Bredin, 2016). In addition, men with high cardiorespiratory fitness display significantly lower levels of abdominal adipose tissue compared with those with low cardiorespiratory fitness (Wong et al, 2004). Considerable evidence indicates that high levels of cardiorespiratory fitness significantly attenuate or even eliminate the elevated risk of cardiovascular disease and all-cause mortality in overweight and obese individuals (Elagizi et al, 2018; Fletcher et al, 2018; Kennedy et al, 2018; Lavie et al, 2015a; Wisloff & Lavie, 2017). Lavie et al. (2019) summarises some of the potential physiological benefits of cardiorespiratory fitness on prognosis for cardiovascular disease. These benefits include reduced blood pressure (BP), improved heart rate variability, improved endothelial function, improved insulin sensitivity, increased mitochondrial density, reduced systemic inflammation, decreased myocardial demands, maintenance of lean muscle mass, reduced visceral adipose tissue (VAT), increased capillary density, improved mood and stress levels and improved sleep quality. It is perhaps unsurprising that cardiorespiratory fitness has been demonstrated to be an independent predictor of cardiovascular disease risk, cardiovascular disease related mortality and total mortality in uninjured populations (Kaminsky et al, 2013; Kodama et al, 2009; Lee et al, 1999).

A relationship between a reduced aerobic capacity and the ability to walk with prosthesis has previously been observed (Chin et al, 2002; Chin et al, 2006; Sansam et al, 2009; Wezenberg et al, 2019). It is therefore widely believed cardiorespiratory fitness is an outcome of habitual physical activity. Epidemiological data linking cardiorespiratory fitness with long-term health outcomes in able-bodied adults, at least indirectly, implies that PA has a key role to play in the prevention of chronic disease in individuals with traumatic LLA, particularly its role in energy balance and preventing obesity.

2.6. BODY COMPOSITION FOLLOWING TRAUMATIC LOWER-LIMB AMPUTATION

2.6.1. Obesity

Excess adiposity typically evolves slowly over time, due to a long-term positive energy balance. Human energy balance can be expressed as:

$$\textit{Energy Balance} = \textit{Energy Intake} - \textit{Energy Expenditure}$$

Although an increase in total body adipose tissue is associated with an increase in health risk, body fat location may have a more important impact on the development of cardiometabolic risk factors and related diseases than excess total adiposity (Despres & Lemieux, 2006). The amount of VAT has been associated with an increased risk of comorbidities such as type 2 diabetes mellitus, coronary heart disease, stroke, sleep apnoea, hypertension, dyslipidaemia, insulin resistance, inflammation, and some types of cancer (Tchernof & Despres, 2013). Excess adiposity also imposes a mechanical load on joints, making obesity a risk factor for the development of osteoarthritis (Goldring & Otero, 2011). The role of excess VAT on insulin resistance and chronic inflammation will be discussed in more detail in section 2.7 of this literature review.

Unfortunately weight gain and obesity are common following traumatic amputation (Bouldin et al, 2016; Eckard et al, 2015; Foote et al, 2015; Kurdibaylo, 1996; Littman et al, 2015; Rose et al, 1987; Shahriar et al, 2009). Greater waist circumference (used as a measure of central adiposity) is associated with poorer levels of physical function (Gaunaurd et al, 2013) and is a predictor of cardiovascular disease in people with traumatic LLA (Mozumdar & Roy, 2006). Eckard et al. (2015) investigated the body composition changes during the first year following traumatic amputation in US servicemen using DEXA. They found pre-injury BMI was exceeded 6 months following amputation in both unilateral and bilateral groups with significant increase in fat mass during the first 9 months following unilateral amputation. Additionally, there was clinically significant, but not statistically significant, weight gain of 14.5 kg from baseline to 12 months in individuals with bilateral LLA. However, Eckard et al. (2015) only commented on 'total fat mass', 'trunk fat mass' and 'lean mass' measurements (and their respective percentages of total body mass), not the distribution of adipose tissue (i.e., gynoid and android fat mass or visceral adipose tissue (VAT) area measurements) which are more commonly reported and known to be more strongly associated with unfavourable cardiometabolic health changes over time.

In a 22 year follow-up study of 327 Iranian males with combat-related traumatic bilateral LLA, Shahriar et al. (2009) observed abdominal obesity in 82.5% of the study sample and high levels of triglycerides (32%). The authors conclude that the majority of individuals with trauma-related bilateral amputation are at an increased risk of cardiovascular disease in the near future. Numerous factors have been identified that may predispose individuals with LLA to increased rates of abdominal obesity. These include living a sedentary life, low physical activity, lack of nutrition education leading to unhealthy eating habits (increased calories, low-nutrient dense foods and excessive alcohol intake), pain and psychotropic medications, psychological issues, using a wheelchair to mobilise instead of prosthesis, and unemployment (Dougherty, 1999; Fazizi et al, 2004; Frugoli et al, 2000; Kurdibaylo, 1996; Modan et al, 1998; Naschitz & Lenger, 2008; Rose et al, 1987; Shahriar et al, 2009; Trappe et al, 2007). Thigh fat percent is significantly greater in the residual limb following transfemoral and transtibial amputations compared with the intact limb (Sherk et al, 2010). Excess thigh fat mass is thought to impair prosthetic control and negatively impact socket-residual limb mechanics due to increased movement of the limb within the socket compared to the interface with the socket that may come from lean muscle tissue, thereby unfavourably altering gait (Haboubi et al, 2001; Kuiken et al, 2018). Increased BMI is also a contributing factor to decreased prosthetic fit rates (Webster et al, 2012), prosthetic use (Rosenberg et al, 2013), and ambulatory (K) level classification (Kulkarni et al, 2015). MFCL (K) levels are described in table 2.1, further complicating the ability of this population to engage in regular ambulatory-based PA. At present there is a lack of information on energy balance in this population, due to the lack of accurate methods to measure habitual EE (discussed in section 2.10). Furthermore, there are several limitations with subjective diet diaries which include a large responsibility burden on the participant (literacy and high motivation required and increased possibility of under reporting). Diet diaries are also time-consuming if there is a requirement for several days of measurement and possible recall bias (Shim et al, 2014).

2.6.2. Muscle Atrophy and Weakness

Skeletal muscle is an adaptable tissue that responds to multiple stressors during a lifetime to modify/maintain muscle mass. During adulthood, the maintenance of muscle mass and adaptive growth is dependent on two primary factors: external loading and neural activation. Decreases in the amount of external loading and/or neural activation of a muscle will lead to a loss of muscle mass and are often referred to as 'disuse atrophy' (Atherton et al, 2016). Severe illnesses and injuries typically impose a period of immobilisation that, depending on the clinical context, may be temporary or permanent and either localised (e.g., limb casting) or generalised (e.g., bed rest). Although immobilisation can sometimes be of benefit to a patient (e.g., allowing a fracture to heal), it always has a negative side effect of facilitating skeletal muscle disuse atrophy (Atherton et al, 2016). Skeletal muscle is one of the most vital tissues impacted by the effects of physical inactivity (Ferrucci et al, 2016). It is widely recognised that muscle mass loss can negatively impact the ability to perform daily tasks, increase the incidence of future injury, prolong the period of rehabilitation, increase the final cost to the health care provider, and in severe cases reduce overall prognosis (Hunter et al, 2004). The role of skeletal muscle tissue in the metabolic control of humans is explored in greater detail in section 2.7.

The most frequent employed models to study disuse atrophy in humans are unilateral limb suspension using a knee brace or cast, and bedrest (Rudrappa et al, 2016). Studies have demonstrated losses of muscle strength and mass early on in disuse. For example, 5 days of cast immobilisation lead to ~3.5% reductions in quadriceps cross-sectional area and ~9% in strength (Dirks et al, 2014). This had progressed to ~8% reductions in cross-sectional area and ~23% reductions in strength by 14 days (Wall et al, 2013). Men who had their lower-limb immobilised following a tibial fracture (thus having 6-weeks of casting) reported reductions in quadriceps cross-sectional areaof ~17% (Gibson et al, 1987). Furthermore, it has been reported that 90 days of bed rest can lead to ~10 and 16% reductions in quadriceps and triceps surae mass after 29 days, with rates of weekly loss slowing during the last 2 months to roughly half that observed during the first month (Alkner & Tesch, 2004). It appears atrophy occurs more rapidly in the first 3 to 14 days of unloading and eventually reaching a nadir where further loss of muscle occurs at a slower rate despite continued unloading of muscle (Bodine, 2013).

Depending on the severity of the injury, extended bed rest in a hospital environment is often essential for military personnel with combat related LLA. They are therefore predisposed to considerable muscle atrophy and strength deficits early in their treatment care pathway (Isakov et al, 1996; Jaegers et al, 1995; Moirenfeld et al, 2000; Renstrom et al, 1983a; Renstrom et al, 1983b; Sadeghi et al, 2001; Schmalz et al, 2001; Sherk et al, 2010; Tugcu et al, 2009). This is likely due to decreased use of the muscle tissue (Tugcu et al, 2009), as well as reduced muscle fibre size which may negatively influence the function of the residual limb and prosthesis (Renstrom et al, 1983b) and complicate the ability to perform ADL (Moirenfeld et al, 2000). The level of amputation and length of the residual limb are significant factors in the severity of muscle atrophy (Isakov et al, 1996; Jaegers et al, 1995). Muscle recruitment strategies for gait and joint stabilization change after amputation (Centomo et al, 2008; Sadeghi et al, 2001) with numerous studies reporting differences in thigh strength and circumference between amputated and non-amputated limbs (Isakov et al, 1996; Renstrom et al, 1983a). Renstrom et al. (1983b) found that muscle fibre size in the vastus lateralis of individuals with transtibial amputation were less than the intact limb. The amputated limb had a greater proportion of type IIb fibres, and the intact limb had a greater proportion of type IIa fibres. Schmalz et al. (2001) has reported a 15% to 30% reduction in muscle cross-sectional area and muscle volume of the quadriceps, in the amputated limb of individuals with unilateral transtibial amputation compared to the intact limb. Another study found concentric muscle strength of the quadriceps and hamstrings in the amputated limb ranged from 40% to 60% of the intact limb, with eccentric quadriceps strength being weaker in individuals with shorter residual tibia, and individuals with shorter residual tibia had weaker hamstrings (Isakov et al, 1996).

Individuals with unilateral/bilateral transfemoral amputation are missing osseous structures and original insertion sites for the quadriceps and hamstring muscles and therefore have a disadvantage to individuals with transtibial amputation when generating force and maintaining balance during high-level mobility (Gaunaurd et al, 2013). The presence of the knee joint enables individuals with transtibial amputation to change direction faster and maintain posture and balance with greater ease (Gailey et al, 2013b). Individuals with transfemoral amputation are more susceptible to postural asymmetries at the pelvis and hip and degenerative changes to the intact hip, knee, and ankle joints that may impair function and restrict activity over the long-term (Gailey et al, 2008; Gaunaurd et al, 2011). Asymmetry in weight bearing between the contralateral and amputated limb during sit-to-stand and stand-to-sit tasks has been demonstrated in individuals with transfemoral, concluding that the activity was primarily performed with the contralateral limb (Highsmith et al, 2011).

The ability to descend and ascend stairs is a leading determinant of higher levels of mobility in individuals with traumatic LLA (Gaunaurd et al, 2013). This task requires dynamic single-limb balance and stabilisation of the limb/musculature within the socket, sufficient lower-limb strength (via eccentric and concentric contraction) and range of motion to raise and lower the body to the next step (McFadyen & Winter, 1988). Therefore, impaired lower-limb muscle strength from quadriceps, hamstrings, and gluteal muscles could affect high-level mobility performance. Due to the inability to flex their prosthetic knee voluntarily, individuals with transfemoral amputation are unable to take advantage of the energy storage and return properties of carbon fibre prosthetic feet. They are therefore confined to utilising the prosthetic foot in its capacity of support and not energy production (Gailey et al, 2013b).

The above information demonstrates the clinical importance of retaining lean muscle mass and strength for the benefit of short and long-term functional independence of individuals with traumatic LLA. The direct relationship between the amount of muscle and metabolic health will be discussed in the next section.

2.7 BIOMARKERS OF CHRONIC DISEASE

2.7.1. Overview of Metabolic Control

During times of dramatic fluctuations in energy supply and demand, the human body must evolve to manage energy metabolism for optimal substrate storage and use during states of either food surplus or famine, and periods of either rest or increased energy demand (Smith et al, 2018). The ability to efficiently adapt metabolism depending on demand or supply is known as 'metabolic flexibility'. Maintaining metabolic homeostasis during fasting or feeding (postprandial) relies on the coordinated control of available fuel across multiple organs. During the postprandial period of a carbohydrate-rich meal for instance, pancreatic β-cells respond to the rise in nutrients by releasing insulin into the bloodstream. The liver is triggered to absorb glucose from the circulation under the influence of insulin and stop glycogenolysis (breakdown of glycogen into glucose) and gluconeogenesis (generation of glucose from fats or protein). Skeletal muscle tissue assists in glucose removal as insulin receptor binding of insulin results in translocation of glucose transporters to the plasma membrane, allowing glucose to enter the cell (Smith et al, 2018). Adipose tissue responds to insulin by decreasing the rate of lipolysis and stimulating fatty acid and triglyceride synthesis from glucose and lipids (Dimitriadis et al, 2011). Collectively, this ensures hyperglycaemia is minimalised within exposed tissues and nutrients are stored in adipocytes for release and oxidation in times of scarcity. Postprandial fatty acids trigger adipose tissue to reduce the release of nonesterified fatty acid (NEFA) and encourage hepatocytes to reduce the release of endogenous triglyceride, together stimulating the clearance of circulating triglycerides (Frayn, 2002).

However, prolonged positive energy balance leads to obesity which directly impedes metabolic flexibility. Obesity is predominantly associated with elevated levels of plasma free fatty acids. High circulating levels of free fatty acids inhibit glycogen synthase activity, which leads to reduced disposal and oxidation of glucose. Excess calories are then stored in subcutaneous fat depots as triglycerides; when these depots reach their maximum capacity and fail to expand, ectopic fat accumulates. The liver and VAT store this excess energy (ectopic triglycerides accumulation) and in the process, become dysfunctional, leading to low-grade chronic inflammation, dyslipidemia, insulin resistance, and ultimately, type 2 diabetes mellitus and cardiovascular disease (Despres, 2011; Heymsfield et al, 2014; Smith et al, 2018; Speakman, 2013; Tahergorabi et al, 2016). Obesity is associated with a state of chronic low-grade inflammation because ectopic fat depots release more inflammatory mediators than subcutaneous fat depots and infiltration of macrophages (Paniagua, 2016). Therefore, fat deposition and metabolic inflexibility are likely to reinforce one another in a vicious cycle.

Skeletal muscle is a critical glucose disposal site during postprandial conditions, especially if glycogen stores are not full and there is some level of energy demand due to contractile activity. If skeletal muscle is being regularly contracted, adenosine triphosphate demand is elevated, and then there is a need for increased glucose to be actively transported into muscle cells (Thyfault, 2008). Therefore, higher levels of activity and depletion of glycogen stores promote increased insulin sensitivity. However, if skeletal muscles are inactive, adenosine triphosphate demand is low (skeletal muscle mitochondrial capacity and β-oxidation are reduced), and glycogen is elevated, demand for glucose is low and thus insulin sensitivity decreases. Depots of glucose (stored in muscle and liver glycogen) and fatty acids (stored in adipose tissue or within muscle and liver) can be rapidly mobilized to fuel muscle contraction and other essential processes until the work is done and food is procured. Therefore, higher levels of activity and depletion of glycogen stores promote increased insulin sensitivity (Booth et al, 2017). Glucose is tightly regulated as it is a substrate in limited supply (only ~4 g in circulation and ~300–500 g of glycogen in muscle and liver (Wasserman, 2009)), and maintenance of glucose levels are critical for brain metabolism and consciousness. Taken together, metabolic flexibility can be understood as an adaptive response of an organism's metabolism to maintain energy homeostasis by matching fuel availability and demand to periodic fasting, varying meal composition, physical activity, and environmental fluctuations (Carstens et al, 2013; van Ommen et al, 2009).

2.7.2. Insulin Resistance

Insulin resistance often precedes hyperinsulinemia and hyperglycaemia is a pre-requisite for type 2 diabetes mellitus (DeFronzo & Tripathy, 2009). Insulin Resistance can be characterised as the failure of insulin to exert the normal cellular effects on various tissues, leading to the impairment of insulin mediated glucose disposal. Fasting hyperglycaemia can persist due to the insensitivity of the liver to the suppressive effects of insulin on gluconeogenesis and reduced glycogenolysis (Lee et al, 2009). Subsequently this can lead to impaired glucose tolerance, which is clinically diagnosed as a fasting plasma glucose concentration between 5.6 – 6.9 mmol·l-1 (American Diabetes, 2014). Persistent hyperglycaemia can lead to significant complications in the future such as coronary and peripheral artery disease (King et al, 2005).

Daily PA is highly correlated to insulin sensitivity, and this is only modestly attenuated by adiposity (Balkau et al, 2008). Indeed, inactivity is a key etiological factor in the development of insulin resistance through two mechanisms: 1) inactivity itself lowers insulin sensitivity, and 2) inactivity provides a permissive environment whereby signalling molecules can impair insulin signalling processes and further reduce insulin sensitivity (Booth et al, 2017). Immobilisation results in the development of whole body and muscle insulin resistance in people within 3 to 5 days (Hamburg et al, 2007; Smorawinski et al, 2000; Sonne et al, 2011). Insulin resistance can develop after 3 to 14 days of reduced ambulatory activity (Knudsen et al, 2012; Krogh-Madsen et al, 2010; Olsen et al, 2008). Young men who were physically active (walking ~10,500 steps.day) and changed their lifestyle so they approached 1,400 steps.day for 2 weeks (Krogh-Madsen et al, 2010; Olsen et al, 2008), indicated a 57% increase in insulin area under the curve (AUC) response to an oral glucose tolerance test (OGTT). Even with a shorter and smaller change in daily activity levels, insulin sensitivity can be lowered. After reducing from ~10,000 steps per day to ~5,000 steps per day, for only 3 to 5 days provided similar results in terms of elevated glucose, and insulin responses to an OGTT and elevated free-living postprandial glucose levels measured by continuous glucose monitors (Mikus et al, 2012; Reynolds et al, 2015). One of the key factors linking the expansion of adipose tissue with the development of insulin resistance is chronic low grade inflammation (Weisberg et al, 2003). This is because immune cells (macrophages and T lymphocytes) stored within adipose tissue, are a source of inflammatory cytokines (e.g., Tumor Necrosis Factor Alpha and Interleukin-6) (Fain, 2010), which can interfere with skeletal muscle insulin signalling. Systemic inflammation and metabolic inflexibility can cause a vicious circle because metabolic inflexibility can also trigger systemic inflammation (Smith et al, 2018).

Exogenous essential amino acids (needing to be acquired through dietary protein intake) stimulates muscle protein synthesis, insulin suppresses muscle protein breakdown (and stimulates muscle glucose uptake). Essential amino acids and insulin are therefore vital in maintaining muscle metabolic homeostasis and failure of these mechanisms inevitably leads to skeletal muscle atrophy and insulin resistance (Rudrappa et al, 2016). Loss of local contractile stimulation induced through sitting leads to both the reduction of glucose uptake and the suppression of skeletal muscle lipoprotein lipase activity (which is necessary for HDL cholesterol production and triglyceride uptake) (Bey & Hamilton, 2003; Hamilton et al, 2004). Insulin resistance (using homeostatic model assessment - HOMA-IR) is associated with reduced quadriceps muscle strength (Kalyani et al, 2013; Leenders et al, 2013); power (Kalyani et al, 2013) and muscle mass (Leenders et al, 2013) in able-bodied adults. Therefore, not only does inactivity promote insulin resistance through reduced utilisation of glucose in muscle, but it may also promote the increased storage of free fatty acids and glucose into adipose tissue. As such, adiposity and insulin resistance may develop at the same time through these processes.

Impaired metabolic health has been reported in individuals with traumatic amputation (Modan et al, 1998; Peles et al, 1995). Both Peles et al. (1995) and Modan et al. (1998) investigated the plasma insulin and glucose response to an oral glucose load of 100g in older individuals with traumatic LLA. Hyperinsulinemia was evident (Peles et al: 128 ± 67.4 $\text{pmol}\cdot\text{L}^{-1}$, Modan et al: 123 ± 59 $\text{pmol}\cdot\text{L}^{-1}$) and both studies reported elevated glucose responses after one hour (~10.3 $\text{mmol}\cdot\text{L}^{-1}$) and two hours post-glucose loading (~9.3 $\text{mmol}\cdot\text{L}^{-1}$). Elevated insulin response was also observed after one hour (Peles et al: 612 ± 315 $\text{pmol}\cdot\text{L}^{-1}$, Modan et al: 563 ± 421 $\text{pmol}\cdot\text{L}^{-1}$) and two hours (Peles et al: 802.8 ± 423.6 $\text{pmol}\cdot\text{L}^{-1}$, Modan et al: 795.9 ± 462 $\text{pmol}\cdot\text{L}^{-1}$), respectively, indicating insulin resistance. These data suggests an increased risk of insulin resistance in individuals with traumatic LLA. Given the previously argued reasons for reduced PA in traumatic LLA, it seems reasonable to assume that physical inactivity plays a considerable role in the development of insulin resistance and in the long-term the progression to type 2 diabetes mellitus in this population. This is in conjunction with the pathophysiology of lower extremity skeletal muscle atrophy and increased relative adiposity observed in this population.

2.7.2.1. Measurement Techniques

The pathophysiology of type 2 diabetes mellitus involves insulin resistance and β-cell dysfunction (Kahn, 2003; Matthews, 2001; Meier & Bonadonna, 2013). The ability to accurately estimate insulin resistance and β-cell function is therefore essential for screening high-risk subjects for type 2 diabetes mellitus. There are several methods for estimating insulin sensitivity/resistance and β-cell function, however, hyperinsulinemic euglycemic clamp, developed by Defronzo et al. (1979) is considered the gold standard measurement (DeFronzo et al, 1979). This method involves intravenous administration of insulin at a constant rate to raise and maintain systemic insulin levels (i.e., hyperinsulinemia). The concentration of plasma glucose is monitored every 5 to 10 minutes, and kept constant by the manipulation of the exogenous glucose infusion rate. Theoretically the rate of glucose infusion required to maintain constant plasma glucose concentrations can be equated to the rate of glucose disposal into all tissues in the body. When combined with isotopic glucose tracers this technique can provide information on tissue specific insulin sensitivity. Based on practical limitations, such as the requirement for experienced clinical staff to manage any technical difficulties and it being expensive and time consuming to perform, hyper-insulinemic euglycemic clamp measurement techniques were deemed unsuitable for chapter 7 of this thesis. Furthermore, the clamp utilises supra-physiological steady state insulin concentrations and ignores absorption from the gastrointestinal tract. It is therefore unable to adequately reflect the normal physiological conditions that a dynamic test such as an OGTT or a mixed meal might deliver.

The OGTT is simple and cost-effective to perform and is widely used to detect patients with impaired glucose tolerance and type 2 diabetes mellitus (American Diabetes, 2014). Following a 10 to 12 hour overnight fast, baseline blood samples are taken prior to and at regular time points for 2 hours following the ingestion of a standard oral glucose load (75 g). Concentrations of glucose and insulin are analysed from these blood samples to provide an indication of metabolic health. Whilst the OGTT provides useful information regarding glucose tolerance, the test itself does not provide specific information on insulin sensitivity/resistance *per se* (Muniyappa et al, 2008). However, simple surrogate indices can also be derived from fasting samples, such as the Homeostasis Model Assessment (HOMA) model, that can be used to indirectly measure insulin resistance (HOMA-IR) (Turner et al, 1979) and β-cell function (HOMA-β) (Matthews et al, 1985) using the formulas provided below. Previous studies show that an increase of HOMA-IR and a decrease of HOMA-β are associated with an increased incidence of diabetes and future cardiovascular events in patients with type 2 diabetes mellitus (Bonora et al, 2002; Sung et al, 2010).

Homeostasis model assessment for insulin resistance (HOMA-IR) is calculated using the following formula (Turner et al, 1979):

$$HOMA\text{-}IR = Fasting\ glucose\ (mmol{\cdot}l^{-1})\ x\ fasting\ insulin\ (mU{\cdot}l^{-1})\ /\ 22.5$$

Homeostasis model assessment for pancreatic β-cell function (HOMA-β) is calculated using the following formula (Matthews et al, 1985):

$$HOMA\text{-}\beta = (Fasting\ insulin\ (mU{\cdot}l^{-1})\ x\ 20)\ /\ (Fasting\ glucose\ (mmol{\cdot}l^{-1})\ -3.5)$$

Formulae which incorporate at least the two main parameters (i.e., insulin and glucose) are preferable over those utilising insulin alone. Such formulae represent the exchangeable kinetics between both parameters which ultimately estimate insulin resistance (Borai et al, 2011). The reasons for the wide application of HOMA based models in the scientific literature are their simplicity and practicality as only one fasting sample is required and there is no requirement for administration of a glucose load. However, simple indices based on fasting levels of glucose and insulin (e.g. HOMA-IR Model) is not always reliable because it does not consider the variations in glucose resistance of the peripheral tissue and liver (i.e. the reduction in the suppression of hepatic glucose output, by hyperglycaemia, and the reduction of peripheral glucose-stimulated glucose uptake) (Wallace et al, 2004). Hepatic insulin resistance is considered the major determinant of fasting hyperglycaemia and as such is the major factor contributing to the pre-diabetic state, impaired fasting glucose (Howard et al, 1996; Sanyal et al, 2004). Whilst hepatic and peripheral insulin resistance correlate with each other, the relative contribution of each varies between individuals. In most circumstances peripheral tissue insulin resistance develops later than hepatic insulin resistance (Sanyal et al, 2004). This is an important limitation of fasting simple indices. Indices based solely on fasting measurements of glucose or insulin cannot always reliably estimate insulin resistance, since it is possible for subjects to be significantly insulin resistant without having fasting hyperinsulinaemia. Furthermore, some individuals may be euglycaemic when fasting but hyperglycaemic and hyperinsulinaemic two hours following a 75g oral glucose load. It is recognised that even in healthy individuals with hepatic insulin resistance regular diet, exercise or glucose lowering medications can restore both fasting glucose and insulin levels to well within normal ranges (Ferrannini & Balkau, 2002; Mason et al, 2011). There is now an updated HOMA calculator (HOMA2), recalibrated to give steady-state β-cell function (% B) and insulin sensitivity (% Sensitivity of 100% in normal young adults) (Levy et al, 1998). The upgraded HOMA2 model has been observed to be more closely associated with reference test derived indices and consequently more reliable and accurate (Borai et al, 2011) with the HOMA2 model modified to allow for increased plasma and insulin and glucose levels (Wallace et al, 2004). HOMA2-IR, HOMA2-β and HOMA2-S are now recommended to use as they obtain the strongest correlation with reference methods (Borai et al, 2011). The HOMA2 outputs can be determined using the calculator downloaded and provided online at https://www.dtu.ox.ac.uk/homacalculator.

When selecting an index that reflects the physiological insulin response to a glucose load, it is appropriate to use one derived from parameters measured during an OGTT (Borai et al, 2011). These surrogate indices are derived from a dynamic response which generally incorporates both peripheral and hepatic insulin resistance. The larger the number of specimens collected from an individual, the more accurately the derived parameters are likely to reflect insulin sensitivity. The advantage of the OGTT-derived indices over those derived from fasting insulin and glucose alone is that they can detect subtle disturbances in glucose metabolism not apparent from the latter (Kanauchi et al, 2007).

Whole body insulin sensitivity can be measured using the Matsuda Index (ISI Matsuda), which can be calculated from the ratio of plasma glucose to insulin concentration over the course of the 120 minutes following a glucose load (see formula below). This method is highly correlated ($r = 0.73$, p

< 0.001) to the rate of whole-body glucose disposal during the gold standard euglycemic insulin clamp (Matsuda & DeFronzo, 1999). The Insulin Sensitivity Index (ISI Matsuda) is calculated using the following formula (Matsuda & DeFronzo, 1999):

$$ISI_{Matsuda} = 10{,}000 \surd \text{ [Fasting glucose } (mg{\cdot}dl^{-1}) \text{ x fasting insulin } (\mu U{\cdot}ml^{-1})] \text{ x [mean OGTT glucose value } (mg{\cdot}dl^{-1}) \text{ x mean OGTT insulin value } (\mu U{\cdot}ml^{-1})]$$

It is also noteworthy that the early glucose response during a OGTT can be considered an index of hepatic insulin resistance, while the drop in glucose levels from peak to nadir estimates peripheral insulin resistance predominantly of skeletal muscle with a smaller contribution from adipose tissue (Abdul-Ghani et al, 2007).

2.7.3. Metabolic Syndrome

Metabolic syndrome is a clinical diagnosis characterised by the clustering (≥ 3) of the following metabolic abnormalities; abdominal obesity (i.e. waist circumference), raised blood pressure (BP), depressed HDL cholesterol, elevated fasting concentrations of glucose and triglyceride (Grundy et al, 2006). Individuals diagnosed with metabolic syndrome present the same clinical threat as those with type 2 diabetes mellitus (Ford, 2005) and demonstrate a two-fold increase in developing cardiovascular disease over the following 5 to 10 years compared to controls (Alberti et al, 2009). Consequently, individuals with metabolic syndrome have increased mortality and a shortened lifespan (Guize et al, 2007).

Numerous studies have demonstrated that the pathogenesis of metabolic syndrome is largely attributable to a lack of fitness and PA (Roberts et al, 2013). Greater prevalence of metabolic syndrome has been indicated in participants with lower levels of fitness. Lakka et al. (2003) noted associations of cardiorespiratory fitness and leisure-time PA activity with metabolic syndrome in a sample of 1,069 middle-aged men without cardiovascular disease, type 2 diabetes mellitus, or cancer. Men who engaged in one hour/week or less of moderate-intensity leisure-time PA were 60% more likely to have metabolic syndrome than those engaging in three hours/week or more (Lakka et al, 2003). Greater muscle strength and lean muscle tissue has also been associated with a lower prevalence of abnormal metabolic syndrome components. Muscular strength, as determined by one-repetition maximum (1-RM) bench press and leg press, has been shown to be inversely associated with each of the five components of metabolic syndrome (Jurca et al, 2004).

The International Diabetes Foundation published criteria for metabolic syndrome (Zimmet et al, 2005). These criteria (for men) include:

- Increased ‘abdominal’ obesity: Waist circumference ≥ 94 cm

- Hypertriglyceridemia: ≥ 1.7 mmol/L
- Reduced HDL cholesterol: < 1.03 mmol/L
- Hyperglycaemia: Fasted plasma glucose ≥ 5.6 mmol/L
- Hypertension: Systolic BP ≥ 130 mm Hg and/or diastolic BP ≥85 mm Hg

There is evidence to suggest these component risks may occur at a heightened frequency in individuals with traumatic LLA. Ejtahed et al. (2017) found a higher prevalence of metabolic syndrome in Iranian military veterans with bilateral LLA compared to the general populations. In this cross-sectional study, the prevalence of metabolic syndrome among amputees was 62.1% (95% CI: 55.9% to 68.4%) (Ejtahed et al, 2017). Health-related QoL scores were comparable between the two groups; however the mean weight, waist and hip circumferences, systolic and diastolic BP, triglyceride, HDL and LDL cholesterol were significantly higher in the amputation group with metabolic syndrome compared to participants without amputation ($p < 0.05$). Patients with traumatic LLA appear to be two times more at risk of developing metabolic disorders including obesity, hypertension, hyperlipidaemia, and hyperinsulinemia (Magalhaes et al, 2011; Naschitz & Lenger, 2008).

2.7.4. Chronic Inflammation

A positive energy balance will ultimately result in an increase in visceral and subcutaneous adipose tissue followed by the infiltration of pro-inflammatory T cells and macrophages leading to chronic low-grade systemic inflammation (Gleeson et al, 2011). Adipose tissue is an endocrine organ capable of secreting a number of proteins, collectively called adipokines. These include various hormones and cytokines, such as C-Reactive Protein (CRP), Interleukin-6, leptin and adiponectin which can all impact energy homeostasis, metabolism, inflammation and immunity. Chronic low-grade inflammation caused by altered adipokine secretion may alter glucose and lipid metabolism and contribute to cardiometabolic risk of individuals with visceral obesity (Ferrante, 2007; Trayhurn & Wood, 2005). Adipocyte size and adipose tissue distribution are key determinants of inflammatory cytokine secretion (Skurk et al, 2007). Many adipokines involved in the aetiology of insulin resistance and inflammation (e.g., Tumor Necrosis Factor Alpha , Interleukin-6) are up-regulated with increased adiposity while other insulin-sensitising or anti-inflammatory adipokines (e.g., Adiponectin) are down-regulated as fat mass increases resulting in metabolic complications (Kaur, 2014; Maury & Brichard, 2010; Tchkonia et al, 2013) Of these circulating inflammatory markers CRP is measured in chapter 7 of this thesis.

CRP is an acute-phase protein of hepatic origin that increases in response to inflammation, trauma and/or infection. Following Interleukin-6 secretion by macrophages and T cells, its physiological role is to bind to dead or dying cells in order to activate the complement system thus promoting cellular removal by macrophage phagocytosis (Crossland et al, 2019). It is therefore characterised as a well standardised marker of systemic inflammation and is considered the most accurate inflammatory marker to predict future risk of cardiovascular events (Nimmo et al, 2013; Schillinger et al, 2003; Tsimikas et al, 2006). CRP is regulated by other pro-inflammatory cytokines such as Tumor Necrosis Factor Alpha and Interleukin-6 and as such circulating CRP may be an upstream representative of their activity (Ridker, 2016). CRP has been shown to be highly associated with VAT, insulin resistance, BP, LDL cholesterol, and triglycerides (Roberts et al, 2013). CRP appears to be inversely associated with self-reported PA, aerobic fitness (Aronson et al, 2004a; Aronson et al, 2004b; Lavie et al, 2011) and loss of lean body mass (Dutra et al, 2017; Schaap et al, 2006).

Physical function status, physical inactivity and the subsequent alterations in body composition, predisposes individuals to an increased likelihood of chronic disease and impaired metabolic control compared to able-bodied adults. Various psychosocial and environmental barriers mean that it is difficult for individuals with traumatic LLA to engage in PA. Moreover, due to functional limitations typical of complex blast and/or blunt force-related trauma, exercise options can often be limited. The current available evidence on PA levels in individuals with LLA shows that the majority of this population is predisposed to living sedentary lifestyles. Considering the independent role that PA has on metabolic health and inflammation and subsequently body composition, it is important to understand how it is quantified. This review will now focus on the components of EE, and the measurement of these in individuals with LLA.

2.8. ENERGY EXPENDITURE: COMPONENTS

It is important to recognise the difference between EE and PA. Total energy expenditure can be subdivided into basal metabolic rate, diet-induced thermogenesis and PAEE. Basal metabolic rate typically represents the largest component of total EE (Landsberg et al, 2009) and is the energy required to sustain metabolic activities of cells and tissues within the body to maintain homeostasis. Basal metabolic rate and resting metabolic rate (RMR) are often used interchangeably. To estimate basal metabolic rate accurately, participants are required to sleep in a specifically designed respiration chamber (a calorimeter), where carbon dioxide (CO_2) production and oxygen (O_2) uptake can be measured. Due to logistics and equipment limitations, this is not always possible. Therefore, the resting component of total EE is often used instead. RMR is influenced by body mass, specifically fat-free mass (Schofield, 1985), which, as mentioned previously (section 2.6.2) is known to be reduced in individuals with traumatic LLA due to the loss of metabolically active tissue and difficulties in eliciting site specific muscle hypertrophy following injury.

PAEE is characterised by the thermic effect of any movement produced by a skeletal muscle contraction which exceeds RMR (Westerterp, 2009). Due to its high variability amongst free-living individuals, it is often considered the most important component of total EE. However, due to its variable nature it can be very problematic to accurately translate human movement into units of PAEE. This task is even more challenging in individuals with LLA during free-living, attributable to atypical gait.

2.9. ENERGY EXPENDITURE: CRITERION MEASURES

2.9.1. Direct Calorimetry

Despite being considered the gold standard method for measuring total EE, direct calorimetry is not widely used due to its large cost and operational complexity. The method involves quantifying heat exchange between the human body and the environment of a participant whilst they remain unaccompanied in an airtight isolation chamber for 24 hours or more. Measuring heat released by the body, as well as water vapour released through respiration and from the skin, the measurements recorded represents the combustion of energy in the form of carbohydrate, protein or fat (Schutz, 1995). As this method of measuring EE is not practical or possible within the rehabilitation environment of DMRC, this review will focus on applicable measures.

2.9.2. Doubly Labelled Water

Doubly labelled water is a method used to measure free-living total EE in humans. The doubly labelled water method is based on the principle of isotope dilution, and total EE is measured via estimating whole-body CO_2 production (Speakman, 1998). Participants orally ingest a dose of water containing stable isotopes of both oxygen and hydrogen. Within a couple of hours the isotopes (O^{18} and H^2) mix with the more abundant forms of hydrogen and oxygen found in the endogenous body water pool throughout the body. As energy is expended by the body, H_2O and CO_2 are produced. Hydrogen is removed from the body via urination and sweating; O^{18} is removed via these same routes, but also lost as CO_2 which is expelled via the lungs during exhalation. Consequently, with both isotopes being ingested simultaneously, their rate of elimination will differ as O^{18} will be eliminated at a faster rate than H^2. The disappearance rate of both isotopes is determined by measuring their concentrations in urine samples collected over an allotted period of time (typically 7 to 14 days). The difference in the elimination rate of H^2 and O^{18} reflects the rate at which CO_2 is produced (Ainslie et al, 2003). EE can then be estimated using the Weir equation, assuming a mean respiratory quotient value of 0.85, indicative of a standard westernised diet (Westerterp, 1999).

The doubly labelled watertechnique is widely considered the ‘gold standard’ for measuring total EE during free-living as EE can be accurately reported during habitual daily routines over extended periods without the interference of equipment attached to a participant. However, this method is not without its limitations. Minimal information regarding frequency, duration or intensity of activity can be obtained (Plasqui & Westerterp, 2007). This technique is also expensive, requires sophisticated equipment and trained personnel to operate it. Nevertheless, the values it produces provide the closest measure of free-living EE, making the doubly labelled water method an extremely valuable criterion measure for validating estimates of energy requirements obtained by other methods. The validation of the doubly labelled water method has been previously described (Schoeller & Hnilicka, 1996). To our knowledge one paper has used doubly labelled water as a method of assessing PA with an amputation cohort (Kaufman et al, 2008). However, it is unclear whether the various assumptions for the prediction of EE remain with clinical populations who are considered predisposed to secondary health complications such as LLA.

2.9.3. Indirect Calorimetry

The principles underpinning indirect calorimetry is based on the rate and type of substrate utilisation whereby energy metabolism is estimated from respiratory gas exchange measurements (i.e. oxygen consumption [$\dot{V}O_2$] and carbon dioxide production [$\dot{V}CO_2$]), (Ainslie et al, 2003), as an approximate measure of an individual’s respiratory quotient. Respiratory quotient refers to the quantity of CO_2 produced in relation to O_2 consumed at tissue level. This indirect measurement of respiratory quotient is referred to as the respiratory exchange ratio (RER). Carbohydrates, fats and proteins differ in their chemical composition and in the amounts of O_2 needed and CO_2 produced when oxidised. Therefore based on measures of the ratio (O_2 and CO_2) it is possible to obtain measures of the mixture of fuels being oxidised under different conditions (Jeukendrup & Wallis, 2005).

Whilst direct and indirect calorimetry was equivalent when using a whole room calorimeter (Seale et al, 1990), the indirect calorimetry method provides unique information, is non-invasive and more adaptable than direct calorimetry. One of the most common indirect calorimetry methods, the Douglas bag technique (Douglas, 1911) is still referred to as the gold standard for measuring oxygen uptake in the laboratory (Gladden et al, 2012). The Douglas bag method is usually confined to a laboratory, light-weight and fully-portable equipment can continually measure gas exchange variables typically between 1 to 5 hours (Ainslie et al, 2003). When best practice is adhered to this method has high reliability, CVs of 0.5% for both CO_2 and O_2 (Hopker et al, 2012).Technology has now advanced to online systems to enable instantaneous breath-by-breath pulmonary gas exchange measurements that are both valid and reliable (Carter & Jeukendrup, 2002) and not available using classic Douglas Bag methodology. These portable systems are reliable instruments for measuring respiratory gas exchange (Meyer et al, 2001) and more applicable to the assessment of everyday activity due to its ability to be used outside of the laboratory. However, the short memory capacity and battery life can make them impractical for use for extended periods of time. Furthermore, the wearing of portable units may interfere with everyday habitual activities during free-living assessment and therefore not always deemed practical.

Indirect calorimetry methods for energy expenditure measurements are based on the following assumptions (Jeukendrup & Wallis, 2005; McLean et al, 1987; Porter & Cohen, 1996): (1) Any fuel consumed has an intrinsic energy content that upon Metabolic modifications in the living system will result in heat or energy production. (2) The combustion or synthesis of carbohydrate, fat, or protein is the end result of all the biochemical reactions occurring in the body. (3) The oxidation of glucose, fat, or protein results in a substance specific fixed ratio between the quantities of O_2 consumed and CO_2 produced. (4) Loss of substrates is negligible in faeces and urine. (5) RER adequately reflects respiratory quotent. While $\dot{V}O_2$ will reliably reflect tissue O_2 uptake, $\dot{V}CO_2$ will only be a reliable estimate estimate of tissue CO_2 production I the presence of a stable bicarbonate pool. At low to moderate exercise intensities this is unlikely to be a problem since no accumulation of hydrogen ions occurs and lactate production in the muscle will be matched closely by lactate clearance through oxidation and gluconeogenesis. At higher exercise intensities (above intensities corresponding to maximal lactate steady state), it could be argued that shifts in the acid base occur. Under circumstances of increase glycolytic flux, lactate will accumulate in the contracting muscle and begin to move into the extracellular fluid. The increase in hydrogen will be buffered by bicarbonate and ultimtely excess (non-oxidative) CO_2 will be excreted through hyperpnea. This will have the effect of elevating $\dot{V}CO_2$ and therefore overestimate carbohydrate and underestimate fat oxidation. This would imply that as soon as Hydrogen ions and lactate accumulate in the muscle, estimations of fat oxidation by indirect calorimetry are flawed. Notwithstanding the limitations of those assumptions, as previously stated, indirect calorimetry has been found to be consistent and in close agreement with direct calorimetry.

2.10. ENERGY EXPENDITURE: PREDICTION IN INDIVIDUALS WITH TRAUMATIC LOWER-LIMB AMPUTATION

Recognising it is not always feasible to use criterion methods to measure free-living EE, researchers have endeavoured to develop accurate, user-friendly, unobtrusive methods to predict EE during free-living. The development of these methods usually involves a validation study using one of the criterion measures mentioned above. This section will provide a summary of the available prediction tools to measure PA and which methods have been previously used by researchers investigating populations with amputation.

2.10.1. Commonly Used Methods to Measure Physical Activity

Considering the definition of PA as any body movement produced by the skeletal muscles that results in an EE (Caspersen et al, 1985), there is a great difficulty in measuring it accurately. PA is highly heterogeneous and there is no single outcome measure that can capture all the relevant information about a given individual (Thompson & Batterham, 2013). As a construct, PA is multi-dimensional comprising of four key components; frequency, intensity, duration and type. The intensity component is an important determinant of metabolic health benefits resulting from PA in able-bodied populations (Haskell et al, 2007). The activity counts derived from wearable devices capture raw movement signals and can be used to describe both duration and intensity of a given task (Matthews et al, 2012). Intensity can also be captured by metabolic equivalents (METs) which are often used to express the energy cost of a PA as multiples of RMR. One MET is defined as an oxygen uptake of 3.5 $mL.kg^{-1}.min^{-1}$, however, it is unclear whether this conventional MET value is applicable to individuals with traumatic amputation due to their loss of metabolically active muscle tissue. It is beyond the scope of this thesis to address this question.

2.10.2. Self-Report Measures

Self-reported PA monitoring is not routinely used within UK DMS. However, self-reported questionnaire have previously been used in research investigating activity levels of individuals with amputation elsewhere. The most commonly used questionnaire in LLA populations is the Physical Activity Scale for Individuals with Physical Disabilities (Devan et al, 2012; Littman et al, 2014). The Physical Activity Scale for Individuals with Physical Disabilities contains 13 questions and follows a similar structure to the International Physical Activity Questionnaire (Craig et al, 2003). Questions refer to information about an individual's leisure time (including walking, and use of a wheelchair), household and work related activities conducted over the past 7 days. The intensity is determined by multiplying the standard MET value developed using healthy populations by the mean hours/day for each item. Scores are determined by multiplying the mean number of hours/day by the number of days/week. The International Physical Activity Questionnairehas also been used in individuals with amputation (da Silva et al, 2011), however, caution must be applied as this questionnaire has not been validated using individuals with amputation.

2.10.3. Strengths and Limitations of Self-Report Questionnaires

Self-report PA questionnaires are cheap, easy to administer, widely used and have an advantage over other direct methods, such as movement sensors, as they enable participants' behaviours to be recorded (type of activity), which is a relevant factor for the evaluation of exercise programs and PA interventions (Dollman et al, 2009). However, the results from these questionnaires depend on the accuracy of the participants' memory and recall. It has been suggested that self-report measures are unable to adequately quantify the lower end of the PA continuum (Shephard, 2003) and often lend themselves to recall bias and participant over-reporting (Lagerros & Lagiou, 2007). One of the main limitations of the Physical Activity Scale for Individuals with Physical Disabilities questionnaire is the use of standard MET values as a measure of activity intensity irrespective of the participant's level or type of disability. Furthermore, the standard MET values employed were developed for healthy individuals; it is not clear whether these values can be directly applied to a population with LLA.

Patients with LLA at DMRC are required to complete a range of clinical outcome measures as part of their admissions and these can be considered quite burdensome. Therefore, as part of this thesis, no additional self-reported outcome measures will be measured. However, objective body-worn sensors will be used to estimate EE. Conveniently objective sensors overcome these shortcomings of self-reporting by removing the subjective recall element.

2.10.4. Pedometers

Pedometers record movements performed in response to the body's vertical acceleration via a mechanical counter. A pedometer is traditionally attached to an individual's waist and counts the number of steps in a time interval. It enables the cumulative measurement of leisure, domestic, occupational and transportation activities. A pedometer is also an objective and sensitive method to quantify individual's PA (Colpani et al, 2013). However, although it is suitable to evaluate movements (steps/day) by vertical oscillations of the body, it is unable to quantify horizontal displacement (Butte et al, 2012). Pedometers are also unable to evaluate activities such as cycling, resistance training, PA in water based environments (i.e. swimming) or discriminates between the intensity and type of PA. Adults are considered to be sedentary if they take less than 5,000 steps/day (Tudor-Locke et al, 2011).

2.10.5. Heart Rate

Heart rate (HR) is known to increase linearly and proportionately with exercise intensity and thus oxygen uptake (Chen et al, 2012). After adjusting for gender, age, body mass and fitness, HR has been found to be an accurate predictor of PAEE (Keytel et al, 2005). However, the relationship between EE and HR weakens during lower intensity PA (Luke et al, 1997). This is most likely due to small postural changes causing alterations in stroke volume, or that HR during low intensity PA is also affected by external factors such as ambient temperature, hydration status, emotional stress, and illness (Davidson et al, 1997; Melanson & Freedson, 1996). Whilst HR monitoring provides an indication of physiological strain and a general picture of PA which is cheap and reusable, when used in isolation it is unlikely to offer the most accurate method for obtaining estimates of EE.

2.10.6. Accelerometry

The main advantage of accelerometers is they can record not only the total accumulated activity, but also the intensity, frequency and duration of these activities (Lagerros & Lagiou, 2007; Westerterp, 2009). Commercially available accelerometers are compact, in that they are relatively unobtrusive and overall compliance in large population based studies has been shown to be high (Matthews et al, 2012). Increased on-board memory capacity and extended battery life of newer generations of accelerometers allow extended monitoring periods of habitual PA with higher time resolutions. It is perhaps unsurprising that accelerometer-based devices are frequently used to quantify habitual free-living PA in able bodied cohorts (Plasqui et al, 2005). Various studies have also supported the utility of accelerometers to determine variability in activity levels in ambulatory populations with disabling conditions such as multiple sclerosis, rheumatoid arthritis, stroke and Parkinson’s (Cervantes & Porretta, 2010; Hale et al, 2008; Khemthong et al, 2006; Rand et al, 2009). However, at present there is a lack of research focussing on the accurate prediction of PAEE using accelerometers in individuals with traumatic LLA.

2.10.6.1. Methodological Considerations When Using Accelerometry to Predict PAEE

The two main types of accelerometers used widely in PA research include uniaxial and more increasingly tri-axial monitors. Uniaxial accelerometers register movement in the vertical axis only, whereas tri-axial accelerometers register movement in the anteroposterior (X), mediolateral (Y) and vertical (Z) axes. During laboratory and free-living validation studies in uninjured populations, triaxial accelerometers have demonstrated greater sensitivity in detecting movement compared to uniaxial accelerometers, suggesting triaxial accelerometers may better predict PAEE (Van Remoortel et al, 2012). One week monitoring periods have routinely been used in previous research to provide a sufficient quantity of days to achieve intraclass correlations > 80%, whilst also providing the opportunity to measure behaviour during both weekdays and weekends (Matthews et al, 2012). Almost all commercially available movement sensors report their outcomes in counts per unit of time or epoch. Counts derived from movement sensors are arbitrary units which are commonly derived in three ways; summing the integral of the modulus of acceleration from each axis, summing the integral of the route mean squared acceleration from each axis and summing the vector magnitude of acceleration from each axis (Cook et al, 2011; Horner et al, 2011). Waist-mounted accelerometers, positioned within close proximity to an individual's centre of mass, have been frequently used in uninjured cohorts. However, the accuracy of single units worn on the waist can be limiting for certain types of activity capture that have a low ambulatory component and may involve upper body work (Matthews et al, 2012). The measurement error of waist mounted devices is generally related to the inability to detect arm movements as well as static work (lifting, pushing, carrying loads) which are all common components of gym based exercise programme performed during rehabilitation which may underestimate PA (Butte et al, 2012; Lagerros & Lagiou, 2007). Ambiguity regarding the most appropriate anatomical wear location that offers the most sensitivity to estimate PAEE in this population remains. An objective of chapter 5 is to address this issue.

Best practice guidelines for validating activity monitors (Bassett et al, 2012) recommend evaluating devices over a wide range of activities of various intensities. Judging the validity of a movement sensor based solely on the strength of its relationship to a criterion measure should be avoided as it does not indicate the agreement between the two variables (Bland & Altman, 2010). Measurement error is a statistical consideration proved useful for researchers to report in the analysis of accelerometer data (Staudenmayer et al, 2012). To date relatively few studies have attempted to develop and then cross-validate regression equations capable of accurately estimating PAEE in disabled populations, and none to my knowledge in individuals with LLA.

2.10.6.2. Accelerometer-Based Monitoring in Individuals with Lower-Limb Amputation

Table 2.3 provides a summary of the studies to have detailed PA of LLA populations using accelerometer based methods. To date, Bussmann and colleagues (Bussmann et al, 2004; Bussmann et al, 2008) have validated the uniaxial IC-3031 and ADX202 body fixed accelerometer in unilateral amputees. However, the aim of these two investigations was only to assess the accelerometers ability to identify posture and motion by comparing outputs to video recordings. Although useful for identifying movements performed in controlled rehabilitation settings, these studies did not attempt to predict EE, nor the metabolic cost of prosthetic ambulation. The most commonly used PA monitor, which has been validated in amputation populations, is the StepWatch^{TM} Activity Monitor. The StepWatch^{TM} is a step activity accelerometer designed specifically for the amputee population (Hordacre et al, 2014). Typically worn on the lateral side of the prosthetic pylon, this pager-sized device (70 x 50 x 20 mm; 38 g) contains a sensor that records the number of steps taken during a 1-minute sampling interval. After adjustments are set for cadence and motion settings according to the user's stature and walking style, this measure of total steps taken in a day has been shown to be accurate (Raya et al, 2010) and to have high test-retest reliability (Kent et al, 2015; van Dam et al, 2001). However, habitual PA may not always be accurately recorded using a prosthesis based accelerometer as an individual may need or prefer to remove the prosthesis to perform ADL. An Actigraph GT1M accelerometer has previously been used to assess the effects of two types of microprocessor-controlled prosthetic knee joints on perceived performance and everyday life activity level (Theeven et al, 2012). However, it is important to note this uniaxial accelerometer has not been validated in populations with amputation, so although they were able to ascertain raw counts, this information cannot accurately translate to PAEE. Using algorithms intrinsic to certain devices may not be generalisable to a target population (Pedisic & Bauman, 2015), an important consideration for individuals predisposed to significant gait or movement deficiencies such as individuals with LLA (Sagawa et al, 2011; Su et al, 2007).

Table 2.3: Summary of reported physical activity levels and study details

Study	Number and age of participants	Aetiology	Type of physical activity monitor	Location of Monitor	Duration of monitoring	Physical activity variables recorded	Physical activity levels reported
Unilateral Transtibial Amputation							
(Bussmann et al, 2004)	N = 9 Mean age = 55	Vascular	ADX202 accelerometer	Sternum, upper thigh on both limbs	2 days	Time spent active (hours) Time spent walking (hours) Number of sit-to-stand transitions/day	Mean = 10.1 Mean = 0.9 Mean = 43.1
(Bussmann et al, 2008)	N = 9 Mean Age = 55	Trauma	ADXL202 accelerometer	Sternum, upper thigh on both limbs	2 days	Time spent in dynamic activities (hours) Time spent walking (hours) Number of sit-to-stand transitions/day	Mean = 1.7 Mean = 1.2 Mean = 55.8
(Coleman et al, 2004)	N = 13 Mean age = 49	Trauma	StepWatch[a]	Prosthetic ankle	2 week	Steps/day Time spent active (hours)	Range = 2,262 to 4,135 Mean = 3.4 to 5.8
(Gailey et al, 2012)	N = 10 Mean age = 61 and 51	5 Vascular 5 Non-vascular	Step Activity Monitor[a]	Prosthetic ankle	10-14 days	Steps/day	Mean = 2,702 – 4,336 (Vascular) Mean = 6,202 – 7,465 (non-vascular) Range = 1656 - 5971 (Vascular) Range = 4,689 13,274 (non-vascular)
						Time spent active (hours)	Mean = 3.14 - 3.69 (vascular) Mean = 4.26 – 5.22 (non-vascular) Range = 2.3 - 5.7 (vascular) Range = 2.6 – 8.8 (non-vascular)
(Hordacre et al, 2014)	N = 46 Mean age = 60	18 Vascular 19 Trauma 9 Other	StepWatch 3 and GPS	Prosthetic ankle	1 week	Steps/day	Mean = 3,612 (home) Mean = 2,378 (community)

(Hordacre et al, 2015)	N = 46 Mean age = 60	18 Vascular 17 Trauma 11 Other	StepWatch 3 and GPS	Prosthetic ankle	1 week	Steps/day	Median = 3,441 (home) Median = 2,124 (community)
(Kanade et al, 2006)	N = 21 Mean age = 63	Vascular	StepWatch [a]	Prosthetic ankle	1 week	Steps/day	Mean = 1,941
(Kent et al, 2015)	N = 22 Mean age = 52	7 vascular 13 Trauma 2 Other	Actigraph [a]	Prosthetic Pylon	3 week	Steps/day	Mean = 4,581 Range = 2,497 to 8,305
(Klute et al, 2011)	N = 5 Mean age =56	4 trauma 1 vascular	StepWatch 3	Not Specified	2 weeks	Steps/day	Mean = 2,714 and 5,214
(Segal et al, 2014)	N = 10 Mean age = 56	5 Trauma 4 vascular 1 Cancer	StepWatch 3	Prosthetic Pylon	1 week	Steps/day	Mean = 6,269 and 6,728
(Selles et al, 2005)	N = 26 Mean age = 67 and 58	11 Trauma 14 Vascular 1 Other	Temec Instruments activity monitor [a]	Sternum, mid-thigh on both limbs	2 days	Time spent in dynamic activities (hours) Time spent walking (hours)	Mean = 1.5 and 2.2 Mean = 1.1 and 1.8
Unilateral Transfemoral Amputation							
(Albert et al, 2014)	N = 9 Mean age = 53	6 Trauma 2 Vascular 1 Other	Fitbit One	Waist	1 week	Steps/day Fitbit activity score Time spent active (hours)	Mean = ~4,000 Range = ~0 to 8,200 [b] Mean = ~400 Range = 50 to 630 [b] Mean = ~3.6 Range = ~1.2 to 6.0 [b]
(Hafner & Askew, 2015)	N = 12 Mean age = 58	10 Trauma 2 Other	StepWatch 3	Prosthetic Ankle	60 consecutive	Steps/day	Mean = 1,942 and 2,239

					days over 2-14 months		
(Hafner et al, 2007)	N = 17 Mean age = 48	10 Trauma 7 Other	StepWatch 2	Not specified	2 months	Steps/day	Mean = ~1,800 [b]
(Halsne et al, 2013)	N = 17 Mean age = 50	10 Trauma 7 Other	StepWatch 2	Prosthetic Ankle	145 to 359 days	Steps/day	Mean = 1,540 Range = 497 to 2675
Mixed (Transfemoral and Transtibial Amputations)							
(Buis et al, 2014)	N= 48 TTA Mean age = 50 and 61	12 Vascular 36 unknown	ActivPAL	Prosthetic Ankle	Up to 6 days	Steps/day	Mean = 9,130 and 7,383 Range = 1,570 16,815
(Klute et al, 2006)	N = 12 TTA N= 5 TFA Mean age = 54 and 48	11 Trauma 4 Vascular 2 Other	StepWatch 2	Distal end of Prosthetic limb	1 week	Steps/day Time spent active (hours)	Mean = 2,657 and 2,976 Mean = 4.2 to 4.7
(Lin et al, 2014)	N= 12 TTA N= 8 TFA Mean age = 51	12 Trauma 7 vascular 1 Other	Impulse Pedometer, model B1	Waist	1 week	Steps/day	Mean = 4785
(Stepien et al, 2007)	N = 54 TTA N = 23 TFA Mean age = 60	39 Trauma 23 Vascular 15 Other	StepWatch 3	Prosthesis	6 days	Steps/day Time spent active (hours)	Mean = 2,284 and 3,395 Mean = 10.1
(Theeven et al, 2012)	N = 24 TFA N = 6 KD Mean age = 59	23 Trauma 6 Vascular 1 Tumor	Actigraph GT1M (uniaxial)	Waist	1 week	Counts per day Time spent active (hours)	All = 117,852 Low MFCL-2 = 48,322 Intermediate MFCL-2 = 114,165 High MFCL-2 = 156,304 Mean = ~7.7
(Parker et al, 2010)	N= 27 Mean age = 55	20 vascular 26 Trauma	StepWatch 3	Prosthetic Ankle	1 week	Steps/day	Mean = 4,217 Range = 249 to 12714

		6 Other				Time Spent active (hours)	Mean = 3.78 Range = 0.77 to 10.5
Bilateral Amputation							
(Sherman et al, 2019)	N = 9 Mean age = 26	Trauma	LAM2 ActiPAL	Prosthesis	2 weeks	Steps/day	Mean = 2,258 (rehabilitation) Mean = 1,387 (home)

Adapted from Pepin et al. (2017)

[a] Type or model of activity monitor is not specified in the reviewed manuscript. [b] Estimated physical activity level based on visual examination of published graphs.

Abbreviations: TTA: transtibial amputation, TFA: transfemoral amputation, KD: knee disarticulation, MFCL: medicare functional classification system, GPS: global positioning system

Accelerometry data alone is unable to capture the physiological strain associated with certain ambulatory behaviours, such as walking up a gradient (Lamonte & Ainsworth, 2001). Multi-sensor devices, incorporating physiological signals, might offer a greater improvement in prediction accuracy (Strath et al, 2005).

2.10.7. Multi-Sensor Physical Activity Devices

Multi-sensor technologies, which incorporate both accelerometry and physiological parameters, have great potential for increasing the accuracy of predicting PAEE as they reduce the weaknesses of using HR and accelerometry in isolation. The Actiheart^{TM} (AHR) (Cambridge Neurotechnology Ltd, Papworth, UK) is a research grade multi-sensor device that utilises branched modelling techniques to estimate PAEE through the combination of accelerometer counts and HR (Brage et al, 2004). Combining HR and accelerometer devices have been used to accurately estimate PAEE during activities of low-to-moderate-intensity (Thompson et al, 2006) and treadmill walking / running in uninjured adults in a laboratory setting (Brage et al, 2005). Other multi-sensor devices used to predict EE include the SenseWear® Armband, a commercially available monitor that is designed to be worn on the upper arm. This device incorporates acceleration signals and physiological measures such as galvanic skin response and heat flux to estimate EE. More detail regarding componentary and specifications of the SenseWear® Armband can be found elsewhere (Chen et al, 2012). It is unclear whether positioning a multi-sensor on the upper arm is suitable location when attempting to predict ambulatory EE in prosthetic users (see table 2.3). Previously the sternum has been used (Bussmann et al, 2004; Bussmann et al, 2008; Selles et al, 2005) and anatomically, this is closer to the accelerometry placement of the AHR than the SenseWear Armband.

Wearable PA monitors like the Fitbit and Apple Watch are growing in popularity and provide an opportunity for large numbers of the public to self-monitor their own PA behaviours (Chowdhury et al, 2017). To date, only one study has used one of these widely available consumer devices in a group of individuals with LLA. Albert et al. (2015) performed a feasibility study monitoring daily function in persons with trans-femoral amputations using a Fitbit. They found that this monitor has the potential to be used to assess the PA levels of people with LLA. However, it is important to note that commercially available activity monitors such as the Fitbit or Apple Watch are yet to be validated in individuals with LLA, therefore the information they provide must be viewed with caution.

2.10.8. Summary

In order to enhance research and practice in this field, it is important to develop valid and reliable tools to estimate free-living PAEE in individuals with LLA. The development of tools specific to individuals with traumatic LLA would help both researchers and clinicians better understand the link between PA and metabolic health in this population. This is of primary importance in individuals with traumatic LLA, who exhibit lower levels of self-reported PA and an increased risk of chronic disease. To date, work in this area has focussed self-reported questionnaires and/or the use of step counts relating to different prosthetic componentry using uniaxial accelerometry (see table 2.3). The ability to accurately predict free-living PAEE in individuals with unilateral and/or bilateral LLA with modern day triaxial accelerometers has yet to be explored. To date, there are no published studies to determine the most appropriate anatomical placement of accelerometers to accurately predict PAEE in unilateral or bilateral amputees. Consequently, there are no peer-reviewed articles, which have attempted to develop population specific algorithms for the prediction of PAEE in lower-limb amputees. It is unclear whether the addition of a physiological variable (such as HR) improves the accuracy of these models against a criterion measure or whether inherited proprietary predictive algorithms of devices like the AHR can accurately predict ambulatory EE in individuals with traumatic LLA. These questions will form the bases of chapter 5 of this thesis.

2.11. SUMMARY OF LITERATURE REVIEW AND PLAN FOR THE FOLLOWING CHAPTERS

As stated in the introduction, strategies to increase physical function, whilst reducing the development of secondary health disorders in severely injured military personnel is a very important issue for UK DMS. Their primary concern relates to the premise that disability is known to negatively impact PA behaviour and that injured personnel with LLA are considered a population at risk of developing future cardiometabolic health conditions (pathways described in chapter 1 figure 1.1). Based on the available literature, individuals with traumatic LLA are predisposed to a range of long term secondary health conditions (section 2.3). The review then describes some of the functional challenges associated with LLA, which may explain the reported reductions in PA in this population (section 2.4). There is limited available literature detailing changes in body composition and cardiometabolic health following traumatic LLA. Unfortunately, in the studies that are available the short to long-term prognosis is not always positive.

Although informative, much of the available evidence used to support the findings stated above are not directly applicable or transferable to UK DMS, in particular UK military personnel with traumatic LLA currently receiving treatment for their injuries from the most recent conflicts in Iraq and Afghanistan. Many of the patients treated at DMRC with traumatic LLA survived injuries that would have once proved fatal. They therefore bring a unique rehabilitation challenge not previously seen before, making comparisons to older conflicts and civilian groups problematic. UK military personnel have access to advance prosthetic technology and receive residential IDT rehabilitation unique to DMS and therefore not comparable to any other amputee rehabilitation centres in the world. Military personnel with traumatic LLA are typically young, previously very active men, with a resilient and determined outlook. Much of the available evidence described in this literature review relies on older military conflicts using older veterans and/or civilian populations with LLA who have used their prosthetic for a considerably greater length of time. Much of the literature investigating PA in LLA cohorts used self-report techniques and/or outdated accelerometers not validated to assess PAEE in this population. The few papers that did objectively measure cardiometabolic health in individuals with LLA are now quite dated, use older participants and fail to utilise the more commonly used indices for predicting metabolic health (i.e. HOMA and ISI Matsuda following OGTT).

There are many gaps in the literature surrounding the topic of traumatic LLA rehabilitation, health and wellbeing in young serving military personnel currently engaged in rehabilitation. Despite having a combat casualty care pathway described as ‘exemplary’ by the Care Quality Comission, to date UK DMS have not published any data to determine the functional and mental health status of its traumatic LLA population injured on operation in Afghanistan or Iraq. Therefore, at this time DMS are unable to comment on how this unique group of severely injured adults have responded to their rehabilitation. It is unclear if UK military personnel attain clinical outcomes comparable to other militaries with LLA or levels of function and psychosocial health comparable to the general population and therefore indicative of integration back into society. This topic will be explored in chapter 4.

This review details the importance of PA to promote long-term health and wellbeing in individuals with LLA (particularly cardiometabolic health). Despite the growing area of wearable technology and PA monitoring in surveillance research, no study has attempted to validate a PA monitor to predict PAEE in individuals with LLA. Before using PA monitors during free-living environments it is important to first ensure the methods used are accurate and valid for the population of interest. Chapter 5 will investigate the validity of a commonly used triaxial accelerometer and multi-sensor methods to estimate PAEE in UK military personnel with traumatic LLA. Once validated, these PA monitors will be used to assess the difference in ambulatory EE in two different living environments; (i) in-patient rehabilitation at DMRC and (ii) recovery at home (see chapter 6). This will be the first time population specific models are validated to predict ambulatory EE will be used in individuals with traumatic unilateral and bilateral LLA. Their PA data will also be compared to an active normative control group to see how previously active military personnel with LLA now compare to their peers.

The review details unfavourable changes in body composition (i.e., obesity) and elevated risks associated with cardiovascular disease and metabolic disorders following traumatic LLA. The majority of these studies are based on long term follow-up and older cohorts, therefore their results are not easily transferable to DMS and patients still engaged in intensive exercise based rehabilitation. As part of the same longitudinal study as chapter 6, chapter 7 will investigate changes in clinical outcomes, body composition and cardiometabolic health over a 20 week duration in unilateral and bilateral LLA nearing the end of their rehabilitation care pathway at DMRC.

Before addressing the research areas outlined above it is useful to provide some detail of the DMS combat casualty care pathway. Chapter 3 therefore describes the journey from point of injury to last admission to DMRC Headley Court. This will also provide an overview of the heterogeneous injury profiles of the participants with LLA who will be investigated in the later chapters of this thesis.

CHAPTER 3. UK DEFENCE MEDICAL SERVICES COMBAT CASAULTY CARE PATHWAY

This chapter describes the UK combat casualty care pathway (i.e. ‘methods’ used by DMS) from point of injury to final rehabilitation admission at DMRC Headley Court for severely injured UK military personnel (i.e., individuals with blast-based traumatic LLA). The chapter documents the traditional journey that UK military personnel injured during combat in Iraq/Afghanistan would experience and aims to provide greater clarity on the challenges faced by the patient, surgical and rehabilitation teams along the pathway. The secondary objective is to provide the reader with a greater understanding of the unique heterogeneous nature of the participant groups who later enrolled in the studies described in future chapters of this thesis.

3.1. BLAST INJURY DEMOGRAPHICS AND PATTERNS OF INJURY

Specific injuries are determined by the energy level of the blast and the individual’s protective equipment (Cannon et al, 2016). Blast injuries sustained by casualties outside a vehicle are termed ‘dismounted’ those sustained in a vehicle are termed ‘mounted’ injuries. Dismounted events are classified into two categories; low and high energy blast. Low-energy blast events are when the explosion occurs at a significant distance from the individual or from a relatively small explosive device. Common injuries may include relatively minor wounds to the extremities and perineal soft tissues (Cannon et al, 2016). High-energy blasts from explosions in close proximity to dismounted personnel can result in traumatic LLA, severe pelvic fractures, open fractures or amputations to the upper extremity and destructive injuries to the perineal soft tissues (Andersen et al, 2012; Fleming et al, 2012; Mamczak & Elster, 2012). Despite protection from heavy armour incorporated into military vehicles, severe injuries still occur during mounted blast events, including traumatic brain injuries, blunt thoracic injuries, vertebral fractures, long bone extremity fractures and comminuted calcaneal fractures (Cannon et al, 2016; Possley et al, 2012). Severe limb trauma in military personnel is now considered the signature combat injury from the Afghanistan conflict and the most common cause of disabling conditions leading to medical retirement (Cross et al, 2011). Table 3.1 describes the characteristics and types of injuries that may occur from the four categories of blast injury. Figure 3.1 demonstrates how these different mechanisms of injury can occur following an explosion (i.e., from an improvised explosive device - IED).

Table 3.1: Blast Injury Demographics and Patterns of Injury

Category	Characteristics	Body Part Effected	Types of Injury
Primary	Interaction of the blast wave with the human body. The blast wave quickly exerts over-pressure outwardly from the centre of the blast and rapidly followed by under-pressure. The abrupt pressure change stretches then contracts human tissue beyond normal limits.	Gas-filled organs (lungs, GI-tract and middle ear) are most effected	Barotrauma (pressure injury) • Blast Lung (pulmonary barotrauma) • TM rupture and middle ear damage • Abdominal haemorrhage and perforation • Globe eye rupture • Blast induced m-TBI
Secondary	Bomb fragments and other projectiles energised by the explosion (fragmentation) causing penetrating or non-penetrating wounds.	Any body part maybe effected. But usually unprotected body regions not covered with armour (i.e. extremities, buttocks, groin, neck and face)	Penetrating ballistic (fragmentation) or blunt trauma • Mangled limbs • Immediate amputation • Haemorrhage • Severe vascular and soft-tissue injury • Heavy contamination and wound infection risk
Tertiary	Displacement of the body (individuals being thrown by the blast) or structural collapse of buildings (causing crush injuries)	Any body part maybe effected	Blunt (crush) based injuries • Fractures and soft tissue injury • Head injury, • TBI
Quaternary	All explosive-related injuries, illness or diseases not directly due to the blast mechanism	Any body part maybe effected	• Burns • Asthma, COPD or other respiratory disorders from dust smoke or toxic fumes • Blunt trauma from nearby building collapse

Abbreviation: GI = Gastrointestinal, TM = Tympanic Membrane, m-TBI = mild-Traumatic Brain Injury, COPD = Chronic Obstructive Pulmonary Disorder. Adapted from (Horrocks, 2001), (Fleming et al, 2012) and (Stevenson, 2009).

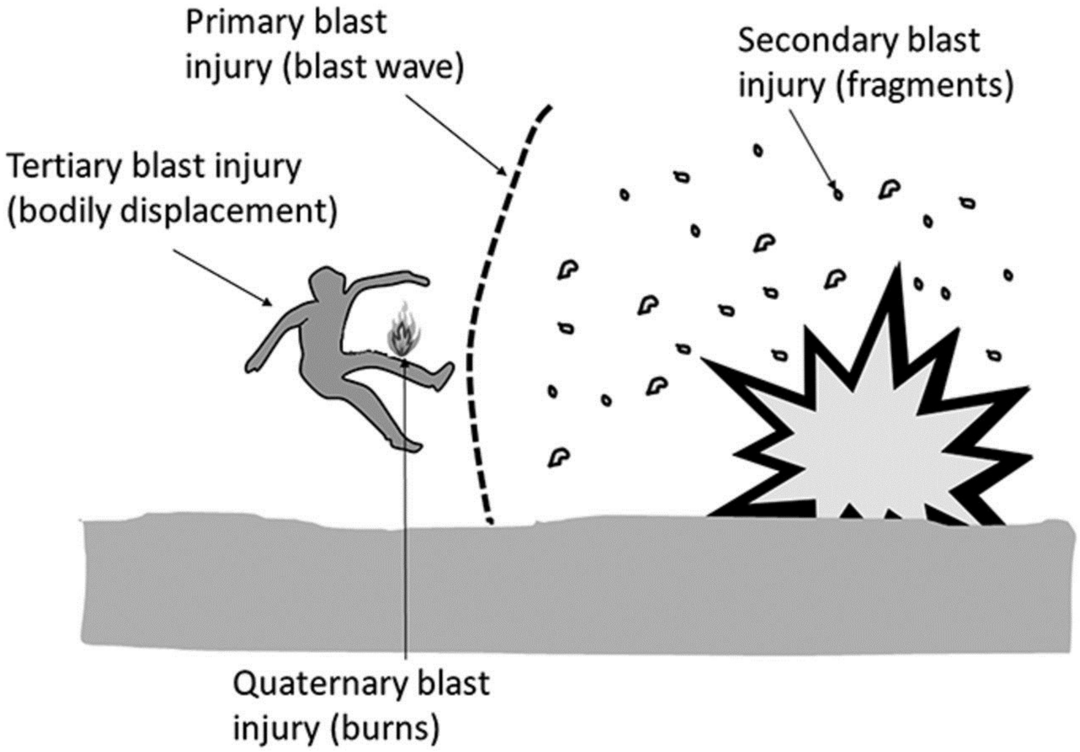


Figure 3.1: Mechanisms of injury from explosions. Image taken from (Ramasamy et al, 2013)

3.2. COMBAT CASUALTIES FROM THE CONFLICTS IN IRAQ AND AFGHANISTAN

The numbers of severely injured military personnel was recently at its highest during and after the conflicts in Iraq and Afghanistan. Between 2003 and 2012 the number of UK military servicemen wounded in action was 2,184 (Penn-Barwell et al, 2015), in Afghanistan 265 individuals sustained 416 amputations (Edwards et al, 2015). Although deeply traumatic to each casualty, their family and serving colleagues, each military conflict often brings great medical advances in battle care emergency medicine that enables military personnel to survive injuries that would have once proved fatal (Gulland, 2008). The most recent conflicts in Iraq and Afghanistan proved no different. Penn-Barwell et al (2015) estimated the Injury Severity Score values associated with a 50% probability of fatality for each year between 2003 and 2012. They found a 50% survival injury severity score value rose from 32.5 in 2003 to 59.6 in 2012 (using the ‘new injury severity score’ (NISS) outcome measure). Supporting the observation that patients were surviving injured that would have once proved fatal. NISS records the 3 most severely injured body parts and scores each of them out of 25, with higher scores signifying greater severity of injury. Injury severity is scored from 1 to 75 with “Major trauma” defined as > 15 (Russell et al, 2011). Subsequently, this created a relatively large population of young servicemen and women with exceptional long-term healthcare and rehabilitation needs not seen or treated in the UK or globally before.

In an effort to grasp a better appreciation of the unique challenges facing this combat injured population, it is important to understand the mechanisms/patterns of injury elicited and the combat casualty care pathway that maintained their survival and ability to integrate back into the community.

3.2.1. Battlefield Trauma Care: First Responders

When a UK Serviceman or woman is injured in combat (for example, the recent conflicts in Iraq or Afghanistan), immediate first aid is administered at the point of wounding from other members of the combat team (often a fellow infantry soldier). The most common cause of death from dismounted combat blast injury is catastrophic haemorrhage (Clasper & Ramasamy, 2013; Eastridge et al, 2012). Therefore, the highest priority on the battlefield is haemorrhage control (Jansen et al, 2012). Haemorrhage control bandages made of a positively charged material that attracts negatively charged red blood cells and rapidly stops blood flow and a tourniquet are immediately applied at the point of injury by a fellow soldier (Gulland, 2008). The casualty will often be evacuated via helicopter carrying a crew of emergency medics, one of whom will either be a consultant anaesthetist or a consultant emergency specialist (Gulland, 2008). The prehospital deployment of physicians provides advanced resuscitation techniques and senior decision making at the point of wounding and during transport (Penn-Barwell et al, 2015). The casualty is evacuated to the Role 3 British Military Field Hospital in Camp Bastion, Afghanistan or Basra in Iraq.

3.2.2. Field Hospital in Iraq / Afghanistan

On arrival at the British Military Field Hospital, patients typically have multiple tourniquets and if necessary a pelvic binder in place and ‘initial stabilization’ of the casualty is performed (Cannon et al, 2016). The principles of war surgery: arrest of haemorrhage; thorough wound debridement (cleaning or washout and removal of dead or heavily contaminated tissue); temporary mechanical stabilization of fractures (i.e. external fixators); removal of contamination and foreign bodies; copious wound lavage and administration of antibiotics are immediately delivered (Evriviades et al, 2011).

The casualty will receive a thorough examination of their entire body and receive appropriate radiological imaging (such as X-rays and computed tomography scans) to diagnose all injuries (Dharm-Datta & McLenaghan, 2013). Blast-related traumatic limb-loss is initially managed with completion of the amputation using a length-preserving technique (Cannon et al, 2016). Dismounted blasts often result in highly unstable pelvic fractures. In a dismounted combat blast injury with lower-extremity wounds, the probability of concomitant pelvic fracture increases significantly with more proximal injuries (Cannon et al, 2016). Individuals with bilateral transfemoral amputations have a nearly 40% incidence of an associated pelvic fracture (Mossadegh et al, 2012). Fractures of the non-dominant upper extremity are also common in these patients, due to the forward stance or "low-ready" position used by most service members on dismounted patrol (Andersen et al, 2012). More than 80% of such fractures are open and severely contaminated (Cannon et al, 2016). Temporary mechanical stabilisation of fractures, using external fixators device are commonly used (see figure 3.2). Wounds are left open at this stage as they are contaminated with bacteria and will become infected if closed too early (Dharm-Datta & McLenaghan, 2013).

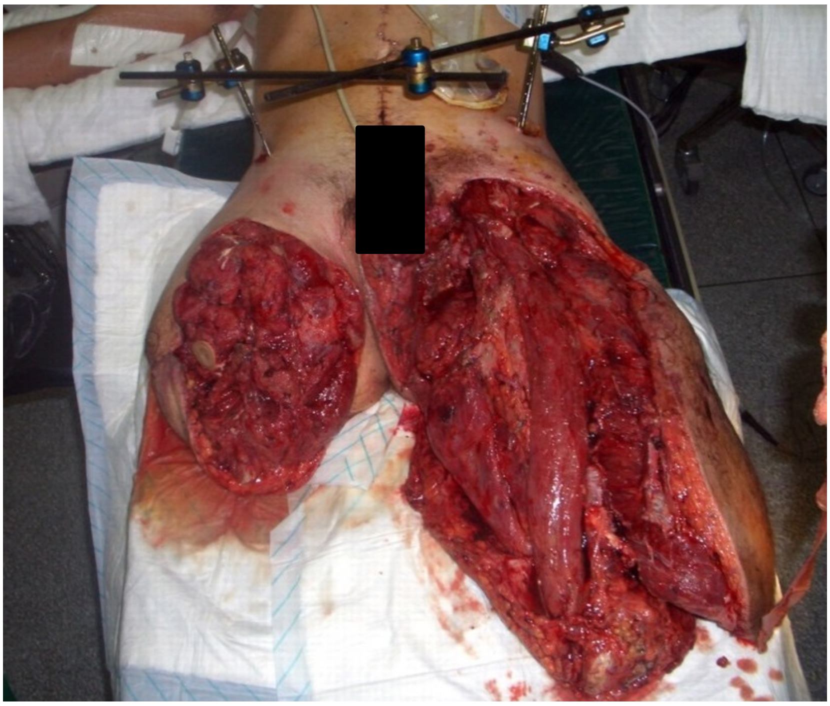


Figure 3.2: A typical presentation of an IED blast. Bilateral trans-femoral amputation with inadequate skin coverage, external fixator to pelvis, shrapnel wounds across upper limbs attached. Taken from, Evriviades et al. (2011).

3.2.3. Common Complications / Challenges Associated with Trauma Surgery

The greatest challenges or complications confronting the surgeon include haemorrhage control, infections, venous thromboembolic events (including deep venous thrombosis and pulmonary embolism), hypothermic coagulopathy and managing multiple casualties at one time (Cannon et al, 2016; Stevenson, 2009). In combat casualties with severe open fractures, bacterial infection occurs in 24% to 40% (Brown et al, 2010), and either bacterial or fungal infections occur in up to 63% of those injured in an explosion (Mossadegh et al, 2012). Blast-based weaponry (like an IED) can create a considerable number of casualties for first responders to manage and the surgical team back at the field hospital.

Despite victims of trauma frequently survive the initial insult owing to improvements in the control of haemorrhage, rapid volume replacement and haemostasis, the danger is not over once bleeding has been arrested and blood pressure restored. Two-thirds of patients who die following major trauma now do so as a result of causes other than exsanguination. Trauma evokes a systemic reaction that includes an acute, non-specific, immune response (Lord et al, 2014). A systemic inflammatory response to trauma is required for tissue repair and optimises the healing potential of an organism. In uncomplicated trauma patients the systemic inflammatory response is temporary, predictable and well balanced between pro- and anti-inflammatory mediators (Brochner & Toft, 2009). Following severe major trauma the systemic response involves interactions across the haemostatic, inflammatory, endocrine, and neurological systems. Endothelium activated by exposure to inflammatory cytokines becomes more porous, allowing mediators of tissue damage to gain access to the intercellular space (Lord et al, 2014). If the casualty is exposed to severe major trauma an initial exaggerated pro-inflammatory response may be observed. If the pro-inflammatory response is greater than the anti-inflammatory, the patient is at risk of the systemic inflammatory response syndrome (SIRS). If the anti-inflammatory response is greater, the immune system is suppressed thereby exacerbating the initial organ damage caused by shock, but also reduces the body’s ability to fight infection; this process can lead to an increased risk of sepsis, which, in turn, triggers a further vicious cycle of inflammation, immune-paralysis, and infection (Lord et al, 2014)). In most cases, pro-inflammatory activation predominates in the first 36 hours, followed by moderate immune suppression for the next few days (Gutierrez et al, 2011). Following trauma and uncomplicated surgery, the inflammatory response is usually normalised within 3 weeks (Brochner & Toft, 2009). It would therefore be anticipated that any residual inflammation response unique to the severe traumatic injury would eventually subside and any consequential increases in inflammation may be a reflect subsequent revision surgeries, acute infections or a result of long-term unfavourable lifestyle behaviours leading to chronic health conditions such as cardiovascular disease.

3.3. ROYAL CENTRE FOR DEFENCE MEDICINE AT QUEEN ELIZABETH HOSPITAL, BIRMINGHAM

Once stabilised and the immediate threat to life has subsided, all casualties receive aeromedical transfer to Queen Elizabeth Hospital Birmingham for ‘definitive’ surgical treatment and care (Dharm-Datta & McLenaghan, 2013). The Queen Elizabeth Hospital includes the Royal Centre for Defence Medicine, where military personnel complement the NHS therapy services provision.

Patients often arrive at includes the Royal Centre for Defence Medicine within 48 hours of wounding (Evriviades et al, 2011). In terms of 'definitive' management, open wounds will require several inspections and debridements as necessary, followed by delayed closure at a later date with fractured bones treated with either internal or external fixation methods (Dharm-Datta & McLenaghan, 2013).

Robinson and colleagues summarise the aims of surgical interventions when reconstructing the residual limb. Advice from the rehabilitation consultant or their clinical team is often sought at this stage as the decisions made and implemented by the surgeon has large consequences for prosthetic fitting and physical rehabilitation. To optimise short to long-term clinical outcomes of the patient and minimise the likelihood of additional surgery in the future, the surgeon will endeavour to (Robinson et al, 2010):

- Reduce the risk of developing pressure ulcers.
- During prosthetic use, ensure that scars are mobile and located away from areas vulnerable to shear damage (due to pressure or friction).
- To facilitate appropriate force generation and movement control of the residual limb, ensure muscles of the residual limb are secure to the periosteum (i.e. move the prosthesis in the desired way).
- Shape muscles and the overall residual limb such that the patient can safely and effectively use their prostheses. Sufficient amounts of muscle should remain to act as a protective cushion against pressure ulcers over bony prominences. A bulbous residual limb can make prosthetic fitting very challenging.
- To maintain correct anatomical alignment of the prosthesis (in patients with trans-femoral amputations), the femur should be positioned into its normal anatomical alignment before suturing the remaining adductor and abductor muscles to its periosteum or bone.
- To reduce neuropathic pain during prosthetic limb use, bury nerve ends such that when a neuroma forms as the nerve attempts to regenerate, this is not subjected to contact or pressure.

Early rehabilitation focuses on regaining a functional level for basic tasks, for example, bed mobility, transfers from bed to wheelchair, eating, drinking and washing (Pope et al, 2017). Early rehabilitation interventions have previously demonstrated reduced length of stay in hospital, reduce mortality (McWilliams et al, 2009; Morris et al, 2008) and improve long-term functional outcomes (McWilliams et al, 2018). Therapists at Queen Elizabeth Hospital were able to replicate these findings after introducing an 'early rehabilitation team' in 2012 (McWilliams et al, 2015). Examples of their practice include, early (sometimes while ventilated) physiotherapy-led mobilisation, comprising sitting on the edge of the bed, sitting out and walking. Gym attendance for individuals with severe traumatic injury (such as LLA) is encouraged in the critical care setting in reducing the well documented deleterious effects of prolonged bed-rest (Brower, 2009).

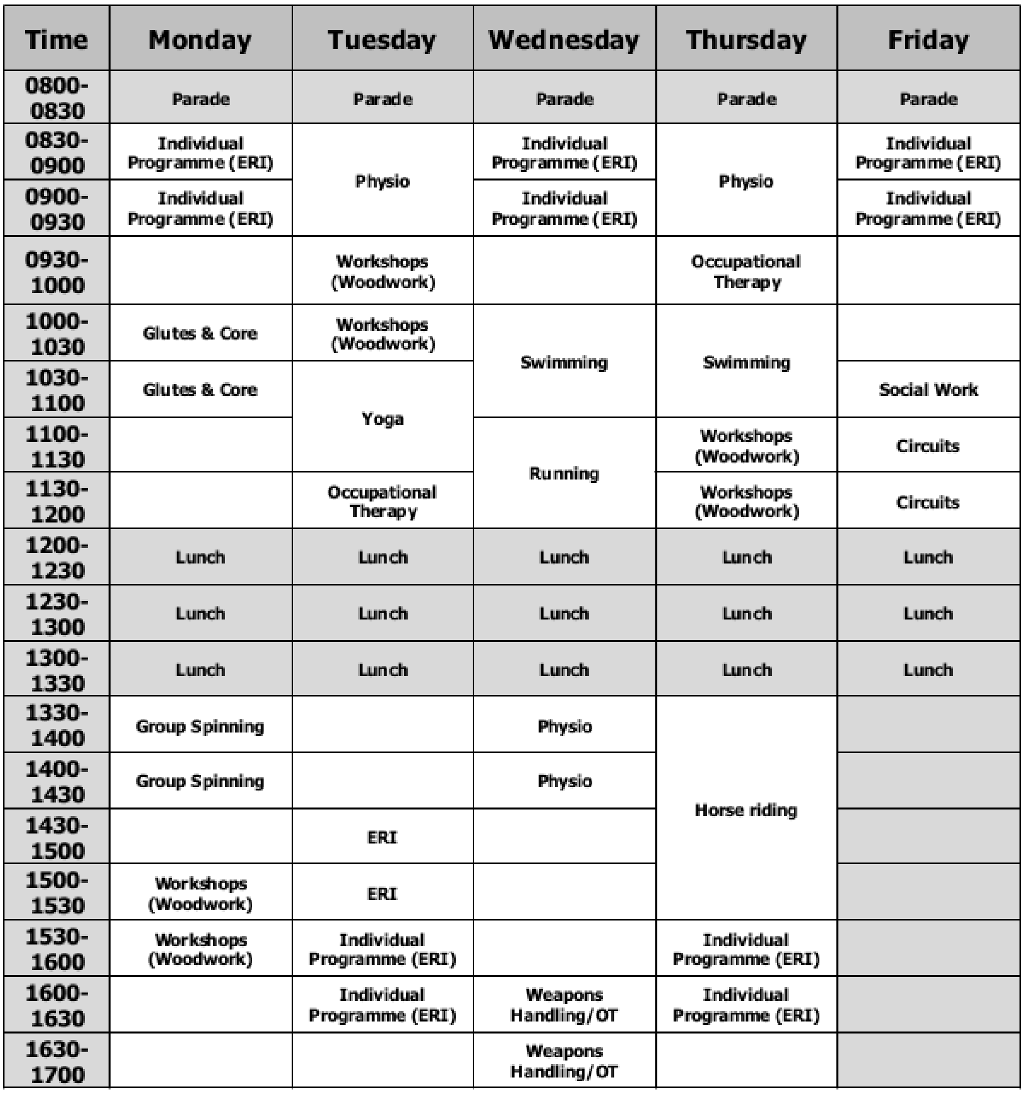
Enhanced exercise therapy includes the use of an over-bed bike, provision of exercise bands and adjustable dumbbells by the intensive care bed (Pope et al, 2017). IDT rehabilitation is delivered at Queen Elizabeth Hospital until the patient with amputation is considered clinically appropriate for transfer to the DMRC Headley Court. Timelines associated with length of stay are determined on a case-by-case basis, however, a proactive relationship between Royal Centre for Defence Medicine and DMRC ensure joined-up care throughout the rehabilitation pathway.

3.4. REHABILITATION CARE AT DEFENCE MEDICAL REHABILITATION CENTRE (DMRC), HEADLEY COURT

All individuals with amputation/s are referred directly to the complex trauma department at DMRC Headley Court for in-patient rehabilitation admissions and subsequently referred to the prosthetic limb fitting centre. Rehabilitation of the combat casualty requires the integration of the IDT working in a partnership with the patient to optimally address the challenges associated with each individual's short-to-long term surgical and medical needs. This is considered a collective effort to meet the high expectations of patients, families, government, and society.

> *'Rehabilitation is a process of assessment, treatment and management by which the individual (and their family and carers) are supported to achieve their maximum potential for physical, cognitive, social and psychological function, participation in society and quality of living.'* - British Society of Rehabilitation Medicine (BSRM) and NHS England.

The complex trauma team at DMRC is a consultant-led interdisciplinary department including medics (consultants, registrars and Senior House Officers), exercise rehabilitation instructors (ERI), physiotherapists, occupational therapists, prosthetists, orthotists, nurses, social workers, podiatrists, psychologists and rehabilitation assistants. In-patient admissions are designed to meet the individual needs of the patient. During the early stages of rehabilitation, each combat injured amputee will have 6 week admissions at a time (or longer if required), with blocks of 'recovery leave' lasting approximately 2 weeks. As rehabilitation progresses the length of in-patient admissions decrease to 4 weeks (2 weeks if the patient only requires minimal input from the rehabilitation team) and the length of recovery time can increase to 6 weeks. Recovery leave can be used for rest at home or return to work depending on an individual's requirement and stage in their rehabilitation. A typical in-patient admission will include 5 to 7 hours per day of IDT rehabilitation, 5 days a week (Sherman et al, 2019). This includes approximately 1 hour of daily physiotherapy (manual therapy and training with the prosthetic); 2 to 3 hours of daily exercise therapy with the ERI (group and one to-one training sessions); occupational therapy to facilitate adaptation of, and training in, ADL and supporting integration into society by promoting socialisation and vocational support; social work services and mental health support. Figure 3.3 provides an example of a weekly timetable for an amputee patient at DMRC, and later in this chapter, figure 3.8 describes how this differs from civilian care in the NHS.



Time	Monday	Tuesday	Wednesday	Thursday	Friday
0800-0830	Parade	Parade	Parade	Parade	Parade
0830-0900	Individual Programme (ERI)	Physio	Individual Programme (ERI)	Physio	Individual Programme (ERI)
0900-0930	Individual Programme (ERI)		Individual Programme (ERI)		Individual Programme (ERI)
0930-1000		Workshops (Woodwork)		Occupational Therapy	
1000-1030	Glutes & Core	Workshops (Woodwork)	Swimming	Swimming	
1030-1100	Glutes & Core	Yoga			Social Work
1100-1130			Running	Workshops (Woodwork)	Circuits
1130-1200		Occupational Therapy		Workshops (Woodwork)	Circuits
1200-1230	Lunch	Lunch	Lunch	Lunch	Lunch
1230-1300	Lunch	Lunch	Lunch	Lunch	Lunch
1300-1330	Lunch	Lunch	Lunch	Lunch	Lunch
1330-1400	Group Spinning		Physio	Horse riding	
1400-1430	Group Spinning		Physio		
1430-1500		ERI			
1500-1530	Workshops (Woodwork)	ERI			
1530-1600	Workshops (Woodwork)	Individual Programme (ERI)		Individual Programme (ERI)	
1600-1630		Individual Programme (ERI)	Weapons Handling/OT	Individual Programme (ERI)	
1630-1700			Weapons Handling/OT		

Figure 3.3: An example of a weekly timetable for an individual with lower-limb amputation admitted to the complex trauma department at DMRC Headley Court

Education on the importance of general health continues throughout rehabilitation care at DMRC Headley Court. Common topics will include the evaluation of skin integrity and wound healing through dressing changes, oedema control, stump maturation and protection, as well as skin/scar mobilisation, pain management (pharmacological and non-pharmacological), the cessation of smoking (if indicated), healthy nutrition practices, and adaptations to promote early ADL. Among the earliest exercise based education topics include flexibility training to reduce/prevent contractures at the hip or knee from developing. Should significant joint contractures develop, they can create difficulty with socket fitting and prosthetic alignment and in some cases result in surgical interventions (Pasquina et al, 2014). Heterotopic ossification represents a significant challenge in the long-term management of combat-wounded patients and can occur in up to 63% of combat-related amputations, and is associated with both blast injuries and final amputation level (Potter et al, 2010).

Figure 3.4: Image of a bilateral amputee who recently arrived at DMRC Headley Court. The image provides an illustration of the range of medication, skin grafting, pelvic fixator and wheelchair requirements to help mobilise during the early phases of rehabilitation.

3.4.1. Exercise-Based Rehabilitation Prior to Prosthetic Fitting

Upon arriving at DMRC, many individuals with amputation have experienced prolonged bed rest, resulting in considerable muscles weakness and atrophy. A comprehensive evaluation of motor, sensory, cardiovascular, and cognitive function will be performed by the IDT. The physiotherapist and ERI will immediately implement an integrated strength and conditioning program designed to prepare the patients for the rehabilitation challenges that lay ahead. This will include upper body strengthening exercises to assist with supporting the body for transfers, propelling a wheelchair or the use of walking aids. It is known that most lower-limb prosthetic devices are ‘‘passive’’ by design (the power needed for mobility must be generated from the remaining musculature), therefore strengthening of the lower-limb musculature and lumbo-pelvic hip complex can be very important to promote early successful prosthetic use (Pasquina et al, 2014). It is also imperative that aerobic conditioning be initiated in the first in-patient admission. Amputees must improve their aerobic fitness levels to meet the increased energy demands associated with prosthetic ambulation (Bussmann et al, 2008).

3.4.2. Prosthetic Limb Fitting

Initial prosthetic fitting will focus on the production of a socket that accommodates both the shape and size of the residual limb. However, during the early stages of rehabilitation following LLA, significant changes in residual limb volume are anticipated due to oedema, fluid shifts, and muscle atrophy (Fitzpatrick & Pasquina, 2010) (see figure 3.4) As a result, multiple revisions to the initial socket are expected. Once limb volume and shape have stabilised and a satisfactory socket design has been attained, final sockets can be fabricated and fitted.

The socket and its interface with the residual limb are crucial to successful prosthetic fitting. Sockets are designed to provide total-surface weight bearing in combination with a design that relieves pressure from pressure-sensitive anatomical structures such as bony prominences (Robinson et al, 2010). If the socket is not comfortable, the patient will often refuse to participate in walking based activities. Individuals with transtibial amputations have areas of bony prominence (the tibia) which need protection, this is usually achieved by wearing a sock (Fitzpatrick & Pasquina, 2010). Due to good soft tissue coverage, individuals with transfemoral amputations do not have the same problems as transtibial; the main concerns with this group of individuals are usually effective transmissions of forces, stabilisation and allowance of muscle function (Robinson et al, 2010). Individuals with knee disarticulation amputations have the advantage that weight can be taken through the distal end of the remaining bone for near normal force transmission, however the distal displacement of the prosthetic joint and bulk of the bony prominence result in restriction on the choice of prosthetic components and a less cosmetically appealing limb for many patients (Robinson et al, 2010).

One of the complexities associated with blast trauma is significant scar tissue that forms and short residual limbs in individuals with transfemoral amputation (see figure 3.5). Due to reduced and disturbed skin surface area interface with the socket, this can often lead to ill-fitted prosthetic and difficulty with ambulation leaving many transfemoral amputees preferring to mobilise in a wheelchair.

Figure 3.5: Image demonstrating the scar tissue on the residual limb after a blast-based trauma

3.4.3. Prosthetic Training

The aim of prosthetic gait training at DMRC is to achieve maximum independence, safely, with minimum additional EE. Incorporating weight bearing activity is dependent upon the healing of wounds, a significant decrease in post-operative oedema and a stable soft tissue envelope that can tolerate prosthetic use (Tintle et al, 2010). Weight bearing with a prosthesis can typically be initiated at 6 weeks depending on additional proximal fractures (i.e. the pelvis) or any planned soft tissue reconstructions, such as myocutaneous or free flap reconstruction, which may further delay prosthetic fitting (Hoyt et al, 2015).

The primary goal is to achieve sufficient muscular force and endurance from the lower extremities to support prolonged periods of standing, walking and performing ADL. Muscular strength and endurance training during prosthetic gait training may become more demanding. Under supervision of the physiotherapist, prosthetist and ERI, gait training begins with weight-bearing and weight-shifting activities (with the parallel bars used for upper-limb support). This is followed by gradually walking back and forth between the parallel bars (see figure 3.6). As patients establish a consistent gait pattern and maintain good technique, they will usually advance from the parallel bars to two crutches and then to unilateral support (when indicated by their physiotherapist).

[redacted] [redacted] [redacted]

Figure 3.6: Image of a triple amputee using the parallel bars whilst performing prosthetic gait training

Early prosthetic training for individuals with bilateral transfemoral amputations is usually initiated with short bilateral prostheses (see figure 3.7). The reduced height lowers the patient's centre of mass, making it easier to stand and walk. Falling and learning how to return to a standing position are important aspects of the gait retraining journey, but easier to learn with shorter prosthetics to begin with. As the patient demonstrates improved stability and ambulation, prosthetic knee components can be added along with height (Pasquina et al, 2014). As the patient demonstrates appropriate balance, ability to recover from falls and improved gait, they are then advanced to stairs, ramps and obstacles and eventually taught how to return to running or other higher level activities (Highsmith et al, 2016). Throughout these progressive activities the physiotherapist and ERI will continue to prescribe personalised muscle exercise programmes to prevent and correct any gait deviations.


[redacted]

Figure 3.7: Image of a bilateral amputee wearing short prosthesis to lower centre of mass

Participation in sport is a traditional part of military life. Many individuals with amputation will view successful rehabilitation as the ability to perform activities they enjoyed prior to injury. Adaptive sports and recreational programs run by charities, such as Battleback, facilitate the engagement of injured military personnel participating in sport. Commonly delivered programmes at DMRC include archery, basketball, hand-cycling, rock climbing, water sports, horse riding and trips away skiing and parachute jumping. These programmes form an integral part of the rehabilitation delivered at DMRC. The physical abilities of some severely injured military personnel have gained international exposure via the Paralympic Games and more recently exemplified by the introduction of the Invictus Games. For many injured UK military servicemen their first introduction to competitive adaptive sports would have been during their in-patient admissions at DMRC.

3.4.4. Integration into Daily Life

It is important to note that not all rehabilitation is exercise based. DMRC have a full time mental health support service and social workers promoting positive psychosocial health outcomes. Due to the severity of injury, many individuals with LLA are unable to return to their previous physically demanding job (i.e., infantry soldier), therefore many patients consider a different line of employment within or outside of the military. Vocational rehabilitation is an important programme delivered at DMRC to encourage independence and successful community reintegration. Occupational therapists introduce specialised adaptive equipment or assistive technology to promote independent living. At DMRC this includes using assets on the unit (including a simulated home and kitchen environment) where the occupational therapist and physiotherapist can work closely with the patient to instrumentally master ADL in the home. This process helps to determine what adaptations might be required around the patient's home to assist with ADL (i.e. access to the shower/bath, and/or making the patient's home more wheelchair friendly). Occupational therapists will also lead efforts on educating patients on driving an adapted motor vehicle and improve confidence in using public transportation and shopping at the supermarket.

Mental health support advisors offer a broader range of services to injured military personnel. Whilst assisting with the patients coming to terms with their injury, they also provide counselling for PTSD, anxiety, depression, alcohol dependency and relationship advice.

3.5. THE CARE PATHWAY FOR UK MILITARY PERSONNEL WITH AN ELECTIVE LOWER-LIMB AMPUTATION

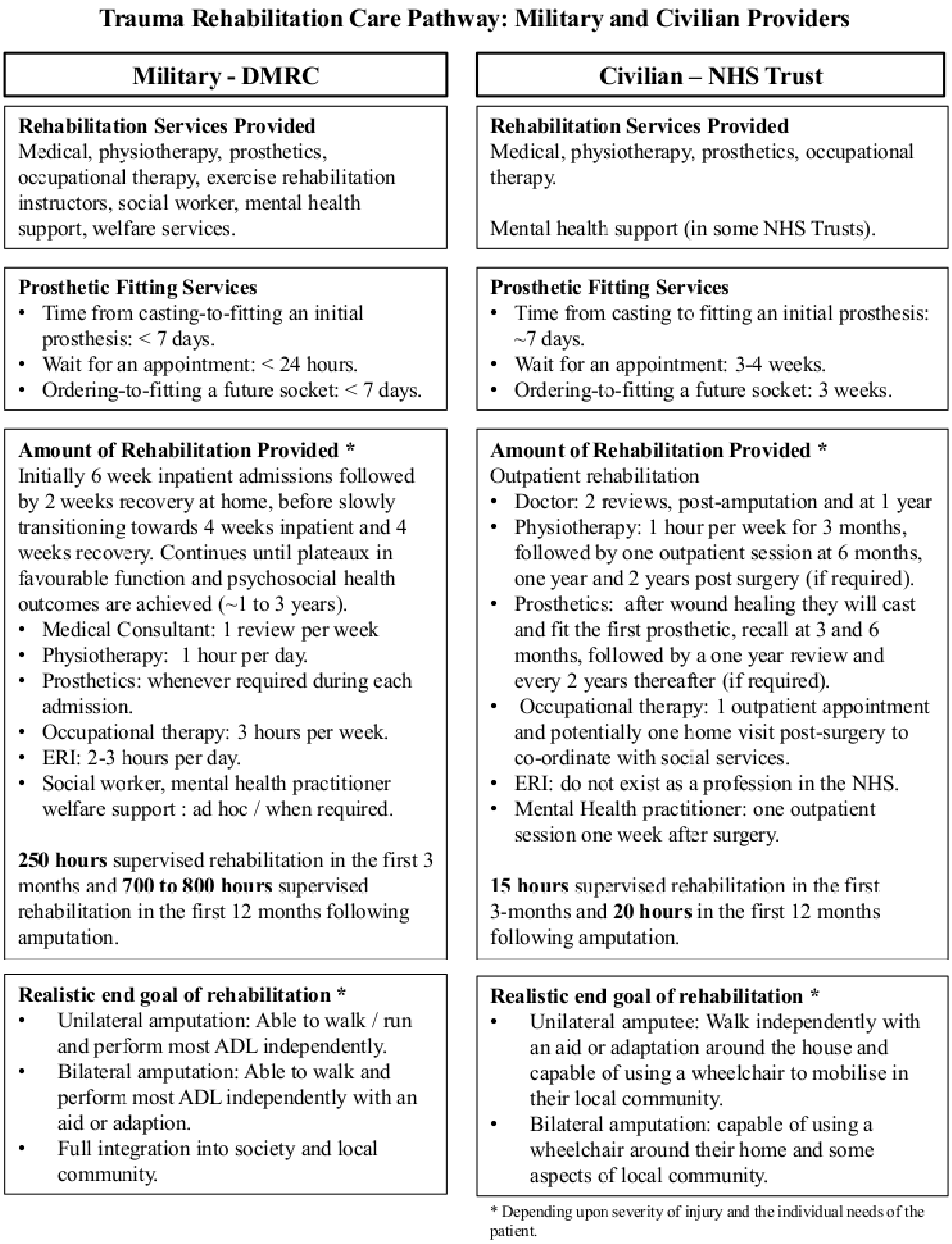
Among the most common reasons for patients electing for LLA in a previously injured and salvaged limb include unsatisfactory levels of pain and physical function. UK military personnel electing for amputation after prolonged limb salvage typically attend a mean of 8 in-patient admissions over 18 months prior to their amputation (Ladlow et al, 2016). Rehabilitation is delivered at the same complex trauma department at DMRC, supervised by the same clinicians, using the same paradigm of treatment. The only difference being, prior to amputation, these individuals would not have attended the prosthetic department. Once a patient is readmitted to DMRC as an amputee, they receive the same rehabilitative care and treatment as described above.

3.6. HOW DO UK MILITARY AND CIVILIAN CARE PATHWAYS COMPARE?

In Gulland's (2008) paper titled "Lessons from the Battlefield," Professor Porter reflects on the most important lessons learnt from battlefield emergency medicine that the UK NHS could learn from:

"The difference with the military model is the high level of consultant input right from the word go. In the UK (NHS) most of that care would be done by juniors."

In the UK military, this consultant-led service continues throughout rehabilitation. Figure 3.8 provides a summary of some the main differences between military (DMRC, Headley Court) and civilian (NHS) trauma care pathways (Nottingham NHS Trust). Each NHS trust and prosthetic contract is different, however it is widely acknowledged that the greatest difference between rehabilitation care in the military and NHS is the waiting time for an appointment to see a prosthetist (and have a socket replaced), the number of professions within the IDT and the amount of contact time they spend with the patient. This intuitively leads to very different outcomes at the end of rehabilitation. Edwards et al. (2016) found that despite a significantly greater severity of injury, military personnel demonstrate far greater mobility grades after rehabilitation. The proportion of patients able to 'walk independent of walking aids except occasionally for confidence or to improve confidence in adverse terrain or weather' was 91% in UK military and 19% in civilian patients with LLA, respectively. The ability to 'walk on uneven ground > 50 metres, with or without use of walking aids' was 95% and 58% in UK military and civilian patients with amputation, respectively (Edwards et al, 2016).



Trauma Rehabilitation Care Pathway: Military and Civilian Providers

Military - DMRC	Civilian – NHS Trust
Rehabilitation Services Provided Medical, physiotherapy, prosthetics, occupational therapy, exercise rehabilitation instructors, social worker, mental health support, welfare services.	**Rehabilitation Services Provided** Medical, physiotherapy, prosthetics, occupational therapy. Mental health support (in some NHS Trusts).
Prosthetic Fitting Services • Time from casting-to-fitting an initial prosthesis: < 7 days. • Wait for an appointment: < 24 hours. • Ordering-to-fitting a future socket: < 7 days.	**Prosthetic Fitting Services** • Time from casting to fitting an initial prosthesis: ~7 days. • Wait for an appointment: 3-4 weeks. • Ordering-to-fitting a future socket: 3 weeks.
Amount of Rehabilitation Provided * Initially 6 week inpatient admissions followed by 2 weeks recovery at home, before slowly transitioning towards 4 weeks inpatient and 4 weeks recovery. Continues until plateaux in favourable function and psychosocial health outcomes are achieved (~1 to 3 years). • Medical Consultant: 1 review per week • Physiotherapy: 1 hour per day. • Prosthetics: whenever required during each admission. • Occupational therapy: 3 hours per week. • ERI: 2-3 hours per day. • Social worker, mental health practitioner welfare support : ad hoc / when required. **250 hours** supervised rehabilitation in the first 3 months and **700 to 800 hours** supervised rehabilitation in the first 12 months following amputation.	**Amount of Rehabilitation Provided *** Outpatient rehabilitation • Doctor: 2 reviews, post-amputation and at 1 year • Physiotherapy: 1 hour per week for 3 months, followed by one outpatient session at 6 months, one year and 2 years post surgery (if required). • Prosthetics: after wound healing they will cast and fit the first prosthetic, recall at 3 and 6 months, followed by a one year review and every 2 years thereafter (if required). • Occupational therapy: 1 outpatient appointment and potentially one home visit post-surgery to co-ordinate with social services. • ERI: do not exist as a profession in the NHS. • Mental Health practitioner: one outpatient session one week after surgery. **15 hours** supervised rehabilitation in the first 3-months and **20 hours** in the first 12 months following amputation.
Realistic end goal of rehabilitation * • Unilateral amputation: Able to walk / run and perform most ADL independently. • Bilateral amputation: Able to walk and perform most ADL independently with an aid or adaption. • Full integration into society and local community.	**Realistic end goal of rehabilitation *** • Unilateral amputee: Walk independently with an aid or adaptation around the house and capable of using a wheelchair to mobilise in their local community. • Bilateral amputation: capable of using a wheelchair around their home and some aspects of local community.

* Depending upon severity of injury and the individual needs of the patient.

Figure 3.8: Comparing UK military and civilian care pathways for individuals with lower-limb amputation.

Abbreviations: DMRC – Defence Medical Rehabilitation Centre, NHS= National Health Service, ERI = Exercise Rehabilitation Instructor, ADL = Activities of Daily Living.

During in-patient rehabilitation at DMRC it is common practice for the prosthetist, physiotherapist, OT and ERI to work collectively with the patient in the same treatment sessions to ensure a shared understanding of the treatment goals and progress during every admission. Depending on the severity of injury, the length of rehabilitation at DMRC may last 1 to 2 years for a unilateral amputee and 2 to 3 years for a bilateral LLA. Therefore, the scope of practice and the amount of personalised rehabilitation delivered is considerably greater in the military model. The primary goal of the physiotherapist and occupational therapist working in the NHS is to ensure that the patient can walk independently with an aid or adaptions around their home. At DMRC it is to regain as much function as possible to ensure the patients attain their goals. This might be running independently or engaging in sports. At DMRC the prosthetist is afforded time and the most advance prosthetic provision available to achieve the best possible outcome for the patient. In the NHS, prosthetists strive for the best possible outcomes with the limited resources that their Trust can afford. At DMRC, ERI's provide the greatest amount of contact time with the patient (focussing on regaining muscle strength, muscle hypertrophy, balance and cardiorespiratory fitness). In the NHS, the ERI profession does not exist.

Despite the complex trauma rehabilitation pathway at DMRC Headley Court being described as exemplary, Winston Churchill was once quoted as saying:

> *"However beautiful the strategy, you should occasionally look at the results"*

In this next chapter, we retrospectively investigate the clinical outcomes recorded for individuals with traumatic LLA at their last admission to DMRC Headley Court, representing the final stage of the DMS combat casualty care pathway. The results will provide an insight into the functional and psychosocial status of UK military amputees after prolonged care and intensive residential rehabilitation.

CHAPTER 4. FUNCTIONAL AND MENTAL HEALTH STATUS OF UK MILITARY AMPUTEES POST-REHABILITATION

In the previous chapter, the UK combat casualty care pathway for military personnel with traumatic LLA was described from the point of injury to last admission at DMRC Headley Court. It is currently unknown what clinical outcomes are achieved by UK military personnel who follow this rehabilitation pathway. In this chapter the functional and psychosocial health status of UK military amputees on their last admission to DMRC are investigated. This chapter has been subject to peer-review and published in the Archives of Physical Medicine and Rehabilitation (Ladlow et al, 2015). The text below has been extracted from the published manuscript.

4.1. INTRODUCTION

Conflicts in Afghanistan and Iraq have led to advances in battle care emergency medicine enabling military personnel to survive injuries that would have once proved fatal (Gulland, 2008). These improved outcomes are credited to the high quality care from trauma on the battlefield to arrival back in the United Kingdom (UK) (Gulland, 2008). However, little empirical data exists regarding outcomes in traumatic amputees following rehabilitation. There is a medical and economic interest in ensuring that the trauma rehabilitation pathway is effective. Measuring physical function and mental health outcomes are essential to inform the development of evidence-based best practice.

There are no published data on the rehabilitation outcomes of UK military members with amputation from recent conflicts. Therefore, the purpose of this study was to evaluate the functional and mental health status of very severely injured UK military traumatic amputees upon completion of the rehabilitation pathway and comparing these data with published normative data.

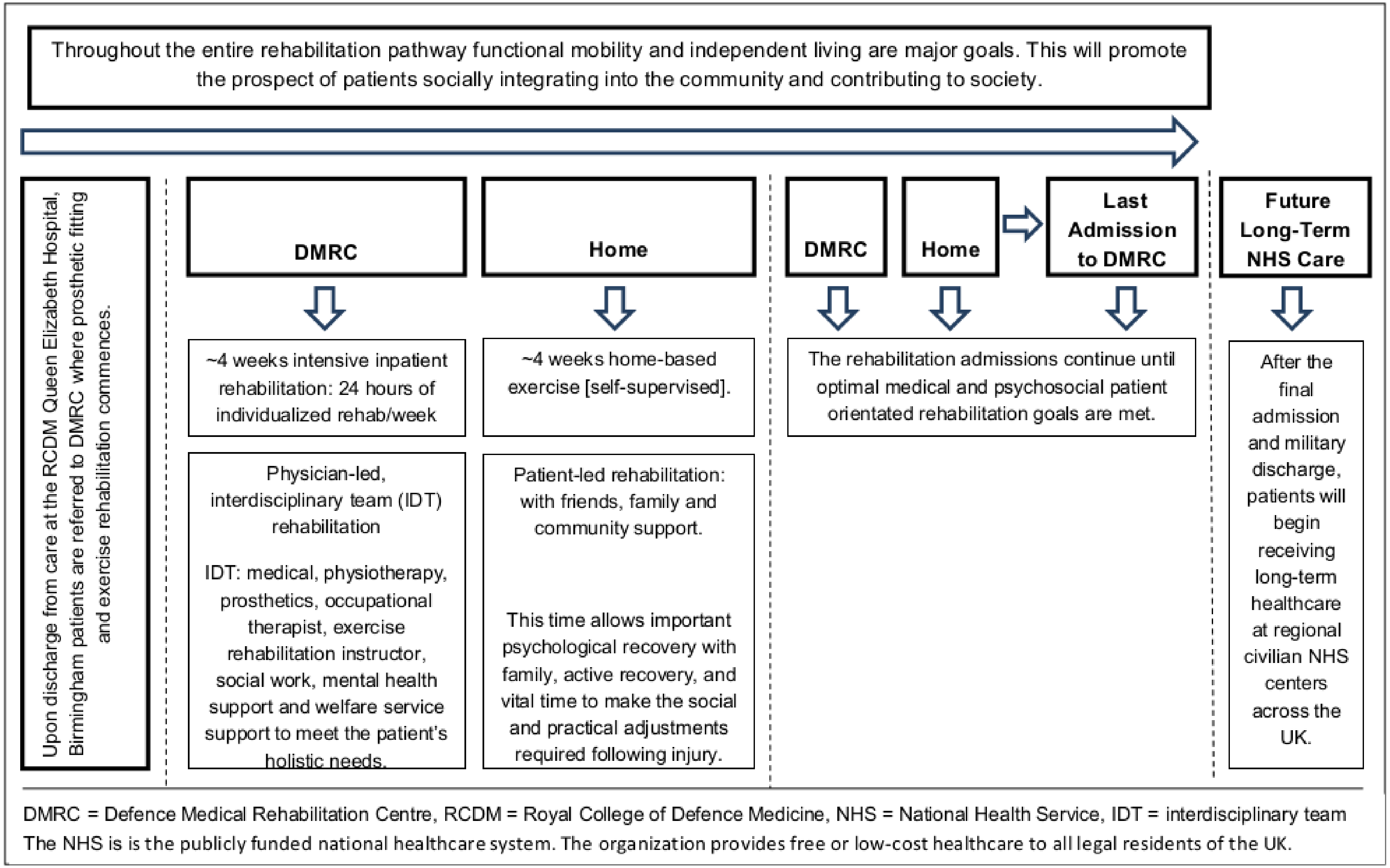
4.2. METHODS

4.2.1. Overview and Data Sources

A retrospective analysis of injury severity and the post-rehabilitation functional and mental health status of rehabilitation were undertaken in a cohort of military amputees. Patients discharged between January 2013 and March 2014 were included in the analyses. These dates were specified due to the online availability of patient records using the Defence Medical Information

Compatibility Program. Amputees were classified as any patient with an amputation above the ankle or above the hand. Patients with digital amputations only were excluded. Permission to access anonymous data was granted by the local Caldecott Guardian (person responsible for protecting the confidentiality of a patient, service-user information and enabling appropriate information-sharing). Ethical approval was granted by the Research Ethics Approval Committee for Health, University of Bath.

It is beyond the scope of this study to complete a detailed comparison of different NATO amputee care pathways. Combat casualty care for the US, Canadian and UK military has previously been described (Dharm-Datta & McLenaghan, 2013), along with a detailed analysis of US combat amputee care (Pasquina et al, 2009). The paradigm of UK military rehabilitation at DMRC was explained in chapter 3 and is summarised in figure 4.1.



Throughout the entire rehabilitation pathway functional mobility and independent living are major goals. This will promote the prospect of patients socially integrating into the community and contributing to society.

Upon discharge from care at the RCDM Queen Elizabeth Hospital, Birmingham patients are referred to DMRC where prosthetic fitting and exercise rehabilitation commences.

~4 weeks intensive inpatient rehabilitation: 24 hours of individualized rehab/week

Physician-led, interdisciplinary team (IDT) rehabilitation

IDT: medical, physiotherapy, prosthetics, occupational therapist, exercise rehabilitation instructor, social work, mental health support and welfare service support to meet the patient's holistic needs.

~4 weeks home-based exercise [self-supervised].

Patient-led rehabilitation: with friends, family and community support.

This time allows important psychological recovery with family, active recovery, and vital time to make the social and practical adjustments required following injury.

The rehabilitation admissions continue until optimal medical and psychosocial patient orientated rehabilitation goals are met.

After the final admission and military discharge, patients will begin receiving long-term healthcare at regional civilian NHS centers across the UK.

DMRC = Defence Medical Rehabilitation Centre, RCDM = Royal College of Defence Medicine, NHS = National Health Service, IDT = interdisciplinary team
The NHS is is the publicly funded national healthcare system. The organization provides free or low-cost healthcare to all legal residents of the UK.

Figure 4.1: The Paradigm of UK Military Rehabilitation at DMRC, Headley Court

Injury severity was calculated at the point of injury and recorded by the UK Joint Theatre Trauma Registry. Severity of Injury is defined and measured using the New Injury Severity Score (NISS). "Major trauma" has been defined as a NISS > 15 (Russell et al, 2011). NISS measures injury severity from 1 to 75, with a higher score signifying greater severity of injury. The distribution of body regions experiencing trauma were categorized as follows; head/face/neck, chest/upper back, upper-limb, spine, abdomen, pelvis, genitals and lower-limb. This information was used to define the number of anatomical regions injured in addition to the amputation. Patient demographics, mechanism of injury and length of rehabilitation were recorded. Predicted body mass and BMI were calculated according to methods outlined by Osterkamp (Osterkamp, 1995). Osterkamp estimated the percentage body mass loss of a limb following amputation for the foot (1.5%), below-knee (3.5%), above-knee (11%) and hip-disarticulation (16%). These estimated values were added to existing body mass to determine predicted body mass and predicted BMI.

Length of rehabilitation was defined as the number of four-week in-patient admissions and the number of months from first to last admission. Functional and mental health outcome measures were recorded upon completion of the rehabilitation pathway. All data were extracted from the Defence Medical Information Compatibility Program or from medical notes.

4.2.2. Functional Measures

The 6-MWT is an internationally recognised, validated outcome measure, used to assess function in a range of conditions (Laboratories, 2002). The test was performed indoors on a 20 meter flat surface with patients instructed to walk back and forth, turning around a cone, as many times as possible in 6-minutes. No assistive devices were used. The AMPQ is a functional outcome measure administered by a physiotherapist who observes and records the amputee patient's ability to perform basic mobility activities (Gailey et al, 2002). The Special Interest Group in Amputee Medicine (SIGAM) mobility assessment is a self-reported outcome tool that measures the potential of lower limb amputees to mobilize with their prosthesis (Ryall et al, 2003). In addition, two DMRC tools were completed by clinicians assessing the patient's ability to mobilize and perform ADL. 'Mobility' was recorded as the ability to 'run', 'walk independently', 'walk independently with an aid/adaptation' or 'requires wheelchair to mobilize'. The ability to perform ADL was recorded as 'independent', 'independent with aid/adaptation', 'assistance with some tasks' or 'requires constant care'. These measures are simple, quick to administer, easy to understand and provide clinically useful information on functional status.

4.2.3. Mental Health Measures

The Patient Health Questionnaire (PHQ-9) and General Anxiety Disorder (GAD-7) are validated self-reported questionnaires used to define severity of depression and general anxiety disorder, respectively (Kroenke et al, 2001; Spitzer et al, 2006). Both report a simple diagnostic grading scale of none/minimal to severe. In addition, two DMRC outcome tools were completed by clinicians to assess the requirement for mental health support - yes/no, and the assessment of ongoing pain; ‘no’, ‘controlled’ or ‘uncontrolled’ pain.

4.2.4. Statistical Analysis

The amputees were grouped according to the number of amputations (Unilateral, Bilateral and Triple) and in the unilateral LLA group whether their injuries were sustained operationally (combat injured) or non-operationally (non-combat injured). All multiple amputees were injured during operations. Statistical analysis was performed using SPSS Statistics v.21. All data were checked for normality using the Kolmogorov-Smirnov test and tests for skewness and kurtosis. One-way analysis of variance (ANOVA) was used to determine if there were significant main effects of the number of amputations (i.e. group) on injury severity, length of rehabilitation and the functional and mental health outcome measures. *Post-hoc* analyses using least-significant difference pairwise comparison tests were performed to determine differences between groups. For non-parametric data the Kruskal-Wallis test was used with *post-hoc* analysis performed using a Mann-Whitney U test. Level of significance was set *a priori* as $p > 0.05$.

4.3. RESULTS

4.3.1. Patient Demographics

Sixty-five amputees were evaluated at the completion of their rehabilitation pathway (unilateral LLA = 23, UNI-NonOp = 11, bilateral = 23 and triple amputee = 8), mean age 29 ± 6 years with 97% male. The number of amputations had a significant effect on predicted body mass ($F = 3.242$, $p = 0.028$) and BMI ($F = 3.569$, $p = 0.019$). Body mass and BMI were significantly higher in the UNI-NonOp compared to the two multiple amputee groups with no significant difference reported between the operational amputees ($p > 0.05$). Patient demographics and descriptive statistics are presented in table 1.

Table 4.1: Comparison of the patient demographics in all amputee groups on the last admission to DMRC, Headley Court. Values are mean ± SD (range)

	Amputee Groups				
Patient characteristics on last admission	Unilateral LLA	Unilateral NonOp	Bilateral LLA	Triple	Total Amputees
Number	23	11	23	8	65
Age (years)	28 ± 4 (20 - 40)	35 ± 10 (23 - 52)	28 ± 4 (22 - 35)	26 ± 3 (22 - 30)	29 ± 6 (20 - 52)
Gender (% male)	95.7	90.9	100	100	96.9
Pre-injury height (cm)	180 ± 10 (164 - 197)	178 ± 8 (163 - 192)	180 ± 7 (166 - 200)	177 ± 7 (166 - 189)	179 ± 8 (163 - 200)
Predicted body mass (kg)*	95 ± 16 (67 - 135)	104 ± 19 (84 - 125)	91 ± 16 (64 - 133)	82 ± 12 (66 - 101)	93 ± 17 (64 - 135)
Predicted BMI kg/m^2 *	29 ± 5 (22 - 40)	33 ± 4 (27 - 40)	28 ± 5 (22 - 44)	26 ± 4 (20 - 33)	29 ± 5 (20 - 40)
BMI $\geq$ 30 kg/m^2 (%)	43	55	22	13	34
Blood Pressure (mmHg):					
Systolic	133 ± 10 (112 - 147)	128 ± 8 (115 - 140)	134 ± 17 (108 - 186)	130 ± 12 (111 - 144)	132 ± 13 (108 - 186)
Diastolic	77 ± 12 (60 - 96)	70 ± 11 (53 - 93)	73 ± 6 (62 - 90)	77 ± 8 (62 - 92)	74 ± 10 (53 - 96)
Smoking Status:					
Non-smoker (%)	43.5	36.4	52	50	46
Previous smoker (%)	13	18.2	9	0	11
Current smoker (%)	43.5	45.4	39	50	43
Military Rank:					
Junior NCO's (%)	91	64	78	100	83
Senior NCO's (%)	9	36	9	0	12
Officers (%)	0	0	13	0	5
Years of service *Ɨ	8 ± 3 (2 - 14)	17 ± 10 (6 - 32)	7 ± 4 (4 - 17)	6 ± 3 (4 - 12)	9 ± 6 (2 - 32)

Abbreviation: LLA = lower-limb amputation, NonOp = non-operationally injured unilateral amputee, BMI = body mass index, NCO = non-commissioned officer (soldier), SD = standard deviation.

*significant difference ($p < 0.05$) between unilateral non-operational amputees and the multiple amputees (Bilateral and Triple)

Ɨ significant difference ($p < 0.001$) between unilateral non-operational and unilateral operational amputees

4.3.2. Injury Severity at Point of Injury

The number of amputations had a significant main effect on (F = 24.295, p < 0.001). The NISS revealed significant differences in injury severity between all operational amputee groups (table 2). Despite all patients having at least one amputation to the lower limb, there was a main effect of group (i.e. number of amputations) on the number of additional injuries sustained by patients (p = 0.005). A large proportion of patients sustained additional injuries to their remaining lower-limb (unilateral LLA = 74%, UNI-NonOp = 36%, bilateral = 43%, Triple = 25%) with 52% of the total cohort experiencing upper-limb injury.

A larger variety of trauma was experienced by the multiple amputees. The other most common injures in the bilateral LLA group included upper-limb (78%), genitals (48%) and head/neck/face (43%). Whilst in the triple amputee group, it was the head/neck/face (75%), genitals (63%), and upper-limb (38%). Improvised explosive devices (IEDs) were responsible for 97% of all operational injuries and 81% of the total cohort. The most common mechanism of injury among the non-operationally injured was through crushing (27%), road traffic accidents (RTA) (18%), and gunshot wound (GSW) (18%), while 88% were non-blast related.

Table 4.2: Comparison of injury characteristics on first admission to DMRC, Headley Court. Values are reported as mean ± SD (range).

	Amputee Groups				
Injury Characteristics	Unilateral LLA	Unilateral NonOp	Bilateral LLA	Triple	Total Amputees
Injury Severity:					
NISS * Ɨǂ	28 ± 11 (12 - 51)	N/A	44 ± 12 (22 - 75)	57 ± 6 (48 - 66)	40 ± 15 (12 - 75)
Additional number of body regions injured (excluding amputation) (m) §	2 ± 2 (0 - 5)	1 ± 2 (0 - 5)	3 ± 1 (1 - 5)	3 ± 1 (1 - 5)	2 ± 1 (0 - 5)

Abbreviation: LLA = lower limb amputation, NonOp = non-operationally injured unilateral amputee, NISS = new injury severity score, SD = standard deviation

*significant difference ($p < 0.001$) between Unilateral LLA and Bilateral groups
Ɨ significant difference ($p < 0.001$) between Unilateral LLA and Triple amputee groups
ǂ significant difference ($p = 0.009$) between Bilateral LLA and Triple amputee groups
§ significant difference ($p < 0.05$) between Unilateral NonOp amputees and all operational amputees

4.3.3. Length of Rehabilitation

The number of amputations had a significant effect on the length of rehabilitation (F = 8.031, p < 0.001) and the number of four-week admissions to DMRC (F = 3.036, p = 0.036). The mean duration of rehabilitation for the entire cohort was 34 ± 14 months and the mean number of four-week admissions was 11 ± 5. The UNI-NonOp amputees required significantly fewer months (20 ± 11 months) to complete the rehabilitation pathway compared to unilateral LLA (39 ± 15 months, p < 0.001), bilateral LLA (33 ± 10 months, p = 0.005) and Triple amputee (44 ± 9 months, p < 0.001). A difference was also found between the bilateral and triple amputees (p = 0.034). Each amputee group varied in the number of admissions (unilateral LLA = 11 ± 5; UNI-NonOp = 8 ± 4; bilateral = 13 ± 4; and triple amputee = 12 ± 4 admissions), with *post-hoc* analysis revealing that the only significant difference in number of admissions was between UNI-Non-Operational and bilateral groups (p = 0.006).

4.3.4. Physical Function

4.3.4.1. Six-Minute Walk Distance

The amputees had a mean six minute walk distance (6-MWD) (n=55) of 489 ± 117m. Ten patients did not perform the 6MWT, 4 required wheelchairs, 3 felt unwell, and 3 refused to perform the test. The number of amputations had a significant effect on the 6MWT (F = 5.844, p = 0.002). The two unilateral LLA groups walked similar distances (table 3). *Post-hoc* analysis revealed the unilateral LLA group walked significantly further than the bilateral LLA (p = 0.004) and triple amputee group (p = 0.002) (table 3).

4.3.4.2. Amputee Mobility Predictor Questionnaire (AMPQ)

The amputees achieved an AMP-Pro Score (n = 64) of 43 ± 5. One patient felt unwell and did not perform the assessment. Ninety-one percent of amputees attained at least a functional mobility score typical of a community ambulator (total AMP-Pro score = 37 to 42) (Gailey et al, 2002). The number of amputations had a significant effect on AMP-Pro outcome (p = 0.012). *Post-hoc* analysis revealed that the unilateral LLA group scored significantly higher than the bilateral LLA (p = 0.030) and triple amputee group (p = 0.039) (table 3) signifying a better mobility outcome, with no significant difference found between bilateral and triple amputee group (p > 0.05). Seventy-five percent of amputees attained an AMP-Pro score typical of an active adult or athlete (total AMP-Pro Score = 43 to 47) (Gailey et al, 2002).

Table 4.3: Comparison of 6-minute walk test (6MWT) and the Amputee Mobility Predictor Questionnaire (AMPQ) between amputee groups. Values are reported as mean ± SD (range)

	Amputee Groups				
	Unilateral LLA	Unilateral NonOp	Bilateral LLA	Triple	Total Amputees
6MWT					
No. patients	18	10	21	6	55
6MWT (metres) *	544 ± 99 (365 - 750)	544 ± 114 (380 - 760)	445 ± 104 (206 - 620)	387 ± 99 (270 - 510)	489 ± 117 (206 - 760)
AMPQ:					
No. Patients	23	10	23	8	64
Score (max 47) *	44 ± 3 (36 - 47)	45 ± 2 (41 - 46)	42 ± 5 (23 - 46)	37 ± 9 (24 - 47)	43 ± 5 (23 - 47)

Abbreviation: LLA = lower limb amputation, NonOp = non-operationally injured unilateral amputee, 6MWT = six minute walk test, AMPQ – Amputee Mobility Predictor Questionnaire, SD = standard deviation.

*Significant differences ($p < 0.05$) between both unilateral LLA groups and the bilateral and triple amputee groups.

4.3.4.3. Special Interest Group in Amputee Medicine (SIGAM)

The proportion of amputees that reported being able to “walk independently anywhere in any weather” was 91% (SIGAM grade F) (unilateral LLA = 100%, UNI-NonOp = 91%, bilateral = 91% and Triple = 63%). Ninety-five percent possessed the ability to “walk > 50 metres independently with an aid outside on level ground” (unilateral LLA = 100%, UNI-NonOp = 100%, bilateral = 96% and triple = 75%) (SIGAM grade D to F).

4.3.4.4. Defence Medical Rehabilitation Centre: Mobility and ADL Score

Results from the DMRC mobility score confirmed that 84% of the amputees were able to either walk or run independently in their prosthesis (Unilateral LLA = 87%, UNI-NonOp = 82%, bilateral = 92% and triple = 63%). When the ability to mobilize independently with an aid/adaptation was taken into consideration the overall proportion increased to 95% (Unilateral LLA = 100%, UNI-NonOp = 91%, bilateral = 92% and Triple = 75%). The proportion of amputees able to perform ADL independently was 43%. Differences were found between the two unilateral groups (61% and 45% in the unilateral LLA and UNI-NonOp, respectively). When the ability to perform ADL independently or independently with the use of an aid/adaptation was analysed, the proportion doubled to 95% (Unilateral LLA = 96%, UNI-NonOp = 91%, bilateral = 100% and triple = 88%).

4.3.5. Mental Health Status

4.3.5.1. PHQ-9: Depression

The mean score for the PHQ-9 was 3.1 ± 4.5, with the majority of amputee patients reporting “none” or “mild” depressive symptoms (75% and 14%, respectively). No patients reported “severe” symptoms. Seven patients (11%) scored ≥ “moderate” and two patients (3.1%) scored ≥ “moderate to severe”. The number of amputations did not have a significant effect on depression scores.

4.3.5.2. GAD-7: Anxiety

The mean GAD-7 score was 3 ± 4, with the majority of amputee patients reporting “minimal” or “mild” symptoms (71% and 22%, respectively). Five patients (8%) scored ≥ “moderate” and one patient (1.5%) scored ≥ “severe”. One-way ANOVA results showed that the number of amputations did not have a significant effect on Generalised Anxiety Disorder. However, a trend was found indicating a better outcome (“minimal” levels of anxiety) in those with the highest mean NISS (Unilateral LLA = 65%, UNI-NonOp = 55%, bilateral = 78%, triple = 88%).

4.3.5.3. Defence Medical Rehabilitation Centre: Mental Health and Pain Score

The proportion of amputees who required mental health support during their final admission was 29% (Unilateral LLA = 39%, UNI-NonOp = 36%, bilateral = 22%, triple = 13%). All operationally injured amputees are able to "control their pain" by the time of discharge. In the total cohort, 26% reported "no pain", 72% reported "controlled pain" and one patient (UNI-NonOp) experienced "uncontrolled pain". The most favourable outcome ("no pain") were reported in the operationally injured cohorts (Unilateral LLA = 30%, UNI-NonOp = 9%, bilateral = 26%, triple = 38%).

4.4. DISCUSSION

The cohort of amputees reported in this study had a mean NISS of 40 ± 15, which constitutes the highest severity of traumatic amputee injury in the available literature (Gaunaurd et al, 2013; Raya et al, 2013). Despite this, on completion of a comprehensive and intensive rehabilitation pathway these patients had functional and mental health status comparable with the general population.

Patient mobility is a critical component of successful rehabilitation and vital in aiding ADL (Burger et al, 1997). Therefore, functional levels that allow community ambulation should be the minimum requirement of a successful amputee rehabilitation program. After completing their military rehabilitation pathway, 64% were able to walk distances comparable to age-matched healthy controls (459 to 738m) (Chetta et al, 2006) (Unilateral LLA = 78%, UNI-NonOp = 80%, bilateral = 52% and triple = 33%), and 84% of amputees were able to walk or run independently. Linberg et al. (2013) reported 6MWD in a large cohort of United States (US) Service Members and Veterans. They reported a greater mean 6MWD in their cohort (503 to 661m). However, they did not state the cohorts' severity of injury, had different inclusion criteria (250m 6MWD and Level 37 on AMP-Pro) and employed a 6MWT protocol that required less pivoting and turning. Therefore, direct 6MWD comparisons between studies are difficult.

In this study, 75% had AMP-Pro scores consistent with active adults with limb-loss (Unilateral LLA = 87%, UNI-NonOp = 90%, bilateral = 65% and triple = 50%) and 91% had functional levels consistent with that of community ambulators with limb-loss (Unilateral LLA = 96%, UNI-NonOp = 100%, bilateral = 91% and triple = 63%). The mean AMP-Pro scores demonstrate that both unilateral and multiple amputees achieved mobility comparable to active adults and community ambulators with LLA, respectively (Gailey et al, 2002). A recent study of US service members with LLA, who had completed rehabilitation, reported comparable findings (mean AMPQ scores between 41 and 46 in unilateral LLA and bilateral groups) (Gailey et al, 2013b). This is important as it demonstrates, for the first time, the similarities in functional mobility achieved between US and UK military personnel injured during the Iraq and Afghanistan conflicts. Although the outcomes reported in this study are positive, it is important to note that a small number of patients leave DMRC not wearing their prostheses. In this group four patients required the use of a wheelchair, two for medical reasons and two due to individual preference.

It is known that individuals who experience major traumatic injuries and amputations have a greater risk of depression, anxiety (Horgan & MacLachlan, 2004) and post-injury suicide (March et al, 2014). The amputee rehabilitation pathway should therefore aim to achieve mental health outcomes that allow for the social re-integration of patients back into the community. Despite the severity of their injuries at the completion of their rehabilitation pathway this cohort of UK military amputees reported depression and anxiety levels similar to that of the general population (Kocalevent et al, 2013; Lowe et al, 2008).

When assessing the mental well-being of US and UK troops deployed to Iraq and Afghanistan the rates of mental health disorder vary, with less prevalence reported in UK military (Richardson et al, 2010; Stevelink et al, 2015; Sundin et al, 2014). Using the Revised Centre for Epidemiological Studies Depression Scale 38% reported depressive symptoms and 13% major depression (Doukas et al, 2013) compared to 11% and 3%, respectively, in this UK military cohort. However, given the descriptive nature of this data, it is not possible to discount the potential for under-reporting of depression and anxiety symptoms in the cohort. It is also important to note that the outcome measures used to define depression were different between studies. A number of reasons have been proposed for differences in mental well-being between the UK and US military, including differences in combat exposure, leader to enlisted soldier ratios, length of deployment, differences in access to long-term health care, socio-political and cultural factors (Hunt et al, 2014; Richardson et al, 2010; Sundin et al, 2014). The aetiology of mental health disorders in this population is varied and likely to be patient specific. Horgan and MacLachlan (2004) suggest that acceptance of a changed body image over time, higher levels of active coping, an optimistic personality disposition, increased levels of social support, greater satisfaction with the prosthesis, and decreased pain levels all contribute to better mental health outcomes over time. I also propose that a goal-oriented exercise rehabilitation program (figure 4.2) will improve function and ADL over time in amputees, thereby promoting a closer identification with their pre-injured self.

Figure 4.2: Triple amputee performing exercise rehabilitation at DMRC, Headley Court.

Interestingly, despite good functional and mental health status at the end of rehabilitation the mean predicted BMI (29 ± 4.8 kg/m^2) classifies the cohort as overweight and at risk of obesity. In this cohort, 34% were classified as obese (BMI $\geq$ 30 kg/m^2) compared to 26% reported in English men in 2013 (Team & Niblett, 2015). It is likely that during the periods spent away from DMRC, there is a reduction in PAEE and/or an increase in energy intake (i.e. eating behaviour) resulting in a positive energy balance and a consequent increase in BMI. It should also be recognized that the majority of these patients were transitioning out of the military and whilst most have made significant progress in the short to medium term, the long term general health and psychosocial outcomes are not known. A long-term follow up cohort study of combat casualties would be the best way to investigate these outcomes. Future studies should also aim to characterize the time course for the re-acquisition of physical function and recovery of mental health.

4.4.1. Study Limitations

Despite the intuitive appeal of an association between reported clinical outcomes and the content of the rehabilitation program, evidence demonstrating a causal link is lacking due to the cross sectional nature of this study design. Consequently, the data do not reflect the inevitable fluctuations that occur in functional and mental health scores throughout a prolonged period of rehabilitation. It also fails to capture the impact of leaving military service on functional and mental health outcomes, as well as important psychosocial factors such as employment status and withdrawal from the military environment.

4.5. CONCLUSION

This is the first time clinical outcomes following rehabilitation have been reported in UK military personnel with traumatic LLA from the recent conflicts in Iraq and Afghanistan. The results demonstrate that despite this being the most severely injured amputee cohort reported to date, functional and mental health status post-rehabilitation were comparable with a normal healthy population and are indicative of preparedness for full integration back into society.

CHAPTER 5. ESTIMATING AMBULATORY ENERGY EXPENDITURE IN UK MILITARY PERSONNEL WITH TRAUMATIC LOWER-LIMB AMPUTATION

The previous chapter described the functional and psychosocial status of UK military personnel with traumatic lower-limb amputation(s) attending their last admission at DMRC Headley Court. Although patients demonstrate clinical outcomes indicative of preparedness for full integration back into society, it is currently unknown whether the severity of their injuries lead to unfavourable secondary health outcomes like those detailed in chapter 2 (i.e., cardiovascular disease, type 2 diabetes mellitus, obesity). Prior to using objective PA devices in surveillance-based research (see chapter 6 and 7) it is essential to first ensure that the method used has been validated in the population of interest (i.e, UK military personnel with traumatic lower-limb amputation(s)). This is the aim of chapter 5.

This chapter has been subject to peer-review and published in the journal PLOS One. The text has been extracted from two published manuscripts (Ladlow et al, 2017; Ladlow et al, 2019). The raw data underlying this research are available from the University of Bath data archive. Data for experiment one can be found at http://doi.org/10.15125/BATH-00422 and data for experiment 2 can be found at http://doi.org/10.15125/BATH-00578.

5.1. INTRODUCTION

As described in chapter 2, there is a paucity of research investigating the impact of regular PA on the health and well-being of individuals following recovery from traumatic LLA. This is despite considerable evidence from longitudinal cohort studies demonstrating a substantially increased risk of this population developing a range of chronic degenerative disease (Foote et al, 2015; Robbins et al, 2009; Stewart et al, 2015). Strategies to mitigate the risk of managing these conditions are of upmost importance. Exercise and PA interventions aimed at improving function, health and wellbeing in individuals with LLA may benefit from the use of objective PA measurements.

In order to enhance research and practice in this field, it is important to develop valid and reliable tools to estimate free-living PAEE in LLA. PAEE has proven inherently difficult to measure, even in humans without mobility related physical impairments. This becomes more challenging within a heterogeneous group of individuals with LLA where the level of amputation results in a varying loss of articular structures and sensory/motor function of the lower-extremity (Czerniecki & Morgenroth, 2015). It is well established that a higher level of amputation (above knee versus below knee) and a greater number of lower-limbs amputated (bilateral versus unilateral) are associated with a higher metabolic cost of walking and reduced ambulatory PA (Gjovaag et al, 2014; Hoffman et al, 1997; Ladlow et al, 2016). Indeed, investigations into the daily PA and HR responses of people with vascular (Bussmann et al, 2004) and traumatic (Bussmann et al, 2008) unilateral trans-tibial amputations have demonstrated that both amputee groups are less active than matched controls without known physical impairments. These studies highlight the impact of physical disability and associated mobility restriction on volitional PA behaviour. However, little is known about the consequences of amputation severity on habitual PA levels or ambulatory PAEE. Consequently, PA requirements for the maintenance or improvement of metabolic health and protection against chronic degenerative diseases, are poorly understood in this population. Hence the ability to accurately measure and predict PAEE are critically important in the long-term management and prevention of chronic diseases in persons with LLA.

To date, the ability to accurately predict free-living PAEE in individuals with unilateral and/or bilateral lower-limb amputation(s) has not been explored. An objective method for assessing habitual PA in this population would allow the development of bespoke PA guidelines, allow appropriate cross-sectional comparisons and enhance research efforts on the efficacy of PA interventions. Criterion or 'gold standard' measures of EE (i.e. indirect calorimetry and doubly labelled water) are highly accurate, but relatively expensive, requiring sophisticated equipment rendering them impractical to use outside of the laboratory when assessing free-living PA. The ability to detect subtle or large variances in the duration spent at different PA levels during free-living conditions, patterns of PAEE (morning, afternoon, evening) and exercise intensity are not possible using doubly labelled water technique (only total EE) during a monitoring period.

Previous research on amputee mobility has relied on subjective amputee specific self-report questionnaires (Miller et al, 2001; Parker et al, 2010; Stepien et al, 2007) in conjunction with objective measures of functional mobility (e.g. step count and timed up and go). Limitations to using self-report measures of PA are well-known and include inaccurate subjective reporting and recall bias (Sallis & Saelens, 2000). Over 17 years ago, Bussmann and colleagues (Bussmann et al, 1998; Bussmann et al, 2001) validated the uni-axial IC-3031 and ADX202 body fixed accelerometer in individuals with unilateral amputation. However, the aim of these two investigations was only to assess the accelerometers ability to identify posture and motion by comparing outputs to video recordings. Although useful for identifying movements performed in controlled rehabilitation settings, these studies did not attempt to predict EE, nor the metabolic cost of prosthetic ambulation.

Having access to an objective measurement of PAEE can facilitate our understanding of a number of other clinically important areas with greater accuracy than we might have had before. However, a recent systematic review (Piazza et al, 2017) of instruments used for the assessment of PA in individuals with amputation demonstrated that most instruments are not specific enough for the population. Accelerometer assessed PAEE, using algorithms intrinsic to certain devices may not be generalisable to a target population (Pedisic & Bauman, 2015), an important consideration for individuals predisposed to significant gait deficiencies such as individuals with LLA (Sagawa et al, 2011; Su et al, 2007). Therefore, the logical first step prior to using objective devices in surveillance research is to ensure that the method has been validated in the population of interest.

Recent technological advancements in the field of PAEE measurement has stimulated the development of sensitive tri-axial accelerometers, which are unobtrusive, low-cost devices, capable of storing higher resolution raw, unfiltered acceleration signals over prolonged periods of time (Intille et al, 2012). Accelerometers are widely used in the assessment of human EE (Crouter et al, 2006; Lyden et al, 2011) and have previously been shown to be best placed on hip or the lower back during free living activity (Trost et al, 2005). Hip based uni-axial and tri-axial accelerometers have been used for the measurement of PA in clinical populations with functional limitations including: stroke (Rand et al, 2009), multiple sclerosis and Parkinson's disease (Hale et al, 2008). The Actigraph™ GT3X+ is a tri-axial accelerometer that has previously been validated in wheelchair users (Nightingale et al, 2014). The ability of the GT3X+ accelerometer to accurately predict PAEE over a variety of ambulatory velocities and gradients in people with unilateral and bilateral LLA remains unclear.

A known limitation to accelerometer based PA data is the inability to capture any physiological strain associated with certain ambulatory behaviours, such as walking up a gradient (Lamonte & Ainsworth, 2001). Multi-sensor devices, incorporating physiological signals, might offer a greater improvement in prediction accuracy (Strath et al, 2005). HR is known to be a valuable physiological signal in the estimation of EE due to its near linear relationship. The Actiheart™ (AHR) is a research-grade multi-sensor device worn on the chest which incorporates HR and uniaxial accelerometry measurements to predict PAEE (Brage et al, 2005). It has been widely used to measure free-living PA in able-bodied individuals but further research in diverse populations, such as individuals with amputation, is warranted. It is unknown whether the proprietary predictive algorithms of the AHR can accurately predict ambulatory PAEE in persons who have experienced LLA.

To date, there are no published studies to determine the most appropriate anatomical placement of accelerometers to accurately predict PAEE in unilateral or bilateral amputees. Consequently, there are no peer-reviewed articles, which have attempted to develop population specific algorithms for the prediction of PAEE in individuals with lower-limb amputation(s). It is also unknown whether the combination of Actigraph GT3X+ triaxial accelerometer and HR data (GT3X+HR) could be used to derive a predictive algorithm with comparable or superior accuracy to the AHR for the estimation of ambulatory-based PAEE. Therefore, the first study in this chapter (experiment one) aims to evaluate the influence of anatomical positioning of the Actigraph GT3X+ accelerometer around the pelvis on the prediction of PAEE in UK military personnel with traumatic lower-limb amputation(s) and to develop valid population specific algorithms during a variety of ambulatory velocities. The second aim of the study (experiment two) aims to test the hypothesis that a bespoke algorithm (GT3X+HR) would demonstrate greater validity and lower random error in predicting ambulatory PAEE in the same group of UK military personnel with traumatic lower-limb amputation(s). These manually derived population specific algorithms will then be compared with the proprietary predictive algorithms of the AHR, a research grade multi-sensor device.

5.2. METHODS

Ethics approval was granted by the UK Ministry of Defence Research Ethics Committee and written informed consent was obtained from each participant. The estimation of ambulatory EE was performed on a treadmill using a range of walking speeds and gradients. Experiments one and two represent two separate research questions; however, all activity monitors (GT3X+, AHR and HR) were worn simultaneously alongside indirect calorimetry based methods of measuring EE, during one visit to the laboratory.

5.2.1. Determining Sample Size

An *a priori* power calculation was performed based on data from a previous study in spinal cord injured humans (Nightingale et al, 2015). It was estimated that a minimum of 8 participants would be required to detect a statistically significant difference in mean absolute error (MAE) between the AHR proprietary predictive algorithms (MAE = 51.4 ± 38.9%) and a bespoke individually calibrated algorithm (MAE = 16.8 ± 15.8%), giving an estimated effect size of (Cohen *d*) of 1.0 (Cohen, 1988). The power was set at 0.8 and the alpha at 0.05. Given the distinct challenges in recruiting and retaining participants from unique clinical populations, I anticipated a ~20% drop-out rate and aimed to recruit 10 participants per group to achieve a final sample of at least 8.

5.2.2. Participants

A sample of ten unilateral and ten bilateral military amputees and ten uninjured healthy controls volunteered to participate in this study. All participants were male and visited the Military

Performance and Rehabilitation Laboratory at the DMRC, Headley Court on one morning after a ten hour overnight fast (including abstinence from caffeine and exercise). Inclusion criteria included all injured participants having experienced traumatic LLA and had previously received at least three 4-week in-patient admissions of intensive exercise rehabilitation at DMRC Headley Court from an interdisciplinary team of health professionals (Ladlow et al, 2016; Ladlow et al, 2015). All patients received a prosthetic fitting prior to commencing the trial and had been given clearance to ambulate on a treadmill by their physiotherapist. Exclusion criteria were based upon the participant's medical history (screened by their physician). This includes severe traumatic brain injury, medication that alters heart rate variability, and any mobility restricting conditions, such as painful heterotopic ossification or insufficient wound healing around the stump. The control group are uninjured physically active men (civilian and military who engage in aerobic or resistance based training at least three times per week). The clinical population being tested are all UK military personnel with traumatic lower-limb amputation(s) who (pre-injury) would have been active uninjured adults. By using an age-matched active male adult population as a control it was possible to comment on the impact of losing a limb and the ability of proprietary algorithms ability to detect such a change against normative activity data from their uninjured peers.

5.2.3. Indirect Calorimetry

Participants wore a sealed face mask and expired gases were analysed using a portable metabolic system (Metamax 3B, Cortex, Leipzig, Germany), which has good accuracy compared to other portable metabolic systems (Vogler et al, 2010). Expired gases passed through a flow meter and are channelled down a sampling line into the analyser unit where the fractions of O_2 and CO_2 in expired gases are measured. Metabolic data were retrieved and analysed using the Metamax software. $\dot{V}O_2$ steady state can be achieved within 3 minutes (Whipp & Wasserman, 1972). Oxygen uptake ($\dot{V}O_2$) and carbon dioxide production (CO_2) were used to estimate EE ($kcal \cdot min^{-1}$) during the final two minute of each five minute activity. The Metamax was calibrated according to manufacturer's instructions prior to use.

5.2.4. Activity Monitors

5.2.4.1. Actigraph GT3X+ Accelerometer

The GT3X+ activity monitor (ActiGraph, Pensacola, FL) records time-varying accelerations within the dynamic range of ±6g and contains a solid-state triaxial accelerometer sensitive to movement along three axes: anteroposterior (*X*), mediolateral (*Y*), and vertical (*Z*). The GT3X+ activity monitor is compact (dimensions, 4.6 x 3.3 x 1.9 cm), lightweight (19 g), and can easily be worn at multiple locations on the body. Each unit is powered by a rechargeable lithium ion battery and has a memory of 512 MB. Approximately 40 days of PA data can be recorded when sampling at a frequency of 30 Hz, although the battery would need recharging after 30 days. To quantify the amount and frequency of human movement, accelerometer outputs are digitized via a 12-bit analog-to-digital converter and passed through ActiGraph's proprietary digital filtering algorithms. To eliminate any acceleration noise outside the normal human activity frequency, digitized signals pass through low-bandwidth (0.25 Hz) and high-bandwidth (2.5 Hz) filters (John & Freedson, 2012). The GT3X+ records time-varying accelerations at a user-defined sampling frequency ranging from 30 to 100 Hz. These are then converted to arbitrary units called ''physical activity counts''. These are calculated through summing the change in raw acceleration values measured during a specific interval of time or ''epoch''.

Throughout the activity protocol, three Actigraph GT3X+ triaxial accelerometer units were worn, one on either side of the waist, above the hip (along the anterior axillary line) and one on the lower back (positioned on L2) all via an elasticated belt. Following the Nyquist principle, the devices were initialised with a sampling frequency of 30 Hz (a similar sampling frequency to the AHR monitor), thereby allowing the capture of general human movement (Chen et al, 2012).

5.2.4.2. Actiheart Monitor

Participants wore an AHR (Actiheart^{TM}, Cambridge Neurotechnology Ltd, Papworth, UK), which integrates accelerometer and HR signals, on their chest. The AHR unit has been described in detail previously (Brage et al, 2005). The main body of the device contains an omni-directional accelerometer with a sampling rate of 32 Hz and a dynamic range of ±2.5 *g*. When exposed to time-varying acceleration the voltage signal generated by the piezo-electric element is converted into a binary signal by an eight-bit analogue to digital converter. The accelerometer in this device has a linear ($R^2 = 0.999$) response to acceleration (Brage et al, 2005). The AHR frequency range is 1 Hz to 7 Hz with a memory capacity capable of storing 21 days of data when recording at 60 s epochs. The AHR consists of two clips which were attached to standard adhesive electrocardiogram (ECG) electrodes (Telectrode T815, Bio-Protech Inc., Exeter, UK), which were then fitted to the participant according to manufacturer's instructions. HR (bpm) is generated from an ECG signal. Participants wore an AHR monitor according to the manufacturer's instructions. AHR devices were initialised to long-term recording with 30 second epochs at a sampling frequency of 32 Hz. PAEE was calculated using Branched Model equations (Brage et al, 2004).

5.2.5. Heart Rate

A Polar T31 HR monitor (Polar Electro Inc., Lake Success, NY, USA) was firmly secured on the chest using an elastic strap and ultrasound gel was applied to the electrodes to improve the connection. HR transmitted by the Polar T31 was captured by a wireless receiver module connected to the Metamax 3B.

5.2.6. Testing Protocol

Anthropometric data were collected at the start of the protocol, including: body mass (with and without prosthesis), stature, hip and waist circumference and, level (below knee, through knee and above knee) and number of amputations (unilateral or bilateral). The time since amputation, which indicates the length of rehabilitation (months), was also recorded. All amputations were performed at an anatomical level above the ankle and below the hip. The Metamax 3B, three GT3X+ accelerometers and AHR device were all synchronised before use and a Polar HR monitor was worn throughout the protocol. RMR (kcal·day^{-1}) was measured in a semi-recumbent position in accordance with best practice guidelines (Compher et al, 2006). Following the measurement of RMR and anthropometric assessment, participants completed a walking protocol on a level treadmill (Woodway Desmo, USA). This protocol consisted of ambulating at 5 progressive velocities (0.48, 0.67, 0.89, 1.12, 1.34 m.s^{-1} or 1, 1.5, 2, 2.5 and 3 mph, respectively) and 2 gradients (3% and 5%) at 0.89 m.s-1 (2 mph). This would provide a wide range of ambulatory velocities to be captured in a heterogeneous group of traumatic LLA. Each activity lasted 5 minutes with no recovery between each intensity increments. The velocities were determined by self-selected walking speeds performed on a similar group of UK military amputees (Jarvis et al, 2017). Participants were asked to complete the entire protocol without resting their arms on the handrail. Participants were told to stop if they experience residuum pain, prosthetic discomfort or difficulty maintaining the speed of the treadmill belt to a point where they felt they were at risk of falling. Rating of perceived exertion (RPE) was collected at the end of each treadmill intensity using the 6 to 20 Borg Scale (Borg, 1982).

5.2.7. Calculating PAEE

Breath-by-breath data was exported into Microsoft Excel from the Metamax 3B software. PAEE was then calculated using the $\dot{V}O_2$ and CO_2 values (L·min^{-1}), averaged over the final 2 minutes of each activity using the Weir equation (Weir, 1949).

As participants were fasted, dietary-induced thermogenesis was considered negligible and criterion PAEE was calculated by subtracting RMR (kcal·min^{-1}) from total EE. METs were then calculated using measured exercise VO_2 divided by resting VO_2 to derive individual METs in the last 2 minutes of each treadmill intensity. The three GT3X+ accelerometer units were downloaded using ActiLife software (ActiGraph, Pensocola, FL, USA). Data were exported to Microsoft Excel in a time and date stamped comma-separated value (CSV) file format. AHR data was ascertained via entering the measured RMR (via indirect calorimetry), age, weight, height and sleeping HR (measured the night before testing) into the AHR software (Version 4.0.23), according to the manufacturer's instructions. Activity counts (counts·min-1) from the three GT3X+, Polar HR (bpm) and AHR readings (kcal·min^{-1}) were then averaged over the final two minutes of each activity (representative of steady-state) and comparisons made to criterion outputs (indirect calorimetry). For experiment two, predicted PAEE using GT3X+HR were derived by combining tri-axial accelerometer counts from the GT3X+ with HR using regression methods.

5.2.8. Statistical Analyses

5.2.8.1. Experiment One

PAEE prediction models were developed using corresponding data from each task for devices at each location, using linear regression analysis. The dependent variable was PAEE (kcal·min^{-1}) during the final 2 minutes of each task (that is 80 values in each group). The independent variable was accelerometer outputs (counts·min^{-1}) for the GT3X+. Pearson product moment correlation coefficients (*r*) and coefficients of determination (R^2) statistics were reported to assess the association between criterion PAEE and outputs from devices at each location. Standard Error of the Estimate (SEE) was calculated for each model (Model 1).

The GT3X+ worn at the anatomical position with the strongest relationship to the criterion PAEE was then selected for further analysis, to develop a predictive model for PAEE. Covariates, which included age, body mass, waist circumference, time since amputation, and level of amputation, were analysed to determine their association with the criterion PAEE depending on if data was discrete or continuous. These covariates were selected due to their influence upon mobility in US military amputees (Gaunaurd et al, 2013). Significant covariates were included in the stepwise regression analysis to strengthen the predictive PAEE equations in each group (Model 2).

These predictive models should be cross-validated using an independent sample. However, this is not always possible in hard to reach populations due to recruitment issues. To overcome this problem prediction algorithms, to determine the PAEE prediction error, were developed using a systematic `leave-one-out' cross validation analysis (Hastie et al, 2005), as performed previously by Nightingale et al. (2015). In summary, this process was repeated where each participant acted as the `held-out' participant and the mean error of all calculations was determined. Error statistics involved calculating the MAE, mean absolute percentage error and mean signed error for each activity; the latter displayed graphically using Bland and Altman plots and limits of agreement (LoA) analysis. A two way mixed model ANOVA was performed to determine differences between criterion PAEE and predicted PAEE at each treadmill task. Where a significant interaction effect was observed, a Bonferroni correction was applied to *post-hoc* tests where multiple comparisons were considered. This was to identify the specific treadmill tasks in which there was a significant difference between the criterion and predicted PAEE.

5.2.8.2. Experiment Two

The GT3X+ worn at the anatomical position with the strongest relationship to the criterion PAEE (as determined by experiment one) was selected for further analysis. PAEE estimation models for the GT3X+HR were developed using corresponding data from each task, using multiple linear regression analyses. The dependent variable was indirect calorimetry PAEE (kcal·min^{-1}). The independent variables were PAC (counts·min^{-1}) from the GT3X+ with HR (bpm). Pearson product moment correlation coefficients (*r*) and coefficients of determination (R^2) statistics were conducted to assess the association between the criterion PAEE and predicted PAEE for GT3X+HR, HR and AHR (AHR data; using proprietary group calibration). SEE statistics was also calculated for each relationship. As mentioned previously, population specific equations (GT3X+HR) would have ideally been cross-validated using an independent sample. As this was not possible, the same leave-one-out ‘bootstrapping’ analysis as used in experiment one was adopted (Hastie et al, 2005). As performed in experiment one, error statistics involved calculating the MAE, mean absolute percentage error and mean signed error for each activity (displayed graphically using modified box and whisker plots) and Bland-Altman plots with 95% LoA analysis. Two-way ANOVA tests by group were performed with *post-hoc* Bonferroni corrections applied when comparing across 8 activities (rest, five progressive treadmill speeds and 2 gradients). All analyses were performed using IBM SPSS Statistics 21 for Windows (IBM, Armonk, NY, USA) with statistical significance set a priori of $p < 0.05$.

5.3. RESULTS

5.3.1. Experiment One

Demographic and physical characteristics of the participants are described in table 5.1. Criterion PAEE (kcal·min^{-1}), ActiGraph GT3X+ accelerometer outputs at each anatomical location, RPE and METs are displayed in table 5.2. Despite the lower mean RMR values in the bilateral amputee group, there were no significant main effects or group differences ($p > 0.05$). Not all amputee participants were able to complete all of the treadmill speeds in this trial. The number of participants that dropped out of each treadmill task is also presented in table 5.2. There was a significant main effect on actual PAEE, predicted PAEE, and METs, with significant differences between all three groups. Mean PAEE, PAC, RPE and METs increased with increasing velocity of the treadmill in the unilateral and control group. In the bilateral group, six to eight participants were unable to complete activities at the higher treadmill velocities (see table 5.2).

All participants with a through and/or above knee amputation, in both groups (i.e. unilateral and bilateral), wore a Genium prosthetic device during all activities. Five of the seven below knee amputees wore a Variflex XC, whilst one used an Echelon VT and the other wore a Panthera CF2. The prosthetic devices worn by the bilateral amputees with an above and below knee combination included a BiOM and a Variflex XC for the below knee prosthesis and a Genium for their above knee amputation.

Across all treadmill walking tasks, PAEE was 1.4 to 1.7 times greater in the unilateral amputees compared to the uninjured controls. At treadmill speeds between 0.48 to 0.89 m.s^{-1} and a gradient of 3% at 0.89 m.s^{-1}, PAEE was 2 to 2.6 times greater in bilateral amputees compared to controls. At these same walking speeds PAEE was 1.4 to 1.6 times greater in the bilateral compared to the unilateral amputees. The difference in PAC between both amputee groups versus control was greatest at slower walking speeds with the relative differences in PAC reducing at faster walking speeds. There was a significant difference in PAC (GT3X+ worn on shortest limb) between all groups at 0.48 m.s^{-1}, 0.67 m.s^{-1} and 0.89 m.s^{-1} ($p < 0.001$). The MET data suggest that, for all walking speeds above 0.48 m.s^{-1}, for the unilateral and bilateral groups, exercise intensity was considered to be of moderate-intensity. For the normative control group, moderate intensity activity only occurred at a walking speed of 1.12 m.s^{-1}.

Table 5.1: Demographic and physical characteristics of the participants. Information displayed as mean ± SD

Variable	**Unilateral**		**Bilateral**		**Control**	
	Mean ± SD	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**
Number of Participants	10		10		10	
Age (years)	32 ± 5	23 - 41	29 ± 4	22 - 34	32 ± 6	25 - 45
Body Mass - without prosthesis (kg)	81 ± 11	63 - 108	82 ± 19	59 - 126	79 ± 7	68 - 89
Waist Circumference (cm) *	92 ± 12	75 - 115	100 ± 20	77 - 149	84 ± 4	76 - 90
Waist-hip ratio	0.90 ± 0.06	0.83 - 1.00	0.94 ± 0.09	0.86 - 1.17	0.86 ± 0.04	0.79 - 0.92
RMR ($kcal \cdot d^{-1}$)	1800 ± 264	1480 - 2158	1596 ± 178	1382 - 2051	1808 ± 217	1463 - 2059
Time Since Amputation (months) ǂ	24 ± 15	4 - 46	39 ± 14	21 - 61	-	-
Level of Amputation:						
Below Knee	6		1		-	
Through Knee	2		2		-	
Above knee	2		3		-	
Bilateral: Below Knee and Above Knee	-		2		-	
Bilateral: Through Knee and Above Knee	-		2			

*Significant difference between bilateral amputees and control group ($p < 0.05$).

ǂ Significant difference between unilateral and bilateral amputees ($p < 0.05$).

Table 5.2: Measured PAEE, accelerometer outputs at each anatomical location, calculated METs, RPE and number of participants for each activity (mean ± SD)

Activity	PAEE Metamax 3B (kcal·min^{-1})	GT3X+ (PAC·min^{-1}) Longest Limb	GT3X+ (PAC·min^{-1}) Spine	GT3X+ (PAC·min^{-1}) Shortest Limb	METS (calculated)	RPE	n
Unilateral Amputees: ǂ # ¥							
RMR	0.0 ± 0.0	0 ± 0	0 ± 0	0 ± 0	1.0 ± 0.0	6 ± 0	10
Treadmill 0.48 m.s^{-1}	2.4 ± 0.7	2361 ± 710	2256 ± 777	2691 ± 831	3.1 ± 0.7	8 ± 1	10
Treadmill 0.67 m.s^{-1}	2.9 ± 0.9	2665 ± 639	2517 ± 577	2945 ± 856	3.5 ± 0.9	9 ± 1	10
Treadmill 0.89 m.s^{-1}	3.6 ± 1.1	3038 ± 583	2904 ± 597	3384 ± 848	4.0 ± 1.0	11 ± 2	10
Treadmill 1.12 m.s^{-1}	4.3 ± 1.4	3723 ± 457	3673 ± 521	4130 ± 670	4.6 ± 1.3	12 ± 2	10
Treadmill 1.34 m.s^{-1}	5.6 ± 1.7	4703 ± 674	4794 ± 660	5126 ± 539	5.3 ± 1.6	12 ± 2	7
Treadmill 3% (0.89 m.s^{-1})	4.1 ± 1.1	3131 ± 514	3044 ± 648	3671 ± 929	4.4 ± 1.1	11 ± 1	10
Treadmill 5% (0.89 m.s^{-1})	4.8 ± 1.2	3370 ± 537	3258 ± 492	4018 ± 948	4.9 ± 1.2	12 ± 2	10
Bilateral Amputees:* §							
RMR	0.0 ± 0.0	0 ± 0	0 ± 0	0 ± 0	1.0 ± 0.0	6 ± 0	10
Treadmill 0.48 m.s^{-1}	3.7 ± 1.4	4132 ± 1645	3449 ± 696	4800 ± 1410	4.4 ± 1.2	10 ± 2	10
Treadmill 0.67 m.s^{-1}	4.6 ± 1.5	4453 ± 2044	3792 ± 829	5264 ± 1603	5.1 ± 1.4	12 ± 2	10
Treadmill 0.89 m.s^{-1}	5.5 ± 1.7	4843 ± 2101	4199 ± 822	5600 ± 1502	5.8 ± 1.6	14 ± 3	10
Treadmill 1.12 m.s^{-1}	5.5 ± 2.9	6596 ± 3943	4846 ± 1363	6123 ± 2823	5.3 ± 1.4	15 ± 3	3
Treadmill 1.34 m.s^{-1}	6.3 ± 2.9	5251 ± 835	4907 ± 518	5235 ± 1212	5.7 ± 1.7	15 ± 0	2
Treadmill 3% (0.89 m.s^{-1})	5.9 ± 2.3	5064 ± 1771	4408 ± 834	5973 ± 1592	6.1 ± 2	14 ± 3	8
Treadmill 5% (0.89 m.s^{-1})	5.8 ± 1.9	5594 ± 2509	4813 ± 1289	5806 ± 2231	5.7 ± 1.1	16 ± 2	4

Control:							
RMR	0.0 ± 0.0	0 ± 0	0 ± 0	0 ± 0	1.0 ± 0.0	6 ± 0	10
Treadmill 0.48 m.s^{-1}	1.4 ± 0.3	1542 ± 495	1352 ± 455	1416 ± 536	2.2 ± 0.3	7 ± 0	10
Treadmill 0.67 m.s^{-1}	1.8 ± 0.3	2006 ± 336	1776 ± 278	1876 ± 403	2.5 ± 0.3	8 ± 1	10
Treadmill 0.89 m.s^{-1}	2.3 ± 0.4	2577 ± 369	2290 ± 332	2478 ± 459	2.9 ± 0.4	9 ± 1	10
Treadmill 1.12 m.s^{-1}	2.8 ± 0.4	3463 ± 398	3162 ± 394	3353 ± 461	3.3 ± 0.4	9 ± 1	10
Treadmill 1.34 m.s^{-1}	3.3 ± 0.4	4321 ± 469	4096 ± 429	4205 ± 471	3.7 ± 0.3	10 ± 1	10
Treadmill 3% (0.89 m.s^{-1})	2.9 ± 0.4	2732 ± 243	2395 ± 265	2591 ± 385	3.4 ± 0.3	10 ± 1	10
Treadmill 5% (0.89 m.s^{-1})	3.4 ± 0.4	2924 ± 280	2598 ± 187	2766 ± 327	3.8 ± 0.3	10 ± 1	10

Abbreviatioons: PAEE = physical activity energy expenditure, PAC = physical activity count, METs = metabolic equivalent, RPE = rate of perceived exertion.

*Due to reduced participant numbers, all statistical analyses comparing the bilateral group with other groups were performed at speeds 1-2 m.s^{-1} and at 3% gradient.
ǂ A significant difference in criterion PAEE and METs were only reported at higher intensities (2.5 m.s^{-1}, 3 m.s^{-1} and 5% gradient at 2 m.s^{-1}) between the unilateral amputees and control group ($p < 0.05$).
§ A significant differences in criterion PAEE, METs, PAC (GT3X+ worn at the longest and shortest limb) were found between bilateral amputees versus the unilateral and control groups all speeds analysed ($p < 0.05$).
# Significant differences in PAC (GT3X+ worn on shortest limb) were only reported at the higher intensities of 3 m.s^{-1} and 5% gradient at 2 m.s^{-1} between unilateral amputees and control group ($p < 0.05$).
¥ Significant difference in PAC (GT3X+ worn on spine) were found at 1 m.s^{-1}, 1.5 m.s^{-1} and 3% Gradient at 2 m.s^{-1} between unilateral amputees and control group ($p < 0.05$).

PAC from each anatomical location were significantly (*p* < 0.01) associated with criterion PAEE. In both of the amputee groups the GT3X+ worn on the hip with the shortest residual limb demonstrated the strongest relationship, smallest LoA (table 5.3) and MAE (See table 5.4, which illustrates the error of the GT3X+ monitor at each anatomical location and the generated predictive model for all three groups). In the control group, the strength of the relationship and level of error was similar at each anatomical location. The correlation between criterion and predicted PAEE at the most accurate anatomical location, for each group, are presented in figure 5.1.

Figure 5.2 (panels a-c) illustrates the difference between criterion and predicted PAEE derived from population specific prediction models (Model 2) through the use of Bland and Altman plots [mean ± 95% LoA]. These reveal a degree of heteroscedasticity (error increases as exercise intensity increases) in the control group. When comparing the two amputee groups these plots demonstrate similar mean bias between groups, which is greater than that in the control group. There are considerably larger LoA in the bilateral amputee group across all treadmill tasks. MAE statistics between the criterion and estimated PAEE from each anatomical location and the generated model for each treadmill task are shown in the table 5.4. Greatest error was reported in the unilateral amputee group (mean absolute percentage error, unilateral: 21±17%, bilateral: 16±15%, control: 15±7%) using the population specific generated models. Modified box and whisker plots depicting the mean percentage error of estimation relative to criterion for each treadmill activity using the cross validated, population specific prediction models (model 2) are found in figure 5.3.

Table 5.3: The relationship between predicted PAEE using the Actigraph GT3X+ and criterion PAEE at the three anatomical positions in all three groups. The table displays the predictive equations used in the most accurate accelerometer location, the shortest residual limb (Model 1) and the impact of significant covariates (Model 2) at increasing the accuracy of the GT3X+ accelerometer at predicting PAEE. Limits of agreement (LoA) expressed as mean ± 95% SD

Location	r	R^2	SEE (kcal·min^{-1})	LoA (kcal·min^{-1})	P Value
Unilateral Amputee Group					
Longest Residual Limb	0.76	0.59	1.23	0 ± 2.39	<0.001
Spine	0.68	0.46	1.40	0 ± 2.73	<0.001
Shortest Residual Limb	0.82	0.67	1.11	0 ± 2.15	<0.001
Model 1.1: PAEE = (0.000979 x PAC·min^{-1}) + 0.2255481					
Model 2.1: PAEE = (0.000928 x PAC·min^{-1}) + (0.027761 x Time Since Amputation[months]) + (0.663267 x Level of Injury [1 or 2]) - 1.139788					
Shortest Residual Limb	0.86	0.73	1.01	0 ± 1.91	<0.001
Bilateral Amputee Group					
Longest Residual Limb	0.80	0.64	1.56	0 ± 3.03	<0.001
Spine	0.80	0.64	1.57	0 ± 3.05	<0.001
Shortest Residual Limb	0.92	0.85	1.03	0 ± 1.99	<0.001
Model 1.2: PAEE = (0.000929 x PAC·min^{-1}) - 0.051541					
Model 2.2: PAEE = (0.000877 x PAC·min^{-1}) + (0.024560 x Waist Circumference [cm]) - 2.263715					
Shortest Residual Limb	0.94	0.88	0.93	0 ± 1.79	<0.001
Control Group					
Left Limb	0.88	0.77	0.54	0 ± 1.06	<0.001
Spine	0.87	0.75	0.57	0 ± 1.10	<0.001
Right Limb	0.87	0.76	0.56	0 ± 1.08	<0.001
Model 1.3: PAEE = (0.000776 x PAC·min^{-1}) + 0.427097					
Model 2.3: PAEE = (0.000782 x PAC·min^{-1}) + (0.033104 x Body Mass [kg]) - 2.191630					
Left Limb	0.89	0.80	0.51	0 ± 0.98	<0.001

Table 5.4: Mean absolute error (MAE); kcal·min-1) and mean absolute percentage error of predicted PAEE using generated linear regression equations for each anatomical location and the most accurate 'generated model' which uses additional covariates (Model 2)

MAE (kcal·min^{-1})

Activity	**Unilateral Amputees**				**Bilateral Amputees**				**Control**			
	Longest Limb	**Spine**	**Shortest Limb**	**Generated model**	**Longest Limb**	**Spine**	**Shortest Limb**	**Generated model**	**Left Limb**	**Spine**	**Right Limb**	**Generated model**
Resting	0.36 ± 0.01	0.79 ± 0.00	0.26 ± 0.00	0.50 ± 0.42	0.98 ± 0.00	0.24 ± 0.00	0.05 ± 0.00	0.36 ± 0.39	0.34 ± 0.00	0.44 ± 0.00	0.43 ± 0.00	0.41 ± 0.24
0.48 m.s^{-1}	0.97 ± 0.51	0.98 ± 0.59	0.83 ± 0.36	0.77 ± 0.55	1.18 ± 0.53	1.30 ± 1.13	1.04 ± 0.65	0.99 ± 0.68	0.30 ± 0.29	0.38 ± 0.22	0.32 ± 0.30	0.21 ± 0.21
0.67 m.s^{-1}	0.78 ± 0.69	0.85 ± 0.75	0.69 ± 0.50	0.62 ± 0.49	1.27 ± 0.95	1.15 ± 1.28	0.91 ± 0.54	0.80 ± 0.53	0.22 ± 0.20	0.21 ± 0.12	0.20 ± 0.18	0.20 ± 0.15
0.89 m.s^{-1}	0.77 ± 0.88	0.91 ± 1.03	0.74 ± 0.80	0.67 ± 0.52	1.49 ± 1.25	1.19 ± 1.27	0.77 ± 0.80	0.68 ± 0.57	0.28 ± 0.21	0.24 ± 0.15	0.25 ± 0.19	0.26 ± 0.16
1.12 m.s^{-1}	0.90 ± 1.17	1.08 ± 1.26	0.89 ± 1.02	0.79 ± 0.72	0.89 ± 0.47	0.91 ± 0.74	0.19 ± 0.20	0.17 ± 0.19	0.43 ± 0.32	0.41 ± 0.30	0.42 ± 0.31	0.38 ± 0.30
1.34 m.s^{-1}	1.11 ± 1.64	1.19 ± 1.72	1.23 ± 1.42	1.05 ± 1.17	1.49 ± 0.15	1.53 ± 0.90	1.16 ± 0.59	0.87 ± 0.97	0.59 ± 0.35	0.62 ± 0.39	0.53 ± 0.38	0.42 ± 0.41
3% (0.89 m.s^{-1})	0.75 ± 0.91	0.90 ± 1.10	0.86 ± 0.71	0.79 ± 0.59	1.40 ± 1.70	1.58 ± 1.66	0.66 ± 1.36	0.61 ± 0.94	0.59 ± 0.33	0.47 ± 0.33	0.44 ± 0.37	0.42 ± 0.24
5% (0.89 m.s^{-1})	0.88 ± 1.11	1.02 ± 1.29	0.75 ± 0.87	0.83 ± 0.64	0.86 ± 0.52	0.79 ± 0.07	0.43 ± 0.28	0.60 ± 0.41	0.79 ± 0.46	0.84 ± 0.43	0.83 ± 0.47	0.82 ± 0.29
All Activities	**0.80 ± 0.92**	**0.96 ± 1.00**	**0.76 ± 0.78**	**0.74 ± 0.63**	**1.22 ± 0.93**	**1.06 ± 1.14**	**0.66 ± 0.77**	**0.66 ± 0.62**	**0.42 ± 0.33**	**0.45 ± 0.33**	**0.43 ± 0.35**	**0.39 ± 0.31**

Mean absolute percentage error (%)

Activity	**Unilateral Amputees**				**Bilateral Amputees**				**Control**			
	Longest Limb	**Spine**	**Shortest Limb**	**Generated model**	**Longest Limb**	**Spine**	**Shortest Limb**	**Generated model**	**Left Limb**	**Spine**	**Right Limb**	**Generated model**
Resting	-	-	-	-	-	-	-	-	-	-	-	-
0.48 m.s^{-1}	40 ± 15.5	43 ± 24.8	36 ± 15.9	35 ± 27.1	34 ± 18.5	40 ± 37.3	30 ± 19.9	29 ± 18.9	23 ± 20.5	29 ± 22.5	24 ± 23.3	15 ± 11.1
0.67 m.s^{-1}	26 ± 16.2	29 ± 20.2	23 ± 12.0	22 ± 18.3	26 ± 14.6	26 ± 23.7	19 ± 9.0	18 ± 12.6	13 ± 14.0	12 ± 8.8	11 ± 9.7	11 ± 10.0
0.89 m.s^{-1}	21 ± 17.7	25 ± 19.9	19 ± 15.0	18 ± 12.4	26 ± 14.9	22 ± 16.7	13 ± 9.2	13 ± 10.3	12 ± 9.4	10 ± 5.6	11 ± 7.0	12 ± 7.6
1.12 m.s^{-1}	21 ± 20.2	25 ± 20.2	20 ± 16.4	18 ± 11.8	20 ± 16.3	25 ± 30.4	6 ± 8.3	3 ± 2.0	16 ± 11.2	15 ± 11.1	15 ± 11.1	14 ± 10.8
1.34 m.s^{-1}	17 ± 19.0	19 ± 20.7	20 ± 16.9	18 ± 16.5	32 ± 14.2	39 ± 38.1	22 ± 0.5	14 ± 11.2	19 ± 12.9	20 ± 14.6	17 ± 14.0	13 ± 13.9
3% (0.89 m.s^{-1})	18 ± 15.7	21 ± 18.2	20 ± 11.2	19 ± 14.2	21 ± 17.3	25 ± 17.2	8 ± 12.5	10 ± 10.4	13 ± 9.3	16 ± 9.6	15 ± 10.9	15 ± 8.0
5% (0.89 m.s^{-1})	16 ± 14.6	18 ± 17.7	14 ± 13.3	17 ± 12.7	14 ± 6.9	15 ± 7.3	10 ± 9.6	14 ± 16.1	22 ± 10.8	24 ± 9.7	23 ± 11.3	24 ± 7.3
All Activities	**23 ± 18.1**	**26 ± 20.9**	**22 ± 15.2**	**21 ± 17.2**	**26 ± 15.9**	**28 ± 24.9**	**17 ± 14.4**	**16 ± 14.7**	**17 ± 13.2**	**18 ± 13.7**	**17 ± 13.8**	**15 ± 7.3**

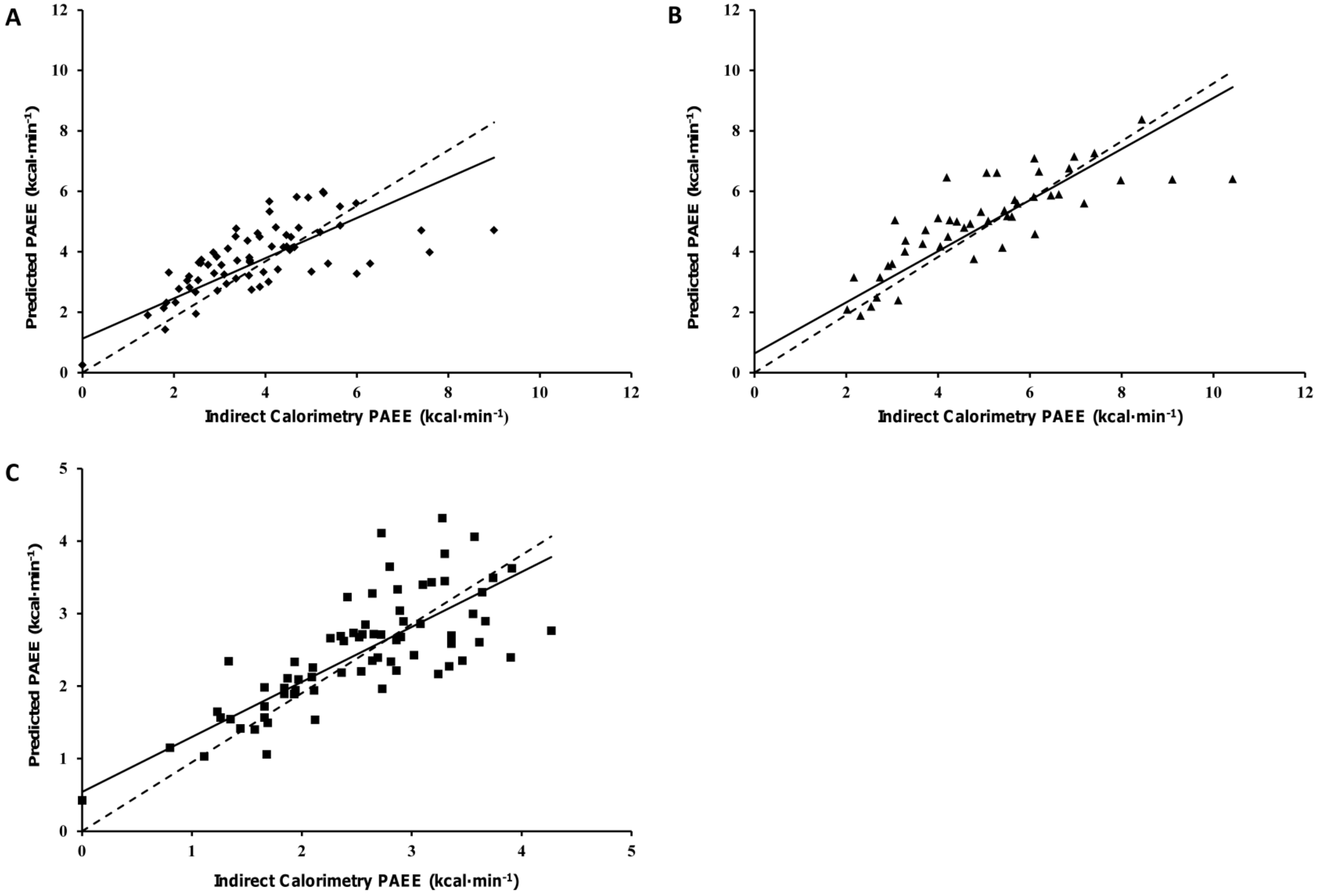


Figure 5.1: Scatterplots showing the relationship between predicted PAEE for the GT3X+ worn on the hip of the shortest limb and criterion PAEE (Model 1). (A) unilateral amputee group, (B) bilateral amputee group and (C) the left hip of the uninjured control group. The straight lines represents the models best fit, and the dotted line indicates the line of identity

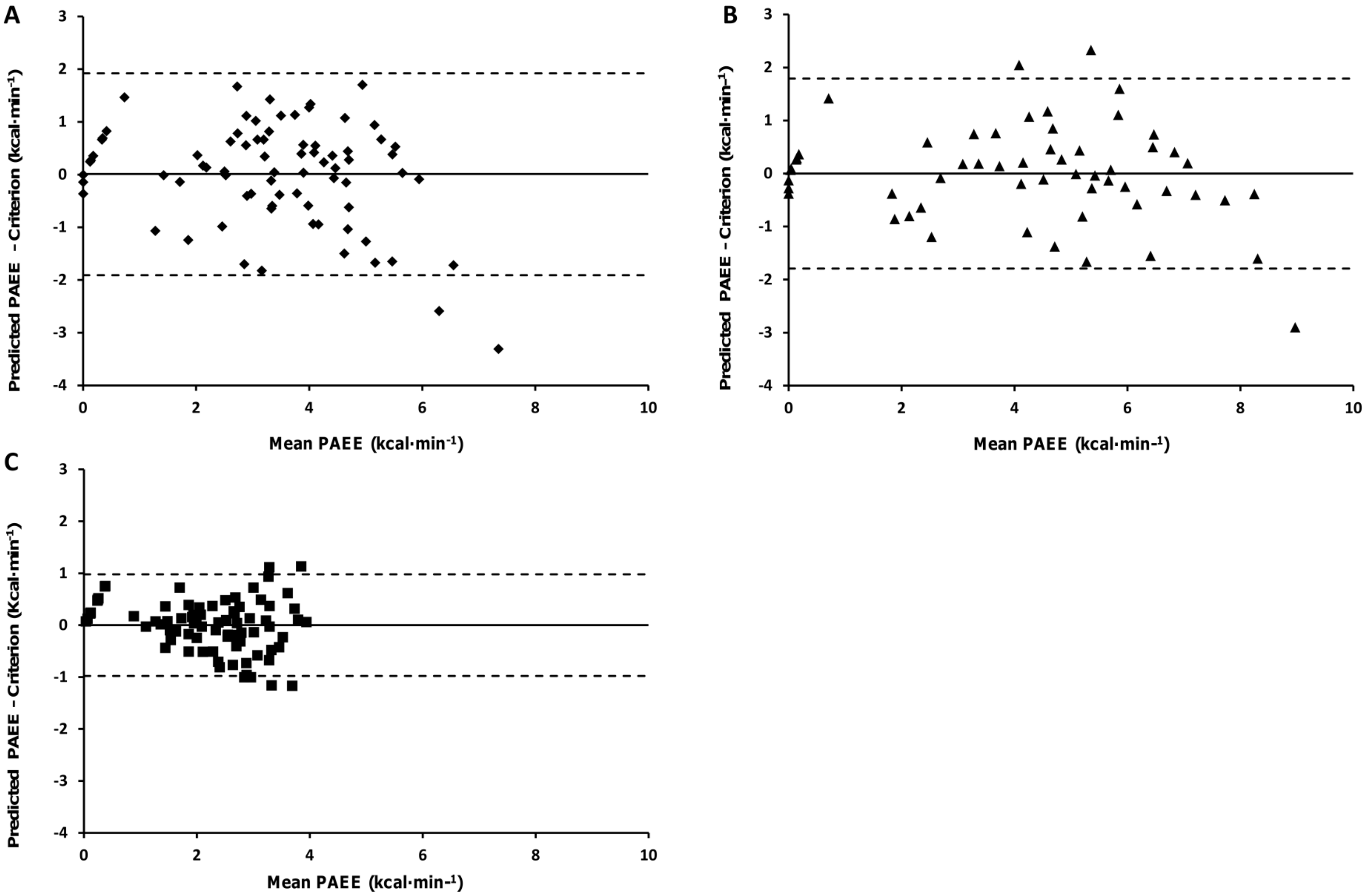


Figure 5.2: Bland and Altman plots for the criterion and predicted PAEE using cross-validated, population specific prediction models (Model 2). Developed for the unilateral group (A), bilateral group (B) and control group (C). The straight line demonstrates the mean and the dotted line indicates the 95% limits of agreement (LoA)

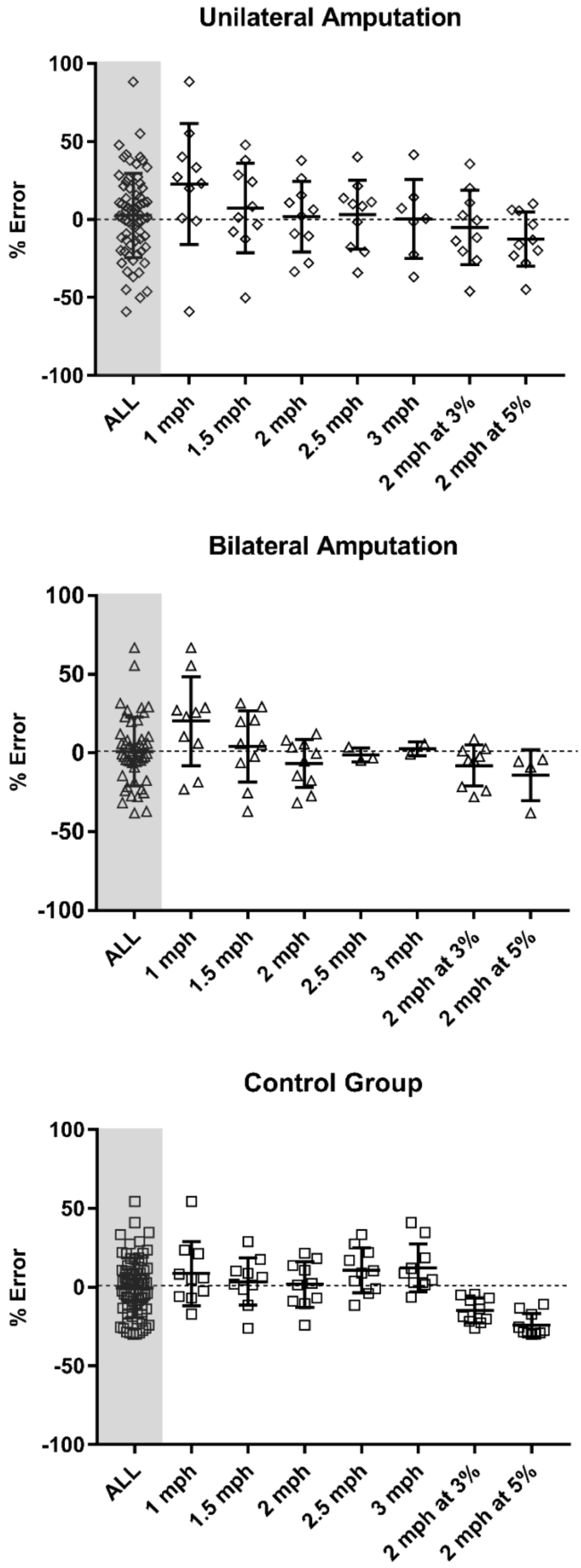


Figure 5.3: Modified box and whisker plots demonstrating the mean percentage error of estimation relative to criterion for each treadmill activity using the cross validated, population specific prediction models (Model 2). The plots show the unilateral group (A), bilateral group (B) and control group (C).

5.3.2. Experiment Two

Demographic and anthropometric characteristics of the participants are described in table 5.5. Two patient's data were removed from these analyses. One due to a mild skin reaction to the electrodes of the AHR requiringremoval, and one due to unreliable data from the AHR due to poor connectivity. Criterion PAEE (kcal·min^{-1}), GT3X+ accelerometer outputs, HR, AHR, and METs are displayed in table 2. Mean criterion PAEE, GT3X+ PAC, HR, AHR PAEE, RPE and METs all increased with increasing treadmill velocity in the unilateral and control groups. In the bilateral group, between six and eight participants were unable to complete activities at the higher treadmill velocities and gradients, three individuals in the unilateral group were unable to complete the highest ambulatory velocity (1.34 m.s^{-1}) which influenced the mean criterion values. There was a significant main effect in criterion PAEE, GT3X+ predicted PAEE, AHR, HR and METs in all groups (table 5.6). In both amputation groups and control participants, the GT3X+HR model demonstrated the strongest relationships, smallest LoA (table 5.7) and mean absolute percentage errors compared to AHR (table 5.8).

Across all treadmill walking tasks, the HR response in individuals with unilateral amputation was 1.3 to 1.5 times greater than the physically active control group. At treadmill speeds between 0.48 and 0.89 m.s^{-1} (1 to 2 mph) and a gradient of 3% at 2 mph, the HR response of individuals with bilateral amputation was 1.5 to 1.7 times greater than control participants. At these same walking speeds the HR of the bilateral amputation group was between 1.1 to 1.3 times greater than the group with unilateral amputation. These differences in HR increase linearly with increases in treadmill intensity (table 4.6). The population specific equations (GT3X+HR) to predict ambulatory PAEE are below:

Unilateral amputation: $PAEE = (0.000453*PAC) + (0.045487*HR) - 2.713284$

Bilateral amputation: $PAEE = (0.000658*PAC) + (0.025308*HR) - 1.795157$

Controls: $PAEE = (0.000550*PAC) + (0.036472*HR) - 1.797866$

Table 5.5: Demographic and anthropometric characteristics of the participants

Variable	Unilateral		Bilateral		Control	
	Mean ± SD	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**
Number of Participants	9		10		9	
Age (years)	32 ± 5	23 - 41	29 ± 4	22 - 34	31 ± 6	25 - 45
Body Mass - without prosthesis (kg)	81 ± 11	63 - 108	82 ± 19	59 - 126	80 ± 7	68 - 89
Waist Circumference (cm) *	92 ± 13	75 - 115	100 ± 20	77 - 149	83 ± 4	76 - 90
Waist-hip ratio	0.90 ± 0.07	0.83 - 1.00	0.94 ± 0.09	0.86 - 1.17	0.86 ± 0.04	0.79 - 0.92
RMR ($kcal \cdot d^{-1}$)	1776 ± 269	1480 - 2158	1596 ± 178	1382 - 2051	1846 ± 191	1505 - 2059
Sleeping Heart Rate #	63 ± 9	46 - 77	57 ± 7	48 - 68	48 ± 3	45 - 55
Time Since Amputation (months) ǂ	23 ± 15	4 - 46	39 ± 14	21 - 61	-	
Level of Amputation:						
Below Knee	5		1		-	
Through Knee	2		2		-	
Above knee	2		3		-	
Bilateral: Below Knee and Above Knee	-		4		-	

*Significant difference between individuals with bilateral amputation and control group ($p < 0.05$)

ǂ Significant difference between individuals with unilateral and bilateral amputation ($p < 0.05$)

Significant difference between control group and both amputation groups ($p < 0.05$)

Table 5.6: Measured PAEE, accelerometer outputs at each anatomical location, calculated METs, RPE and number of participants for each activity (mean ± SD)

Activity	PAEE Metamax 3B (kcal·min-1)	GT3x+ (PAC·min-1)	HR	AHR (kcal·min-1)	METS (calculated)	RPE	RER	n
Unilateral Amputation: ǂ #								
RMR	0.00 ± 0.00	0 ± 0	66 ± 11	0.00 ± 0.00	1.0 ± 0.0	6 ± 0	0.74 ± 0.04	9
Treadmill 0.48 m.s-1	2.40 ± 0.71	2643 ± 866	96 ± 17	1.03 ± 0.52	3.1 ± 0.8	8 ± 1	0.76 ± 0.04	9
Treadmill 0.67 m.s-1	3.00 ± 0.95	2939 ± 908	101 ± 18	1.61 ± 0.74	3.6 ± 1.0	9 ± 1	0.79 ± 0.04	9
Treadmill 0.89 m.s-1	3.61 ± 1.12	3353 ± 892	106 ± 21	2.15 ± 1.05	4.1 ± 1.1	11 ± 2	0.81 ± 0.03	9
Treadmill 1.12 m.s-1	4.39 ± 1.42	4107 ± 707	112 ± 24	2.84 ± 1.93	4.7 ± 1.3	12 ± 2	0.83 ± 0.03	9
Treadmill 1.34 m.s-1	5.89 ± 1.70	4977 ± 581	126 ± 36	4.01 ± 3.32	5.7 ± 1.8	12 ± 2	0.86 ± 0.05	6
Treadmill 3% (0.89 m.s-1)	4.17 ± 1.06	3642 ± 981	111 ± 21	2.32 ± 1.15	4.5 ± 1.1	11 ± 1	0.82 ± 0.04	9
Treadmill 5% (0.89 m.s-1)	4.82 ± 1.24	4020 ± 1005	119 ± 23	2.94 ± 2.09	5.0 ± 1.2	12 ± 2	0.84 ± 0.04	9
Bilateral Amputation:* ¥								
RMR	0.00 ± 0.00	0 ± 0	67 ± 10	0.00 ± 0.00	1.0 ± 0.0	6 ± 0	0.75 ± 0.04	10
Treadmill 0.48 m.s-1	3.72 ± 1.37	4800 ± 1410	108 ± 13	2.41 ± 1.40	4.4 ± 1.2	10 ± 2	0.78 ± 0.03	10
Treadmill 0.67 m.s-1	4.59 ± 1.54	5264 ± 1603	121 ± 15	3.38 ± 1.96	5.1 ± 1.4	12 ± 2	0.81 ± 0.04	10
Treadmill 0.89 m.s-1	5.46 ± 1.74	5600 ± 1502	133 ± 18	4.86 ± 2.81	5.8 ± 1.6	14 ± 3	0.84 ± 0.05	10
Treadmill 1.12 m.s-1	5.54 ± 2.85	6123 ± 2823	139 ± 31	7.80 ± 5.00	5.3 ± 1.4	15 ± 3	0.91 ± 0.18	3
Treadmill 1.34 m.s-1	5.23 ± 2.76	5235 ± 1212	142 ± 41	7.31 ± 4.14	5.7 ± 1.7	15 ± 0	0.86 ± 0.7	2
Treadmill 3% (0.89 m.s-1)	5.93 ± 2.29	5973 ± 1592	138 ± 19	5.95 ± 4.25	6.1 ± 2	14 ± 3	0.87 ± 0.7	8
Treadmill 5% (0.89 m.s-1)	5.77 ± 1.85	5806 ± 2231	146 ± 30	7.80 ± 5.26	5.7 ± 1.1	16 ± 2	0.93 ± 0.17	4

Control:								
RMR	0.00 ± 0.00	0 ± 0	54 ± 4	0.00 ± 0.00	1.0 ± 0.0	6 ± 0	0.75 ± 0.04	9
Treadmill 0.48 m.s-1	1.43 ± 0.31	1299 ± 411	72 ± 7	1.34 ± 0.54	2.2 ± 0.3	7 ± 0	0.78 ± 0.03	9
Treadmill 0.67 m.s-1	1.81 ± 0.32	1811 ± 368	74 ± 6	1.88 ± 0.65	2.5 ± 0.3	8 ± 1	0.82 ± 0.04	9
Treadmill 0.89 m.s-1	2.32 ± 0.40	2430 ± 459	78 ± 4	2.22 ± 0.84	2.9 ± 0.4	9 ± 1	0.82 ± 0.05	9
Treadmill 1.12 m.s-1	2.80 ± 0.43	3325 ± 480	82 ± 5	2.55 ± 0.84	3.3 ± 0.5	9 ± 1	0.82 ± 0.03	9
Treadmill 1.34 m.s-1	3.40 ± 0.34	4144 ± 457	85 ± 5	2.91 ± 0.88	3.7 ± 0.4	10 ± 1	0.83 ± 0.04	9
Treadmill 3% (0.89 m.s-1)	2.88 ± 0.38	2551 ± 385	83 ± 5	2.38 ± 0.92	3.3 ± 0.3	10 ± 1	0.84 ± 0.04	9
Treadmill 5% (0.89 m.s-1)	3.45 ± 0.43	2720 ± 311	87 ± 4	2.52 ± 0.99	3.8 ± 0.3	10 ± 1	0.83 ± 0.04	9

Abbreviations: PAEE = physical activity energy expenditure, HR = heart rate, AHR = Actiheart, METs = metabolic equivalent, RPE = rate of perceived exertion, RER = respiratory exchange ratio, RMR = resting metabolic rate.

Not all participants with amputation/s were able to complete all of the treadmill speeds in this trial. The number of participant completers at each treadmill task is presented here.

*Due to reduced participant numbers, all statistical analyses comparing the bilateral group with other groups were performed at speeds 0.48-0.89 m.s^{-1} and at 3% gradient.

PAC from the GT3X+ combined with HR, HR alone and AHR data were all significantly ($p < 0.01$) associated with criterion PAEE

ǂ A significant difference in criterion PAEE, HR and METs were only reported at higher intensities (1.12 m.s^{-1}, 1.34 m.s^{-1} and 5% gradient at 0.89 m.s^{-1}) between individuals with unilateral amputation and control group ($p < 0.05$).

Significant differences in PAC (GT3X+) were only reported at the lowest intensity of 0.48 m.s^{-1} and the highest intensity of 1.34 m.s^{-1} between individuals with unilateral amputation and control group ($p < 0.05$).

¥ Significant differences in AHR outcomes were reported between the bilateral amputation and the unilateral amputation groups at all speeds analysed and at 0.89 m.s^{-1} and 3% gradient at 0.89 m.s^{-1} versus the control group.

Table 5.7: The relationships between the three devices of the Actigraph GT3X+ with HR, HR alone AHR against criterion PAEE in all groups. Limits of agreement (LoA) expressed as mean ± 95% SD

Location	*r*	R^2	SEE (kcal·min^{-1})	LoA (kcal·min^{-1})	*P* Value
Treadmill Walking					
Unilateral Amputation Group					
GT3X+ and HR	0.92	0.85	0.78	0 ± 1.50	< 0.001
HR	0.89	0.79	0.91	0 ± 1.77	< 0.001
AHR	0.86	0.73	1.02	-1.40 ± 2.00	< 0.001
Bilateral Amputation Group					
GT3X+ and HR	0.93	0.87	0.96	0 ± 1.84	< 0.001
HR	0.88	0.77	1.26	0 ± 2.44	< 0.001
AHR	0.81	0.65	1.53	0.21 ± 4.28	< 0.001
Control Group					
GT3X+ and HR	0.91	0.83	0.48	0 ± 0.93	< 0.001
HR	0.84	0.71	0.62	0 ± 1.82	< 0.001
AHR	0.67	0.45	0.85	0.29 ± 1.82	< 0.001

Table 5.8: Mean absolute error (MAE); kcal·min-1) and mean absolute percentage error of predicted PAEE using the generated models of the GT3X+ with HR, HR signals alone and AHR. Data expressed as mean ± SD

	MAE (kcal·min^{-1})								
Activity	**Unilateral Amputees**			**Bilateral Amputees**			**Control**		
	GT3X+ and HR	**HR**	**AHR**	**GT3X+ and HR**	**HR**	**AHR**	**GT3X+ and HR**	**HR**	**AHR**
Resting	0.53 ± 0.28	1.00 ± 0.73	0.00 ± 0.00	0.22 ± 0.16	0.70 ± 0.50	0.00 ± 0.00	0.20 ± 0.11	0.35 ± 0.24	0.00 ± 0.00
0.48 m.s^{-1}	0.61 ± 0.51	0.78 ± 0.49	1.37 ± 0.41	0.94 ± 0.53	1.06 ± 0.75	1.42 ± 1.06	0.30 ± 0.28	0.51 ± 0.50	0.55 ± 0.36
0.67 m.s^{-1}	0.56 ± 0.47	0.59 ± 0.46	1.40 ± 0.59	0.76 ± 0.55	0.93 ± 0.83	1.36 ± 1.08	0.25 ± 0.22	0.41 ± 0.36	0.66 ± 0.38
0.89 m.s^{-1}	0.59 ± 0.38	0.47 ± 0.46	1.46 ± 0.62	0.69 ± 0.81	1.08 ± 1.06	1.99 ± 1.04	0.30 ± 0.21	0.43 ± 0.30	0.92 ± 0.39
1.12 m.s^{-1}	0.65 ± 0.44	0.64 ± 0.50	1.55 ± 0.93	0.29 ± 0.15	0.79 ± 0.30	2.26 ± 2.32	0.41 ± 0.29	0.40 ± 0.45	0.99 ± 0.49
1.34 m.s^{-1}	0.87 ± 0.49	1.11 ± 0.54	2.37 ± 1.04	0.66 ± 0.02	0.69 ± 0.19	2.08 ± 1.37	0.47 ± 0.16	0.57 ± 0.47	0.89 ± 0.58
3% gradient at 0.89 m.s^{-1}	0.67 ± 0.42	0.58 ± 0.55	1.85 ± 0.70	0.75 ± 1.29	1.06 ± 1.25	2.23 ± 1.38	0.34 ± 0.41	0.51 ± 0.37	0.94 ± 0.62
5% gradient At 0.89 m.s^{-1}	0.73 ± 0.32	0.69 ± 0.47	1.97 ± 1.01	0.47 ± 0.10	0.92 ± 0.39	3.51 ± 2.72	0.60 ± 0.49	0.55 ± 0.50	1.05 ± 0.99
All Activities	**0.64 ± 0.41**	**0.72 ± 0.54**	**1.46 ± 0.94**	**0.64 ± 0.69**	**0.94 ± 0.81**	**1.59 ± 1.50**	**0.36 ± 0.31**	**0.47 ± 0.40**	**0.75 ± 0.61**

	Mean absolute percentage error (%)								
Activity	**Unilateral Amputees**			**Bilateral Amputees**			**Control**		
	GT3X+ and HR	**HR**	**AHR**	**GT3X+ and HR**	**HR**	**AHR**	**GT3X+ and HR**	**HR**	**AHR**
Resting	-	-	-	-	-	-	-	-	-
0.48 m.s^{-1}	26 ± 21.6	35 ± 21.5	58 ± 15.2	26 ± 14.3	27 ± 13.7	39 ± 26.7	24 ± 27.0	42 ± 54.6	41 ± 29.6
0.67 m.s^{-1}	19 ± 16.6	21 ± 18.1	47 ± 19.1	16 ± 7.9	19 ± 11.9	33 ± 27.4	14 ± 13.1	23 ± 24.7	37 ± 24.1
0.89 m.s^{-1}	17 ± 10.4	15 ± 15.6	41 ± 18.6	11 ± 8.3	18 ± 11.8	38 ± 20.4	13 ± 8.8	19 ± 14.9	40 ± 14.4
1.12 m.s^{-1}	15 ± 8.0	16 ± 14.4	38 ± 22.5	7 ± 6.2	15 ± 3.6	37 ± 21.5	15 ± 9.4	14 ± 13.7	35 ± 13.1
1.34 m.s^{-1}	15 ± 7.1	21 ± 14.4	43 ± 21.3	15 ± 8.1	14 ± 3.8	38 ± 6.1	14 ± 5.0	16 ± 12.1	27 ± 16.9
3% gradient at 0.89 m.s^{-1}	16 ± 11.7	16 ± 15.75	46 ± 18.9	10 ± 12.1	16 ± 11.2	38 ± 21.1	11 ± 12.0	17 ± 10.4	33 ± 20.6
5% gradient at 0.89 m.s^{-1}	15 ± 7.9	14 ± 12.1	44 ± 23.7	9 ± 3.5	16 ± 4.3	55 ± 31.2	16 ± 11.7	15 ± 11.6	30 ± 27.6
All Activities	**18 ± 13.9**	**20 ± 16.9**	**45 ± 19.9**	**15 ± 11.5**	**19 ± 11.5**	**39 ± 23.4**	**15 ± 13.9**	**21 ± 25.5**	**34 ± 27.6**

The relationships between criterion PAEE and predicted PAEE, derived from GT3X+HR and AHR, are presented as scatter plots in figure 5.4. Figure 5.5 illustrates the mean bias and 95% LoA differences, when comparing the criterion PAEE data with estimated PAEE derived from population specific prediction models (GT3X+HR) and the proprietary group calibration algorithm of the AHR. Figure 5.6 demonstrates modified box and whisker plots depicting the mean percentage error of estimation relative to criterion for each treadmill activity using the cross validated, GT3X+HR model against the pre-determined algorithm used in the AHR device.

When comparing both methods across all groups the GT3X+HR demonstrates the smallest LoA and the AHR shows the greatest LoA. When comparing populations, the control group demonstrate the smallest LoA and the bilateral group demonstrate the largest LOA for both GT3X+HR and AHR methods (table 5.7).

Participants were asked to complete 35 minutes of walking in total. Participants reasoning for discontinuing with the trial were not asked. However, a large proportion of participants stated an uncomfortable stump due to repeated impact and friction inside their socket. For individuals with transfemoral bilateral amputation the inability to walk quickly enough in their prosthesis without the risk of falling was also reported. Discontinuing with the trial due to excessive physical exertion or fatigue was less common, as demonstrated by the RPE scores in table 5.6. The respiratory exchange ratio (RER) values during the walking tasked range from a mean 0.76 ± 0.04 to 0.86 ± 0.05 and 0.78 ± 0.04 to 0.93 ± 0.02 in the unilateral and bilateral amputation groups respectively. RER values ranged from 0.78 ± 0.03 to 0.84 ± 0.04 in the control group.

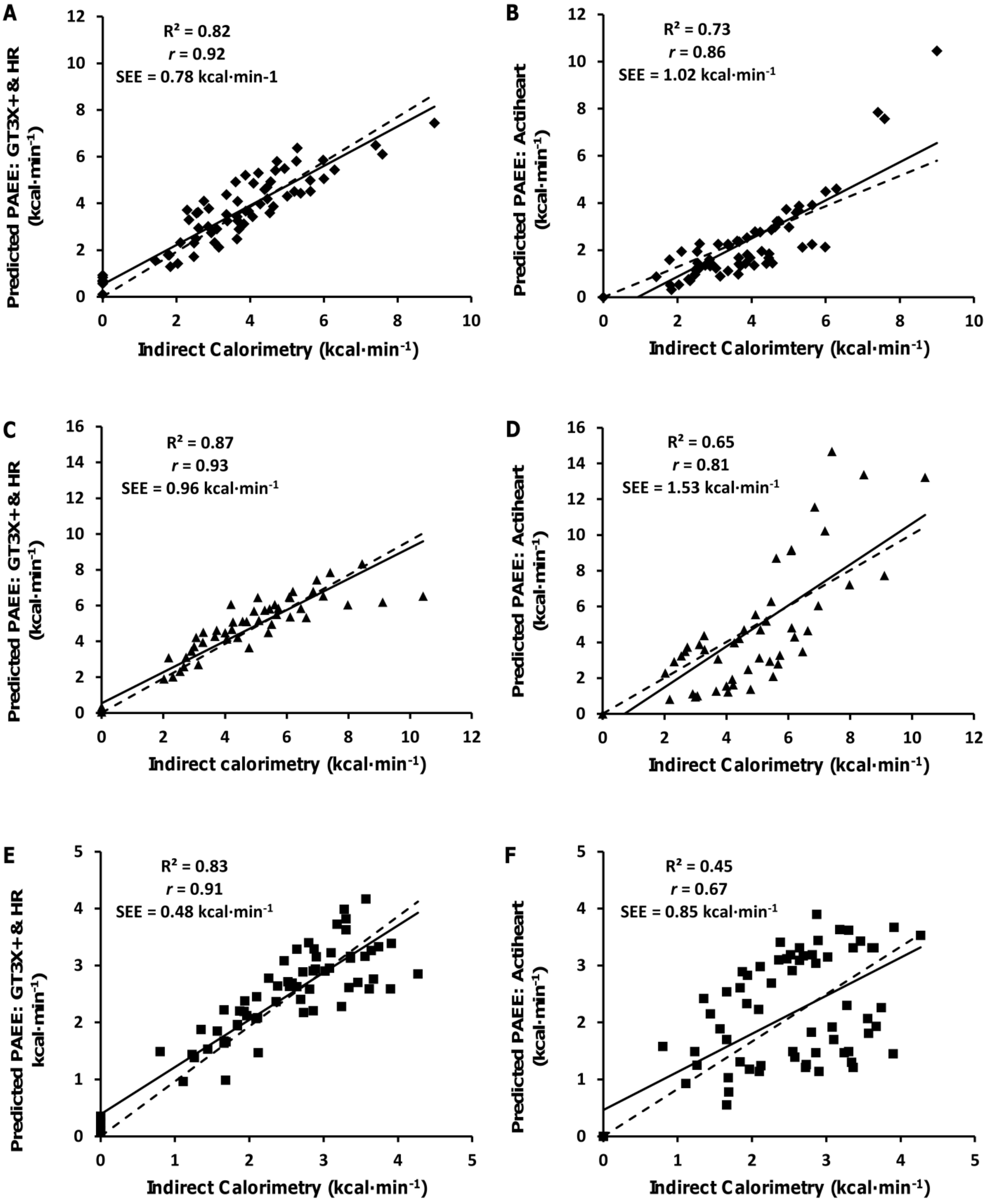


Figure 5.4: Scatterplots showing the relationship between estimated and criterion PAEE. Estimated PAEE from GT3X+ with heart rate and criterion PAEE (A, C, E). Estimated PAEE from the AHR and criterion PAEE (B, D, F). The scatterplots show the unilateral amputation group (A, B), bilateral amputation group (C, D) and the uninjured control group (E, F). The straight line represents the models best fit, and the dotted line indicates the line of identity.

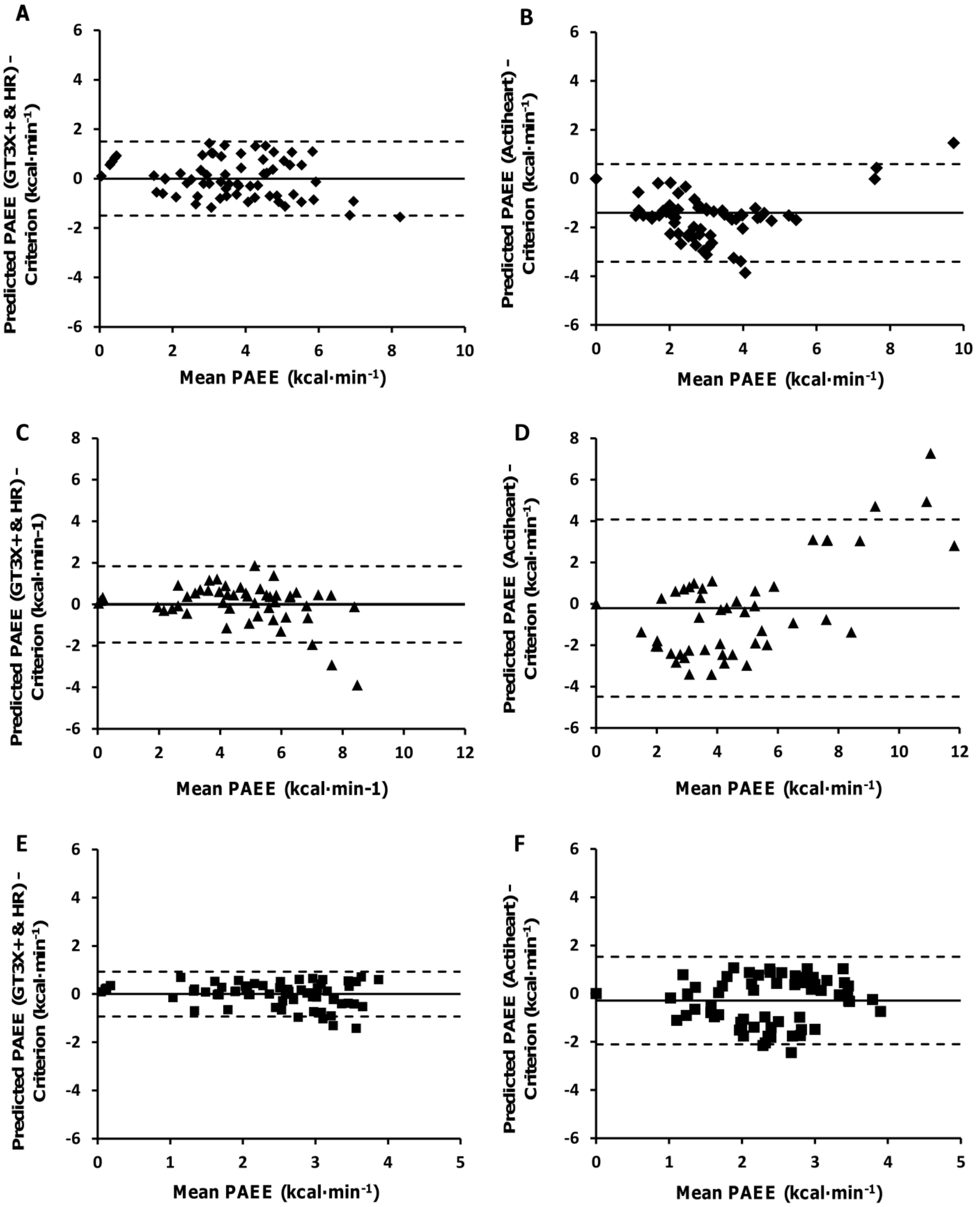


Figure 5.5: Bland and Altman plots for the criterion and predicted PAEE using the GT3X+ with heart rate estimation models (A, C, E), and criterion and predicted PAEE using the AHR (B, D, F). The plots show the unilateral group (A, B), bilateral group (C, D) and uninjured control group (E, F). The straight line demonstrates the mean and the dotted line indicates the 95% Limits of Agreement (LoA).

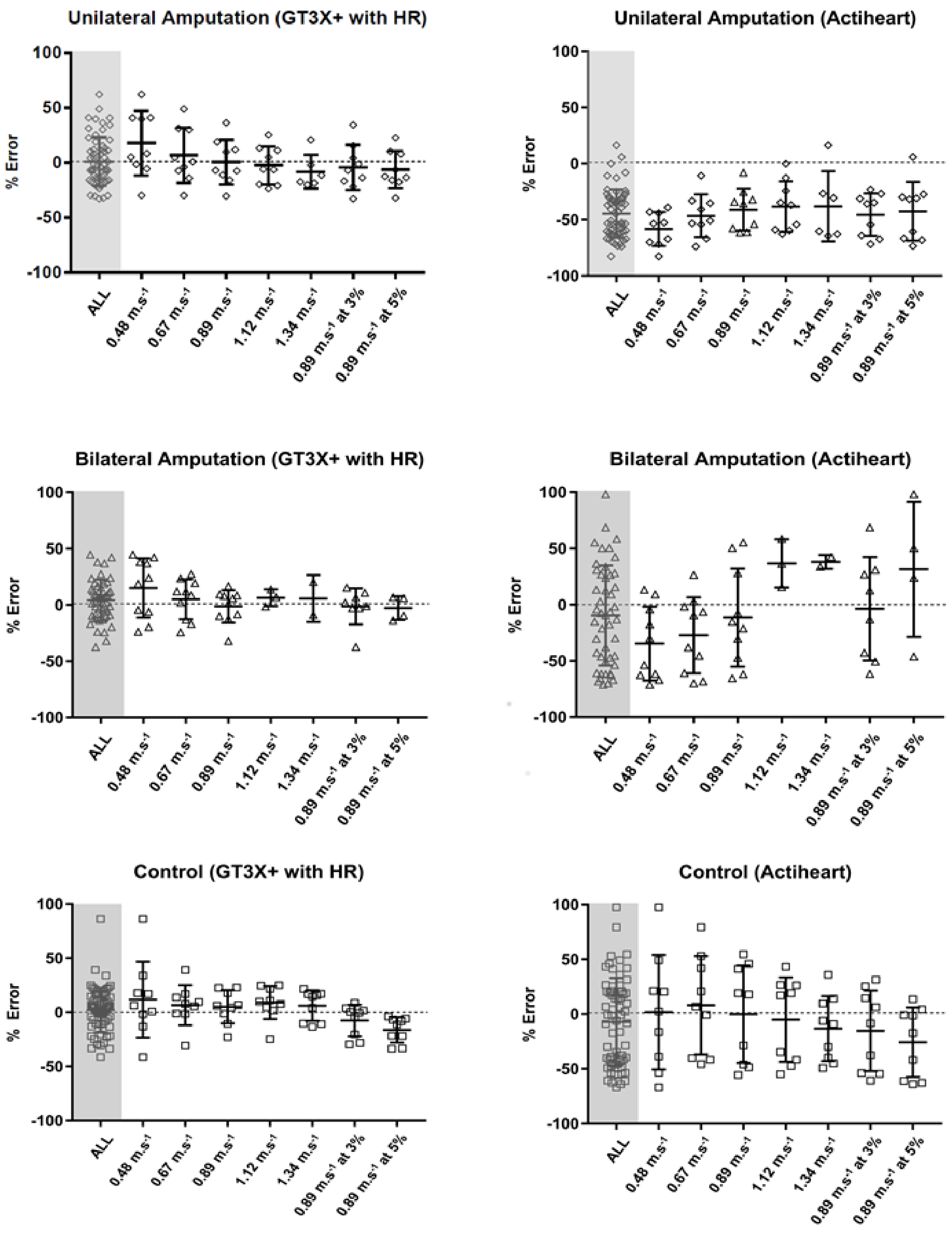


Figure 5.6: Modified box and whisker plots demonstrating the mean percentage error of estimation relative to criterion for each treadmill activity using GT3X+ with heart rate estimation models (A, C, E), and AHR (B, D, F). The plots show the unilateral group (A, B), bilateral group (C, D) and control group (E, F).

5.4. DISCUSSION

To my knowledge, this is the first accelerometer-based validation study to predict ambulatory PAEE in individuals with lower-limb amputation(s). This is also the first controlled-laboratory trial to (i) determine the most appropriate anatomical wear location for a tri-axial accelerometer, (ii) assess the accuracy of a research grade multi-sensor device (AHR) and (iii) develop population specific manually derived equation, cross-validated across a range of ambulatory velocities. The results demonstrate that the anatomical position of accelerometers is an important consideration when assessing PAEE in people with LLA and, potentially, other functional limitations. Of the three anatomical locations considered in this study, the results indicate that the GT3X+ worn on the hip of the shortest residual limb elicits the strongest correlations with criterion PAEE, explaining the greatest amount of variance and displayed the lowest random error in traumatic lower-limb amputees. Of the two multi-sensor activity methods considered in this study, these data indicate the GT3X+HR model, integrating accelerometry data from an Actigraph GT3X+ worn at the hip of the shortest residual limb, with HR data, provided the most valid estimation of ambulatory PAEE in both amputation groups. HR signals alone produced superior accuracy for the estimation of PAEE than the AHR device. Therefore, the proprietary group calibration algorithm, intrinsic to the AHR, has lower validity when predicting PAEE in individuals with unilateral or bilateral transtibial and transfemoral amputations, potentially because the algorithms are not optimized for this population.

One explanation for the lower random error associated with wearing the GT3X+ accelerometer on the shortest residual limb, as opposed to the spine or longest residual limb, is likely due to the increased ability to detect atypical movements (PAC) on that side of the body. It was previously reported that pelvic range of motion in the frontal plane is typically increased following LLA compared to uninjured controls (Su et al, 2007). Amputees often lift their pelvis on the swing side while walking. This compensatory motion, known as hip hiking, is often seen in both trans-tibial and trans-femoral amputees and is believed to compensate for the inability to produce dorsi-flexion in the prosthetic ankle (Sagawa et al, 2011). Hip hiking increases the prosthetic foot clearance (Su et al, 2007), but may also be associated with additional metabolic cost of raising the body centre of mass, thus reducing gait efficiency. Individuals with bilateral amputations may display bilateral hip hiking on both sides, which appears to further increase the energy cost of locomotion (Sagawa et al, 2011). The increase of hip hiking and exaggerated movements on the shortest residual limb may explain the increase in PAC recorded on this GT3X+ accelerometer. However, it was beyond the scope of this study to verify this hypothesis by performing kinematic and/or kinetic analyses of amputee gait. The biomechanics of lower-limb amputee gait have been reviewed elsewhere (Sagawa et al, 2011).

Bilateral amputees typically demonstrate lower levels of physical function than unilateral amputees. Therefore, at the highest ambulation velocities, they have higher PAEEs. The stronger correlation in the bilateral amputee cohort in the current study is likely to be an artefact of there being a wider range of functional mobility in this group as demonstrated in chapter 4 (Ladlow et al, 2015), which may explain the larger range of PAEE. Due to the drop out of participants, only the bilateral amputees who had better ambulatory efficiency were able to perform the higher velocities. This could explain why there was reduced error in the bilateral amputee group compared to the unilateral amputee groups.

The inclusion of the covariates in the predictive models provided small but significant improvements in the variance explained in PAEE (see table 5.3). The preservation of the knee joint and the utilisation of the knee and hip musculature when ambulating appear to provide a significant functional advantage in unilateral amputees (Ladlow et al, 2016). This is evident from the regression analysis that found the level of amputation is a significant predictor of PAEE in the unilateral amputee group. Time since amputation was also significantly associated with PAEE in the unilateral amputee group. It appears that the longer the time since limb-loss, the better the efficiency of ambulating with a prosthesis. All amputee patients in this study have been engaged in the DMS rehabilitation pathway at DMRC, Headley Court since the point of injury (see chapter 3 combat casualty care pathway for more details). This suggests that the longer the recovery time in conjunction with access to intensive exercise rehabilitation, the greater the opportunity to maximise physical function and prosthetic use.

Waist circumference was a significant predictor of PAEE in both amputee groups. Greater waist circumference was associated with increased PAEE when walking at a variety of speeds. Although body mass significantly predicted PAEE in the bilateral amputees it was not included in the predictive model due to the stronger influence of the waist circumference measurement on PAEE. One explanation for this is that total body mass is unable to differentiate between lean and adipose tissue, whereas, waist circumference gives an indication of central adiposity, suggesting the excess body mass in the bilateral amputees group may in fact be increased adipose tissue and not lean skeletal muscle tissue that could contribute to greater strength capabilities and increased levels of function. Gaunaurd et al. 2013 investigated factors relating to high level mobility in US military servicemen with LLA and also found these same covariates were predictors of high mobility.

HR has benefits as a physiological variable as it increases linearly and proportionately with exercise intensity and thus metabolic rate (Chen et al, 2012). HR alone in this study explains 79%, 77% and 71% of the variance in the unilateral, bilateral and control group respectively. The reason HR may have performed so well is because it is highly individualised to the groups of interest and the task intensities were linear in nature. As HR at lower exercise intensities is affected by other factors, such as psychological or thermal stress, it would be intuitive to hypothesise that the integration of acceleration values may offer a more reliable estimation of PAEE. The findings of this current study demonstrate evidence of this when combined with the PAC from the GT3X+. However, the AHR, a multi-sensor device which uses proprietary group calibration algorithms intrinsic to the device, explains less of the variance and demonstrates greater error in the estimation of ambulatory PAEE than HR alone. The proprietary equations, derived from Brage et al. (2004) and utilised here were designed to predict EE during ambulation in able-bodied healthy adults, not those with lower-limb impairments or significant gait abnormalities. The AHR uses a uni-axial accelerometer unlike the GT3X+ which is a tri-axial accelerometer, uni-axial accelerometers have been shown to have less sensitivity when predicting PAEE (Van Remoortel et al, 2012). It is also known that the anatomical location of an accelerometer is important in the accuracy of measuring PAEE (Horner et al, 2013), as evident from experiment one with the positioning of the G3X+ around the pelvis. The anatomical locations of the monitors used in this study are not comparable, as the AHR device was worn on the chest whilst the GT3X+ was worn on the hip of the shortest residual limb. It may be that there is reduced sensitivity with accelerometers being worn at the chest compared to hip during ambulatory movements. The activity protocol adopted in this study captured a wide range of walking speeds, which are representative of the exercise intensities for LLA groups undergoing in-patient rehabilitation. The relative exercise intensities were consequently much lower for healthy uninjured controls (table 5.2 and 5.6). The weaker correlations in the control group compared to the amputation groups may be an artefact of there being a narrower range of exercise intensities. Although not necessarily an accurate reflection of the general population, they provide normative values for physically active military personnel, thereby allowing us to draw closer comparisons to the groups of individuals with an amputation(s) pre-injury and peer-group PAEE values.

When activity counts from the GT3X+ were combined with significant covariates this created population specific estimation models, capable of accurately estimating ambulatory PAEE (mean absolute percentage error); unilateral (21 ± 17%), bilateral (16 ± 15%) and controls (15 ± 7%). When compared to the PAEE predictive models which incorporated a physiological variable (HR) with the GT3X+ monitor (table 5.8) in the unilateral group created stronger correlations, explained greater amounts of variance and displayed lower random error than the GT3X+ with covariates (level of amputation and length of rehabilitation). In the bilateral amputation and control groups the

GT3X+HR model and the GT3X+ with covariates models (bilateral amputation group covariates; waist circumference, and control; body mass) were very similar.

5.4.1. Limitations

A limitation of this study is the relatively small sample size and variations within participants based on the diversity in the severity of lower-limb injuries. However, this diversity may be considered beneficial as the range of walking abilities captured improves the external validity of the regression equations, making them more suitable for the wider amputee population. Also, despite the diversity of the population, the amount of unexplained random error is relatively small. The inclusion of a diverse range of participants is in accordance with best practice recommendations for PA validation studies (Bassett et al, 2012). I recognise that the amputee cohort has received intensive rehabilitation unique to the UK military as described in chapter 3 (section 3.4) and this is likely to have an effect on their walking efficiency. As a population group, military personnel are predominantly male, aged 20-40 years of age and have undergone physical training in the course of their career. Although some of the mechanisms of injury (e.g. blast injuries) may be different to a civilian population, the types of injuries sustained by some of the unilateral amputee cohort are not too dissimilar from what might be expected in RTA (e.g. motorbike) and some adventure sports. Therefore, we believe that these findings are applicable to the physically active civilian LLA population.

It is important to note that participants were not provided with a familiarisation of treadmill walking prior to starting the trial. However, many would have been exposed to treadmill walking/running as part of their rehabilitation or as their preferred mode of cardiorespiratory exercise whilst at home. It is acknowledged that for some individuals (primarily the bilateral amputation group) this may have affected the EE data due to the lack of familiarity with the exercise task. However, it is likely that the GT3X+ still managed to assess increased movement due to atypical gait, thus increased corresponding GT3X+ and HR outputs. This study did not measure self-selected walking speed over-ground. There may have been differences in the energy cost of walking over ground compared with treadmill walking (Traballesi et al, 2008), therefore potentially reducing the accuracy of using these generated equations in the assessment of free-living ambulation. Empirical evidence suggest that civilian bilateral amputees are likely to be reliant on a wheelchair to mobilise during free-living conditions, therefore bilateral amputees from the wider civilian population may be unlikely to complete this treadmill protocol due to the intensity and duration spent in ambulation. Even within this well-trained military population, there was a drop-out of bilateral amputees at the higher ambulatory velocities (see table 5.2). This reduction in participant numbers may negatively affect the accuracy of the prediction models in this group and explain the reduced error in the bilateral amputation group when compared to the unilateral lower-limb amputation group. It was felt the inclusion of bilateral amputees who had undergone intensive rehabilitation was important to allow us to make comparisons with unilateral amputees and uninjured controls. This study design allows greater insight into the energy cost of walking in a wider cohort of severely injured individuals.

It is perhaps unsurprising that a bespoke algorithm, developed for this population specifically, performed better than an algorithm that was generated on a completed independent and physiologically different sample. Nevertheless, if the AHR device is to be applicable to various clinical populations with atypical gait patterns and asymmetries, alternative proprietary equations and/or independent HR calibrations appear necessary. The measurement of PAEE has proven inherently difficult to measure, even in humans without mobility-related physical impairments. Commercial and research grade PA sensors/algorithms are unlikely to have been developed with the movement characteristics and energy demands of amputee populations in mind. Population specific algorithms are important to account for the numerous mobility or physical barriers individuals with amputation are likely to encounter when engaging in physical activity. An individualised approach would improve accuracy, either from a cut-point [50] or HR and PAEE relationship perspective. This approach is more time consuming and not necessarily feasible for large scale trials.

5.4.2. Clinical Implications

Previous research has focused on determining the effects of different prosthetic components and design on the economy of gait in individuals with amputation (Czerniecki & Morgenroth, 2015). Comparing gait, prosthetic fitting and components in a controlled laboratory environment is an important field of study in the development of ambulatory function in individuals with amputation. How these prosthetics function outside of the laboratory however is of great importance to researchers, manufacturers, clinicians and patients. Research in the area of assessing mobility in individuals with amputation has relied on subjective self-reported questionnaires (Miller et al, 2001; Stepien et al, 2007). However, subjective measurements of PA, although low cost, applicable to large populations and practical (Tudor-Locke & Myers, 2001), are commonly found to demonstrate low-moderate predictive validity (Prince et al, 2008) and lack sensitivity (Tudor-Locke & Myers, 2001) when compared to objective measures of PA. The objective models of estimating PAEE that have been developed in this chapter could provide insight into the amount of habitual ambulation-related PAEE individuals with amputations engage in, whilst wearing a range of prosthetic devices outside of the laboratory. The predictive models could allow clinicians and researchers to objectively assess the influence of different environments (e.g. weather and terrain or urban versus rural living conditions) and rehabilitation settings (e.g. in-patient versus home-based) on PAEE and provide insight into different vocational prospects in the community, thus optimising the potential for full integration back into society.

As wearable consumer PA monitors devices become more commonplace there are greater opportunities for people to engage in the self-management of their own care as well as providing lifestyle information to health care providers (Chiauzzi et al, 2015). As a note of comparison, the absolute percentage error of consumer multisensory devices (e.g. Fitbit, Microsoft Band and Apple Watch) (Chowdhury et al, 2017) ranged from 24 to 73% during walking tasks in healthy adults. In this study using individuals with amputation, the accuracy of the research grade AHR demonstrated a comparable level of accuracy (20-28%). The GT3X+HR models developed in individuals with LLA demonstrate a superior level of accuracy (12-14%). Despite the welcomed potential utility of these commercially available multi-sensor devices, until they have been validated in people with lower limb-loss it would be ill-advised to recommend there use for the accurate measurement and monitoring of PA in these groups. While the PAEE estimation models presented here show promising levels of accuracy, the wider scientific community needs to agree upon a threshold to signify an acceptable level of accuracy when using wearable devices.

Future research should consider applying and further developing new data analysis techniques such as artificial neural networks (Staudenmayer et al, 2009; Trost et al, 2012), hidden Markov models (Pober et al, 2006) and classification trees (Bonomi et al, 2009b) which use the rich information to classify certain activities and derive a more accurate estimate of EE. Future models should consider using additional activities (i.e. not just walking) that better resemble free-living conditions for individuals with amputation and evaluating the performance of EE prediction models during recovery after exercise (which contributes to TEE). Using a large diverse sample of participants, by encouraging research groups to work collaboratively and with different aetiologies would provide a more robust model for the assessment of PAEE in individuals with amputations. Future studies should also aim to cross-validate these newly developed population specific equations using a completely independent sample of participants, across a range of ADL or during habitual free-living conditions. The utility of the GT3X+ and the models developed in this chapter to estimate ambulatory PAEE during in-patient rehabilitation and out-patient recovery at home may provide clinically useful information to staff responsible for the rehabilitation care of UK military personnel with lower-limb amputation(s) at DMRC Headley Court and this question will be explored of the next chapter.

5.5. CONCLUSION

Of the three anatomical locations considered, wearing the accelerometer on the side of the shortest residual limb provides the most accurate prediction on PAEE in lower-limb amputees. The manually derived model integrating additional covariates or physiological variables (HR) with triaxial acceleration data, has been shown to possess greater validity for estimating ambulatory-related PAEE in individuals with traumatic lower-limb amputation(s), compared to the proprietary group calibrated algorithm intrinsic to the AHR device. Due to the poorer predictive validity of the AHR device, I would recommend using the GT3X+ together with the algorithms presented in this chapter, which demonstrate lower unexplained variance and lower estimation error.

CHAPTER 6. INFLUENCE OF LOWER-LIMB AMPUTATION SEVERITY AND RECOVERY ENVIRONMENTS ON INDICES OF PHYSICAL ACTIVITY AND FUNCTIONAL RECOVERY IN MILITARY PERSONNEL

The aim of chapter 5 was to validate a PA monitoring device that can be used to estimate ambulatory PAEE during free-living conditions in individuals with LLA. Originally, the plan for this chapter was to use the most accurate method of estimating PAEE determined in chapter 6 (i.e. the GT3X+HR method). I planned to use the HR data from the AHR device. For reasons explained in greater detail within this chapter, the wearing of the ECG electrodes had to be terminated; therefore, I lost the ability to include HR in the prediction models to estimate PAEE. The GT3X+ population-specific models developed in experiment one was used instead.

Chapter 6 will describe the utility of these validated devices to capture PA behaviour during waking hours in a longitudinal observational cohort study. The chapter will also comment on the clinical outcomes (functional and psychosocial) of participants collected at baseline.

6.1. INTRODUCTION

Individuals with LLA have lower PA levels compared to healthy adults (Bussmann et al, 2004; Halsne et al, 2013; Tudor-Locke et al, 2011) with the majority of individuals with amputations living sedentary lifestyles (Pepin et al, 2018). LLA can be associated with elevated anxiety, depression and perceptions of isolation (Deans et al, 2008), which may alter social integration and habitual activities. Unfavourable changes in body composition (Bouldin et al, 2016; Eckard et al, 2015; Kurdibaylo, 1996) characterised by excessive ectopic adiposity and the development of VAT obesity are associated with the development of secondary chronic health conditions. A high prevalence of cardiovascular disease and type 2 diabetes mellitus (Foote et al, 2015; Robbins et al, 2009; Stewart et al, 2015) are observed in individuals with LLA. Despite advanced technology and rehabilitative care, efforts have been unable to completely mitigate the effects of severe war trauma with the majority of US service members with amputation considered to have a high degree of disability (Hurley et al, 2015). Physical activity-based interventions and initiatives could improve QoL (Deans et al, 2008) and mitigate future healthcare costs (Geiling et al, 2012).

The promotion of PA is considered to be a vital therapeutic component in the rehabilitation, recovery and reintegration into society for individuals with LLA (Pepper & Willick, 2009). However, little is known about the consequences of amputation severity (e.g. unilateral versus bilateral LLA amputation) on habitual PA levels or ambulatory physical activity energy expenditure (PAEE). More recently, Sherman et al. (2019) reported significantly higher daily step count during periods of in-patient rehabilitation at DMRC Headley Court (2,258 ± 192 steps per day) compared to free-living out-patient periods at home (1,387 ± 363 steps per day), in individuals with bilateral amputation. The aforementioned study is the first to suggest differences in PA levels of bilateral LLA between these two environments. However, caution when interpreting these finding are warranted (as detailed in chapter 2, section 2.10) as the majority of instruments used to assess PA in individuals with LLA demonstrated are not specific enough for the population (Piazza et al, 2017). Therefore, activity monitors like the one used by Sherman et al. (2019) are unable to discriminate for the known differences in PAEE between persons with and without LLA, particularly multiple amputations. Consequently, it is highly likely that these findings underestimate the 'actual' PA levels of this population. It is also unclear how the activity levels in the individuals with bilateral amputation compare with people with unilateral amputation or a normative age and sex-matched control group.

The primary goal of amputee-based rehabilitation is to optimise physical function (the ability to mobilise and perform ADL independently) and reduce pain to facilitate full integration back into society. Chapter 4 reported mean length of rehabilitation of UK service personnel with amputation at DMRC to be 34 ± 14 months (Ladlow et al, 2015). Periods of in-patient rehabilitation at DMRC Headley Court consists of 4 weeks of intensive supervised IDT rehabilitation. Patients then spend an extended period of time back at home or work, depending on their individual circumstances and requirements. Whilst at home, patients are provided with an individualised exercise programme and encouraged to perform ADL on their prosthetic limb(s) (Ladlow et al, 2015). Accurate monitoring of PA behaviour has the potential to facilitate better care and rehabilitation management of individuals with amputations (Dudek et al, 2008; Redfield et al, 2013; Stepien et al, 2007).

As mentioned in the introduction and literature review to this thesis, PAEE is known to play a critical role in prolonging the cardiorespiratory, musculoskeletal, mental health and vocational opportunities following LLA. Unfortunately, available evidence indicates that individuals with traumatic LLA are pre-disposed to numerous unfavourable changes in health and well-being following their injury. It is currently unknown what impact the UK military's intermittent periods of complex trauma rehabilitation has on body composition, cardiometabolic health and function (this will be explored in chapter 7). However, it is important to first quantify daily PAEE during the different elements of the rehabilitation process, i.e. whilst an in-patient at DMRC and at home. Chapter 5 demonstrated the development of amputee-specific PAEE prediction models using the tri-axial Actigraph GT3X+ accelerometer with an independent physiological variable (heart rate) were superior to the proprietary group calibration algorithm intrinsic to the AHR. Worn at the hip on the shortest residual limb these GT3X+ models provide a valid estimation of ambulatory EE in individuals with unilateral and bilateral traumatic LLA (Ladlow et al, 2017). The aim of chapter 6 is to use these devices and associated validated population-specific models to determine: i) within-group differences in PAEE between in-patient (i.e. DMRC Headley Court) and out-patient (i.e. home) recovery cycles and; ii) between-group differences in PAEE, between unilateral LLA, bilateral LLA and a normative control group. Based on the nature of activities undertaken by UK military personnel with LLA during in-patient rehabilitation (see chapter 3) and the recent observations made by Sherman et al. (2019) I hypothesise that estimated ambulatory PAEE will be greater during periods of in-patient rehabilitation compared to periods of out-patient recovery. Currently no study has attempted to quantify PA at home using an objective and validated wearable device. Furthermore, based on the significant difference in functional outcomes presented in chapter 4 and the energy costs associated with ambulation described in chapter 5, it is hypothesised that ambulatory PAEE will be greater in individuals with a unilateral amputation compared to bilateral amputations.

6.2. METHODS

6.2.1. Study Design

A longitudinal observational cohort study design was used to compares the effect of two 4-week in-patient admissions followed by two 6-week blocks of active recovery at home on a range of function and health parameters in individuals with unilateral and bilateral LLA relative to normative controls. The trial protocol was approved by the UK Ministry of Defence Research Ethics Committee (Reference number: 512/MODREC/14). All participants provided written informed consent. Figure 6.1 provides a schematic description of the study design detailing the measurements taken over the 20 week study duration.

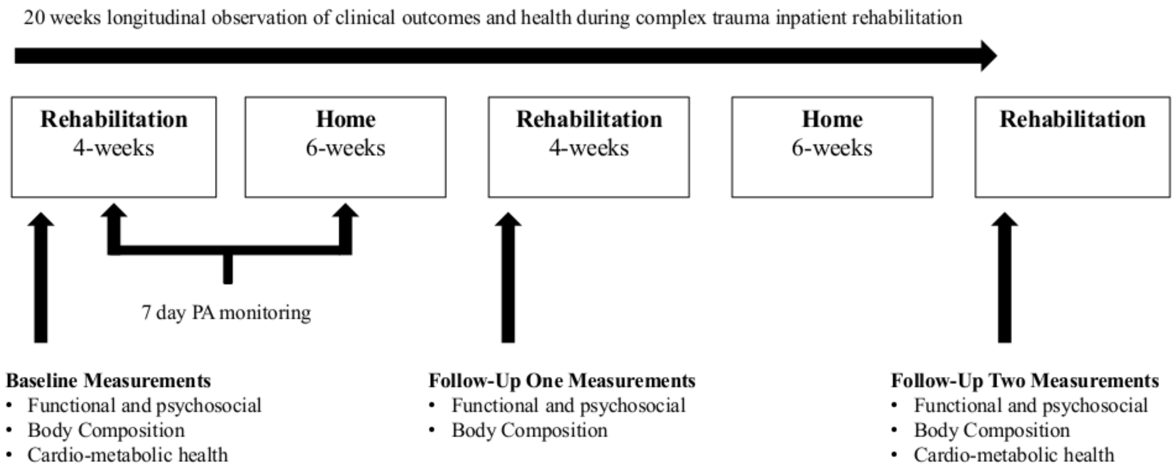


Figure 6.1: Schematic description of the longitudinal observational cohort study design

6.2.2. Participants

Participants who met the following eligibility criteria were included in the study: male, age 18-50 years, with unilateral or bilateral traumatic amputation of the lower limb(s), known to require a further three in-patient rehabilitation admissions at DMRC, Headley Court (confirmed by the patient's physician). Exclusion criteria included: known medical discharge / last admission date that did not allow for three in-patient admissions to DMRC, planned surgery during the data collection study period (potential risk of systemic inflammatory response and interrupted progression of physical function), severe traumatic brain injury, insufficient wound healing around the residuum (screened by physician), unable to ambulate in prosthetic(s) (screened by physiotherapist).

A group of age-matched, active, uninjured males, employed within physically active roles within the UK MOD (physiotherapists and ERIs), who engaged in aerobic or resistance-based training at least three times per week, were recruited to act as normative controls to the amputee cohorts. An age-matched, cohort of individuals with traumatic LLA not receiving rehabilitation at DMRC or a group of lower-limb injured UK military personnel (without LLA) receiving rehabilitation at DMRC may have provided a more suitable control group. However, all UK military personnel with an amputation must receive rehabilitation at DMRC Headley Court; therefore the first option would rely on civilian populations. As described in chapter 3 (section 3.6), civilian and military cohorts are not comparable as a population (mechanisms of injury, rehabilitation exposure and end goals of treatment). Recruiting an age-matched civilian group would also prove very challenging as they would not be allowed to attend DMRC for testing (for security and insurance reasons), thus requiring access to an alternative testing facility with similar testing equipment making this option impractical. Using a lower-limb injured group at DMRC would also bring unique challenges. The lower-limb department at DMRC typically provide one 3-week in-patient admission to their lower-limb injured patients, not the multiple admissions provided by complex trauma, making the two departments not comparable pathways of care. Severe lower-limb salvage groups treated within complex trauma department would be logical, however, this group often present with increased psychosocial complications (elevated anxiety and pain) as the primary barrier to functional progression (Ladlow et al, 2018; Ladlow et al, 2016). In lower-limb amputees physical-related barriers are more often observed (see chapter 2 section 2.4). The limb salvage group at DMRC are also known to require regular surgical revisions throughout their rehabilitation pathway (an exclusion criteria), and therefore not compatible with the study design. The uninjured normative control group recruited for the longitudinal observation cohort study are likely to closer resemble the PA status demonstrated pre-injury in both LLA groups, allowing us to comment on the impact of losing a limb in a previously physical active young male population. Due to their physically demanding jobs roles and regular engagement in aerobic/resistance-based training, the normative control group will not be representative of the wider general population. This will need to be an important consideration when interpreting the PA behaviour, ambulatory function, body composition and cardiometabolic health of the two amputee groups.

Injury characteristics of participants who completed the trial are presented in table 1. Reasons for continued in-patient admission in complex trauma at DMRC are numerous, but are likely to relate to their primary injuries (i.e. residual limb pain, skin/wound healing), secondary injuries (i.e. additional fractures or soft tissue injuries compromising progress) functional (i.e. lack of strength, lean muscle tissue or sensorimotor control) and/or psychosocial status (i.e. depression, anxiety, relationships with friends or family). For greater detail, please read chapter 3 where the rehabilitation journey of UK military personnel with traumatic LLA are discussed.

6.2.3. Clinical Outcomes

A variety of functional and psychosocial outcomes were captured at baseline, follow-up 1 (10 weeks) and follow-up 2 (20 weeks) as part of routine clinical practice (figure 6.1). For this chapter, only baseline clinical outcomes will be reported. This is to identify the function status of the participants (combine physical function outcomes alongside PA findings). As body composition (baseline, 10 weeks and 20 weeks) and cardiometabolic health (baseline and 20 weeks) were recorded at similar time points to clinical outcomes throughout the study duration, all longitudinal data will be reported together in the next chapter. The outcome measures taken were described in detail in Chapter 4 (section 4.2.2), but include: 6-MWT, amputee mobility predictor questionnaire (AMPQ), 9-item patient health questionnaire (PHQ-9), 7-item general anxiety disorder (GAD-7) and DMRC specific questions relating to mobility, ADL and pain status.

6.2.4. Estimating Daily PAEE

Validated as a method of accurately estimating ambulatory PAEE in individuals with LLA (see chapter 5), participants were asked to wear an Actigraph GT3X+ triaxial accelerometer on the hip of their shortest residual limb (right hip for controls) using an elasticated belt. Note: An AHR devise attached to the chest using two ECG electrodes so HR signals could be determined was originally implemented. Following 6 months of data collection, the wearing of the ECG electrodes and therefore the ability to measure HR was terminated for reasons out of my control (see section 6.4.4 for details). Individuals with amputation wore the device for 7 continuous days during in-patient rehabilitation at DMRC and 7 continuous days during active recovery back at home. Participants were provided with their PA monitor during the afternoon of their final day of inpatient rehabilitation. Therefore, the 7 days selected to record home-based activity was the first 7 days following inpatient rehabilitation. All injured personnel were provided with an individualised rehabilitation to complete whilst at home and encouraged to adhere fully to this program where possible. As the control participants were not engaging in rehabilitation they were asked to wear the device for 7 continuous days during a normal week of employment within the MOD (work). All participants were told to remove the monitor before participating in any water-based activity (hydrotherapy, showering) and during sleeping hours. After 7 days the participants returned the device and the data were downloaded onto the ActiLife version 6 software (Actigraph, Pensocola, FL, USA), for subsequent analyses. Wear time validation analysis, an integral part of the ActiLife software, was performed using the Troiano equation prior to converting to an excel file for analysis (Troiano, 2007). The ActiLife software defined non-wear time by an interval of at least 60 consecutive minutes whereby vector magnitude values remained constantly at zero. Based on a typical 8 hours sleep pattern and making allowances for water-based activities, > 14 hours (87.5% of potentially available data) from a16-hour waking day was considered an appropriate cut-point for a valid day. Vector magnitude data from the Actigraph was plotted against the corresponding 24-hr timestamp and visually inspected. Non-wear time vector magnitude data were excluded from the daily averages. Data obtained and derived from the Actilife software included mean physical activity counts (PAC) per day. The population specific prediction models, cross-validated in chapter 5, for estimating PAEE in the unilateral amputees, bilateral amputees and normative controls, respectively (Ladlow et al, 2017) are detailed below:

$$\text{Unilateral amputation PAEE} = (0.000979 \times \text{PAC}\cdot\text{min}^{-1}) + 0.225548$$

$$\text{Bilateral amputation PAEE} = (0.000929 \times \text{PAC}\cdot\text{min}^{-1}) - 0.051541$$

$$\text{Control PAEE} = (0.000776 \times \text{PAC}\cdot\text{min}^{-1}) + 0.427097$$

6.2.5. Statistical Analysis

To test for differences in clinical outcomes measures between the 3 groups at baseline a one-way analysis of variance (ANOVA) with Bonferroni *post-hoc* analysis was used. PAC and PAEE responses within and between amputation groups were determined by a two-way (group [UNI, BI] x environment [Rehabilitation, Home]) mixed-model analysis of variance (ANOVA). Where significant interactions were observed between the two amputation groups, multiple t-tests were applied to determine differences between groups and/or environments. As the control group wore the device in one environment only (work) a one-way ANOVA was used to determine the differences in PA between the three groups (i.e., both amputation groups during a week of rehabilitation versus control participants at work, and both amputation groups during a week at home versus controls at work). All data presented in text and figures are means and standard deviations (SD). Descriptive data reported in tables are presented as mean and prevalence reported as percent in parentheses. The mean change (Δ) of PA measures between environments is reported alongside 95% confidence intervals (CI) in parentheses. Statistical significance was set at *a priori* of $\alpha < 0.05$. All analyses were performed using IBM® SP SS® Statistics 21 for Windows (IBM, Armonk, NY, USA). Standardised effect sizes (Cohens *d*) were used to calculated changes between the two environments (rehabilitation and home) with thresholds of > 0.2 (small), > 0.5 (moderate) and > 0.8 (large) used (Cohen, 1988).

6.3. RESULTS

6.3.1. Injury Mechanisms / Characteristics

Table 6.1 describes the mechanisms and injury characteristics of participants with unilateral and bilateral amputation. There is a notable difference in the quantity and location of secondary trauma between the two groups.

Table 6.1: Injury Characteristics of Unilateral and Bilateral Amputees. Data are presented as number of participants (%)

	Unilateral Amputation	Bilateral Amputation
Mechanism of Injury		
IED	3 (37.5)	8 (100)
GSW	2 (25)	-
RTA	1 (12.5)	-
Training Exercise	2 (25)	-
Timing of Amputation		
Immediate	4 (50)	8 (100)
Delayed / Elective (> 30 days)	4 (50)	-
Level of Amputation		
Below Knee	6 (75)	4 (50)
Through Knee	1 (12.5)	5 (62.5)
Above Knee	1(12.5)	7 (87.5)
Secondary Injuries		
Fractures	3 (37.5)	8 (100)
Nerve damage	1 (12.5)	2 (25)
Soft tissue or vascular trauma	6 (75)	8 (100)
Location of Secondary Injuries		
Head/neck/face	1 (12.5)	5 (62.5)
Chest / upper back	1 (12.5)	5 (62.5)
Upper Limbs	1 (12.5)	8 (100)
Spine	-	3 (37.5)
Abdomen	1 (12.5)	4 (50)
Pelvis	2 (25)	6 (75)
Lower-Limbs	7 (87.5)	8 (100)

Abbreviations: IED = improvised explosive device, GSW = gunshot wound, RTA = road traffic accident.

Table 6.2 demonstrates the heterogeneous nature and severity of injuries of four participants enrolled into the longitudinal observational cohort study. Case studies include two individuals with unilateral transtibial amputation (one with ambulatory function comparable general population norms and the other comparable to active normative controls) and two individuals with bilateral transfemoral amputation (one with ambulatory function comparable to general population norms and the other below general population).

Table 6.2: Injury characteristics, length of rehabilitation and clinical outcomes of 2 individuals with unilateral transtibial amputation and 2 participants with bilateral transfemoral amputation

Injury	Mechanism of Injury	Additional Injuries and Rehabilitation Challenges	Length of Rehabilitation at Baseline	Clinical Outcomes at Baseline
Unilateral Transtibial Amputation	GSW	1. Multiple fractures to tibia/fibula and femur on amputated limb 2. Soft tissue damage to thigh musculature on amputated limb 3. Sciatic nerve transaction on opposing limb	20 Admissions 8 Months	**6MWD:** 475m **AMPQ:** 47/47 **Mobility:** can walk independently **ADL:** requires an aid or adaptation **GAD-7:** 7/21 - mild anxiety **PHQ-9:** 5/27 - mild depression
Unilateral Transtibial Amputation	RTA	1. Tibial plateau fracture 2. Extensive knee ligament damage	7 Admissions 4 Months	**6MWD:** 700m **AMPQ:** 44/47 **Mobility:** can walk independently **ADL:** requires an aid or adaptation **GAD-7:** 0/21 - no anxiety **PHQ-9:** 2/27 - no depression
Bilateral Transfemoral Amputation	IED	1. Open olecranon fracture 2. Open ulna fracture, 3. Multiple fractures to phalanx 4. Genital trauma 5. Perforated tympanic membrane 6. Extensive soft tissue injures to both thighs	20 Admissions 61 Months	**6MWD:** 360m **AMPQ:** 39/47 **Mobility:** can walk independently with an aid or adaptation **ADL:** requires an aid or adaptation **GAD-7:** 1/21 - no anxiety **PHQ-9:** 0/27 - no depression
Bilateral Transfemoral Amputation	IED	1. Extensive soft tissue damage to entire right arm 2. Perforated tympanic membrane 3. Two fractured lumbar vertebrae resulting in low back pain 4. Extensive soft tissue injures to both thighs	10 Admissions 21 Months	**6MWD:** 480m **AMPQ:** 46/47 **Mobility:** can walk independently with an aid or adaptation **ADL:** requires an aid or adaptation **GAD-7:** 1/21 - no anxiety **PHQ-9:** 2/27 - no depression

Abbreviations for table 6.2: GSW = gunshot wound, RTA = road traffic accident, IED = improvised explosive device, 6MWD = six-minute walk distance, AMPQ = amputee mobility questionnaire, ADL = activities of daily living, GAD-7 = general anxiety disorder – 7 item questionnaire, PHQ-9 = patient health questionnaire – 9 item questionnaire (depression)

6.3.2. Rehabilitation Length Prior to Baseline Measurements

At baseline, individuals with bilateral amputations had undergone a significantly greater length (39 ± 15 versus 14 ± 8 months, $p < 0.05$) and number of in-patient rehabilitation admissions (15 ± 15 versus 6 ± 3, $p < 0.001$) compared to individuals with a unilateral amputation, respectively.

6.3.3. Clinical Outcomes at Baseline

6.3.3.1. Physical Function

There was a significant main effect of group on 6-MWD at baseline ($p < 0.001$) (see table 6.3). *Post-hoc* analysis revealed significant differences between all 3 groups ($p < 0.001$) with the controls able to walk the longest and the bilateral the shortest distance. Further *post-hoc* analyses revealed that individuals with a unilateral amputation had greater mean AMPQ scores at baseline compared to bilateral amputees ($p = 0.003$) (see table 6.3).

6.3.3.2. Psychosocial Outcomes

No significant main effects of group were demonstrated on any psychosocial outcome measures. Mean depression and anxiety scores were low (< 5) and indicative of 'none / minimal' symptoms in both amputation groups. The majority (94%) of patients were able to control their pain symptoms at baseline (see table 6.3).

Table 6.3: Functional and psychosocial status of each group at baseline. Data presented as mean ± SD for continuous variables and n = with (%) for categorical variables

	Unilateral (n=8)	Bilateral (n=8)	Control (n=13)
Age (years)	30 ± 5	29 ± 3	28 ± 5
Function			
6 MWD (metres) *	574 ± 66	337 ± 85	705 ± 32
AMPQ #	46 ± 1	40 ± 4	N/A
DMRC - Mobility			
Able to run independently	1 (12.5%)	0 (0%)	13 (100%)
Able to walk independently	8 (100%)	5 (62.5%	13 (100%)
Requires a walking aid / adaptation	0 (0%)	3 (37.5%)	0 (0%)
DMRC - ADL			
Able to perform independently	1 (12.5%)	0 (0%)	13 (100%)
Requires aid/adaptation	7 (87.5%)	8 (100%)	0 (0%)
Psychosocial			
PHQ9 – Depression	3 ± 3	2 ± 2	N/A
< 5 no symptoms	6 (75%)	7 (87.5%)	N/A
> 10 moderate symptoms	0 (0%)	0 (0%)	N/A
GAD-7 Anxiety Disorder	3 ± 3	1 ± 1	N/A
< 5 no symptoms	6 (75%)	8 (100%)	N/A
> 10 moderate symptoms	0 (0%)	0 (0%)	N/A
Pain Status			
No pain	0 (0%)	1 (12.5%)	13 (100%)
Able to control their pain	7 (87.5%)	8 (100%)	13 (100%)

Abbreviations: 6-MWD = six-minute walk distance, AMPQ = amputee mobility predictor questionnaire, DMRC = Defence Medical Rehabilitation Centre, ADL = activities of daily living, PHQ-9 = 9-item patient health questionnaire, GAD-7= 7-item general anxiety disorder questionnaire.

* Significant differences between unilateral amputation, bilateral amputation and normative controls ($p < 0.001$).
# Significant difference between individuals with unilateral and bilateral amputation ($p < 0.05$)

6.3.4. Physical Activity Behaviour

There were no differences in the number of valid days (>14hours) recorded or mean wear time between groups or environments (i.e. rehabilitation versus home).

6.3.4.1. Activity Levels during In-patient Rehabilitation

There was a significant main effect of group on PAC.day ($F(2,26) = 5.378$, $p = 0.011$) and estimated daily PAEE ($F(2, 26) = 51.319$, $p < 0.001$). *Post-hoc* analysis revealed significantly reduced PAC.day ($p = 0.009$) and estimated daily PAEE ($p < 0.001$) in individuals with bilateral amputation when compared to normative controls (see figure 6.2 and table 6.4). Bilateral amputees also demonstrated significantly reduced estimated daily PAEE compared to individuals with unilateral amputation ($p < 0.001$) but no significant difference in mean PAC.day ($p = 0.142$).

Individuals with a unilateral amputation demonstrated mean PAC.day and estimated daily PAEE similar to normative controls ($p > 0.05$).

6.3.4.2. Activity Levels Whilst at Home

There was a significant main effect of group on PAC.day ($F(2,26) = 19.816$, $p < 0.001$) and estimated daily PAEE ($F(2, 26) = 88.507$, $p < 0.001$). *Post-hoc* analyses revealed significantly reduced PAC.day ($p < 0.001$) and estimated daily PAEE ($p < 0.001$) in individuals with bilateral amputations compared to normative controls (at work) (see figure 6.2 and table 6.4). Individuals with a unilateral amputation also demonstrated significantly reduced PAC.day ($p = 0.049$) and estimated daily PAEE ($p = 0.002$) compared to normative controls. Bilateral amputees recorded significantly reduced PAC.day (47%, $p = 0.008$) and estimated daily PAEE (70%, $p < 0.001$) compared to individuals with unilateral amputation.

6.3.4.3. Difference in Activity Levels between Environments

When comparing the two amputation groups, there was a significant main effect of environment on PAC.day^{-1} ($F(1,14) = 39.972$, $p < 0.001$) and predicted PAEE.day ($F(1,14) = 35.814$, $p < 0.001$). However, no significant environment x group interaction effects demonstrated between any PA measurements. However, a trend towards a significant interaction effect in PAC.d^{-1} was reported ($F(1,14) = 3.493$, $p = 0.083$). There was a reduction in PAC.day^{-1} and mean daily PAEE (kcal.d^{-1}) during habitual free-living at home in both amputee groups. However, the difference demonstrated between these two environments was greatest in the bilateral amputee group. Individuals with unilateral amputation reduced their PAC whilst at home by 17% ($d = 1.26$, $p = 0.018$) and their PAEE by 13% ($d = 1.21$, $p = 0.019$). Individuals with bilateral amputation reduced their PAC.d^{-1} by 42% ($d = 2.53$, $p = 0.001$) and their PAEE by 47% ($d = 2.51$, $p = 0.001$).

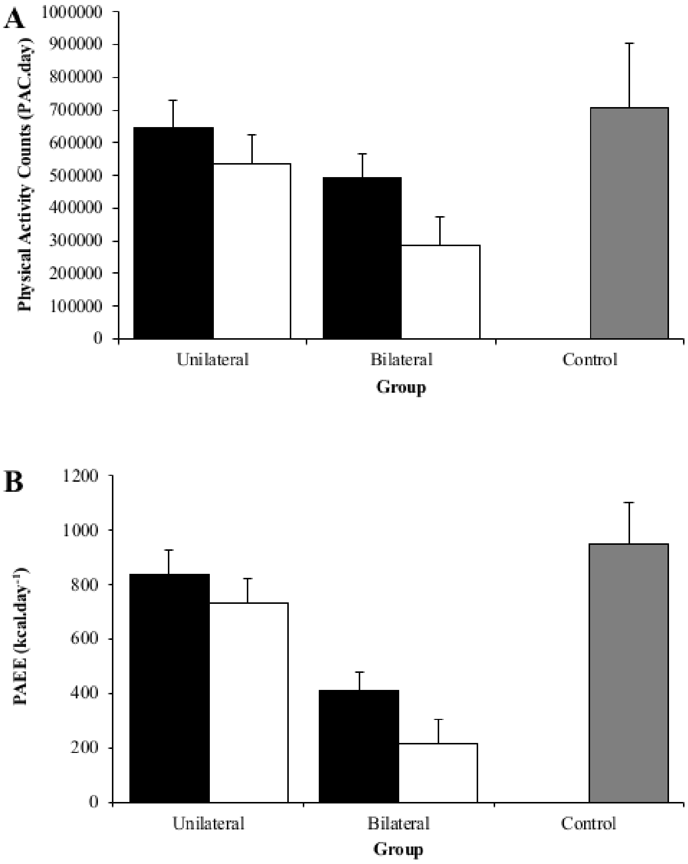


Figure 6.2: Physical activity behaviour. Panel A, shows the mean physical activity counts per day. Panel B, shows the estimated mean PAEE using predictive models validated by Ladlow et al. (2016). Black bars demonstrate activity levels of amputees during rehabilitation, white bars demonstrate activity levels of amputees whilst at home, grey bar demonstrates activity levels of controls whilst at work. Data are presented as mean plus SD.

Table 6.4: Descriptive data demonstrating mean daily physical activity levels of individuals with unilateral and bilateral amputation during in-patient rehabilitation and whilst at home and uninjured normative controls during work. Data presented as m ± SD and Δ m (95% CI)

	Unilateral (n = 8)			**Bilateral Amputee (n = 8)**			**Control (n = 13)**
	Rehabilitation	**Home**	**Δ Change**	**Rehabilitation**	**Home**	**Δ Change**	**Work**
Days (> 14h)	5 ± 1	5 ± 1	0 (-1 to 1)	6 ± 1	6 ± 1	0 (-1 to 1)	5 ± 1
Wear time (minutes)	918 ± 41	916 ± 55	-2 (-39 to 35)	918 ± 45	904 ± 42	-14 (-44 to 17)	934 ± 40
PAC.day # † ¶ @	645084 ± 86078	534248 ± 90125	-110836 (-196065 to -25607	492569 ± 72750	283357 ± 91406	-209212 (-299915 to -118509)	707632 ± 197909
PAEE (kcal.day^{-1}) * # † ¶ @	839 ± 88	733 ± 87	-106 (-188 to -23)	410 ± 68	217 ± 85	-194 (-279 to -108)	948 ± 155

* Significant difference between individuals with unilateral amputation and bilateral amputation during rehabilitation ($p < 0.05$)

\# Significant difference between individuals with unilateral amputation and bilateral amputation whilst at home ($p < 0.05$)

† Significant difference between individuals with bilateral amputation during rehabilitation and normative controls ($p < 0.05$)

¶ Significant difference between individuals with bilateral amputation at home and normative controls ($p < 0.05$)

@ Significant difference between individuals with unilateral amputation at home and normative controls ($p < 0.05$)

6.4. DISCUSSION

6.4.1. Rehabilitation and Baseline Clinical Outcomes

The number of in-patient admissions at baseline in the unilateral amputation group (6 ± 3) was fewer than the mean number demonstrated at last admission in chapter 4 (8 ± 4 admissions over 20 ± 11 months) (Ladlow et al, 2015). However, the mean length of rehabilitation at baseline in the bilateral amputations group (15 ± 15 admissions over 39 ± 15 months) was already greater than the mean length of rehabilitation reported at last admission (13 ± 4 admissions over 33 ± 10 months) in chapter 4. There are numerous reasons why the length of rehabilitation may differ between baseline measure recorded here and previous last admission measures. These reasons are primarily based on the rehabilitation needs of the patient. The complexity and severity of injuries provide different rehabilitation challenges that are unique to each individual patient (i.e. a patient-centred approach). The length of their rehabilitation is governed by the progress of functional and psychosocial health, and patients will only be considered for discharge based on clinical opinion of when they have reached a plateau in their clinical outcomes. It is important to note that no two traumatic injuries or wounds are created equally. While two people may be identified as having a unilateral transtibial amputation or bilateral transfemoral amputations their injuries and their rehabilitation requirements may differ substantially (see case study examples: Table 6.2). All four participants described in table 6.2 were nearing the end of their rehabilitation care pathway with varying amounts of rehabilitation admissions, supporting the premise raised in chapter 3 (combat casualty pathway) that injured UK military personnel with amputation will continue to receive rehabilitation care at DMRC until the end goal of treatment is attained. The majority of service personnel with amputation(s) have multiple disabling conditions that contribute to a very high level of disability and may make rehabilitation more challenging (see chapter 2 section 2.3, 2.4 and chapter 3 section 3.4). The case studies provide a nice summary of how extensive injuries can be from IED or blast-based mechanisms compared to blunt trauma. Table 6.2 clearly demonstrates how, based on the unique nature of their injuries, length of rehabilitation (months and number of admissions) does not necessarily correlate with level of function.

There was a significant difference in ambulatory ability (6MWD) between groups at baseline ($p < 0.001$) with normative controls able to walk a mean 131m and 368m further in 6 minutes than individuals with a unilateral and bilateral amputations, respectively (see table 6.3). It is important to note that this normative control group include very active adults and are not a reflection of a typical adult from the general population (see methods section describing the control group). Mean baseline functional values in individuals with a unilateral amputation (6MWD = 574 ± 66 metres, AMPQ score = 46 ± 1) were similar to the functional value previously reported at last admission at DMRC (6MWD = 544 ± 99 metres, AMPQ score 44 ± 3, $p > 0.05$) demonstrated in chapter 4

(Ladlow et al, 2015). The unilateral amputation group demonstrate a 6-MWD comparable to general population norms > 459 metres (Chetta et al, 2006) and a AMPQ score consistent with a MCFL K-Level of 4 (indicating a prosthetic ambulation ability that exceeds basic ambulation skills, an individual who can exhibit high impact, stress, or energy levels typical of the prosthetic demands of the child, active adult, or athlete) (Gailey et al, 2002).

Mean baseline functional values recorded in individuals with bilateral LLA (6-MWD = 337 ± 85 metres, AMPQ score = 40 ± 4) are less than the normative functional values previously reported at last admission at DMRC (6-MWD = 445 ± 104 metres, AMPQ score 42 ± 5) demonstrated in chapter 4 (Ladlow et al, 2015). Although the mean 6MWD is below age-matched general population norms (> 459m), the mean walking distance recorded at baseline is within the range recorded in US military personnel with bilateral amputation (6-MWD range: 264 to 645 metres) (Linberg et al, 2013). Their baseline 6-MWD and AMPQ scores of this bilateral LLA group are comparable to civilians with unilateral amputation due to peripheral vascular disease (6-MWD range: 298 to 463 metres, AMPQ score 37 ± 2) (Gailey et al, 2012). Considering this group of bilateral amputees are among the most severely injured adults reported in the available literature it can be argued that this group of participants are performing very well from a physical function perspective. Especially as the likely functional status of civilians with bilateral LLA treated within the NHS require a wheelchair to mobilise (see chapter 3 section 3.6). The functional status of the unilateral amputee group is indicative of full integration back into society. Based on the functional outcomes (6MWD and AMPQ) from former UK military personnel with bilateral amputation taken during their last admission to DMRC (chapter 4), there may be potential for further improvements in functional outcomes. This will be investigated during the longitudinal observational trial in the next chapter. Baseline psychosocial health status of both amputee groups (based on depression and anxiety scores) were indicative of 'none/minimal' symptoms and were comparable to the values previously reported in chapter 4 and my previous research investigating clinical outcomes in UK military trauma patients (Ladlow et al, 2015).

Explanations for how these two amputee groups have scored this favourably from a clinical outcomes perspective are provided in chapter 3 and chapter 4, but can be briefly summarised by the provision of advanced prosthesis available to UK military personnel, being a young and previously very active population and provided the time, resources (multidisciplinary group of health care professionals) and infrastructure to progress and facilitate their rehabilitation care.

6.4.2. Activity During In-Patient Rehabilitation and Recovery at Home

This is the first study to investigate the daily PA levels (PAC and PAEE) of individuals with traumatic unilateral and bilateral amputation, using PAEE prediction models developed and validated using a triaxial accelerometer (Actigraph GT3X+), which are bespoke to this population. Results indicate that the environment (i.e. rehabilitation versus home) and number of amputation(s) both effect PAC and PAEE in this population. Both amputation groups were significantly less active (PAC.day^{-1}) whilst at home compared to rehabilitation, albeit less so in the unilateral amputees (17%), compared to the bilateral amputees (42%). Individuals with a unilateral amputation expended significantly greater PAEE (kcal.day^{-1}) than individuals with bilateral amputations in both environments (429 kcal.d^{-1} (105%) greater during in-patient rehabilitation and 516 kcal.d^{-1} (238%) more whilst at home). No significant differences were observed in PAC or PAEE between individuals with a unilateral amputation during in-patient rehabilitation and normative active controls during work ($p > 0.05$). However, when unilateral amputees were monitored during habitual free-living conditions at home against controls at work there was a significant difference in PAC and PAEE demonstrated between these groups. It is important to remain mindful that the active normative control group who enrolled on this study are likely to present with levels of PAEE superior to the general population. This indicates that unilateral LLA, within a supportive environment, may have the 'capacity' to engage in levels of activity comparable to active normative controls, whereas individuals with bilateral amputations may not.

Previous research monitoring habitual activity in lower-limb amputees has relied on pedometers or accelerometers not validated in amputee populations. This makes both intra-group and inter-group comparisons very challenging. Using mixed aetiology (vascular and trauma), older adults (> 49 years of age) with greater than 10 years prosthetic use experience and without intensive supervised IDT rehabilitation provision (Halsne et al, 2013; Klute et al, 2006; Parker et al, 2010; Stepien et al, 2007), mean daily step counts ranged from 1,540 to 4,217 steps per day. A recent study performed at DMRC Headley Court investigated daily step counts of nine bilateral amputees (aged 26 ± 6 years) during rehabilitation and whilst at home (Sherman et al, 2019). At an average of 19 ± 7 months post-injury, these patients demonstrated similar ambulatory behaviour to those in the current study (i.e., significantly greater amounts of ambulation during rehabilitation compared to home). Mean daily step counts significantly reduced by 39% from 2,258 ± 192 during in-patient rehabilitation to 1,387 ± 363 when at home ($p < 0.01$) (Sherman et al, 2019). This study and the one presented here are the first to record and report objectively measured PA levels of individuals with bilateral amputations. The findings from Sherman et al. (2019) suggest mean daily step counts of individuals with bilateral amputations during habitual free-living are less than those of older unilateral amputees with vascular aetiology. Daily step counts below 5,000 have been classified as 'sedentary' (Tudor-Locke et al, 2011). PA behaviours reported in lower-limb amputees (regardless of age, aetiology, number of amputations) are below the recommended PA level for healthy living (7,000 to 8,000 steps per day) (Tudor-Locke et al, 2011). Although I did not measure step counts in the present study, this is the first study to demonstrate that unilateral amputees have the 'capacity' to adopt similar levels of PAC and PAEE as active normative age-matched controls. Their 6MWD are also comparable to the published normative values (Chetta et al, 2006), further supporting the theory that this group have the capacity to achieve fairly normative functional values. Thus potentially showcasing what can be achieved when care providers invest in rehabilitation and advanced prosthetic provision in young individuals with a unilateral amputation.

There remains a paucity of evidence on the implications of various amounts of PA on the health and well-being of adults with LLA. No recommended guidelines exist for the optimal level of PA (i.e. number of steps per day or quantity of PAEE) necessary to optimize health benefits for individuals with LLA.

To my knowledge, only one other study to have reported PAC in LLA populations is Theeven et al. (2012). This study used unilateral transfemoral and knee disarticulations participants (mean age 59 ± 13 years) with mixed aetiologies (23 trauma, 6 vascular and 1 cancer) and a functional status of MFCL (K) Level 2 (see table 2.1 for description of MFCL criteria). Using a uniaxial Actigraph GT1M accelerometer worn on the waist, they reported a mean 117,852 ± 57,982 PAC.day for the entire group and 156,304 ± 44,869 for those considered to be higher functioning based on self-selected walking speeds (Theeven et al, 2012). This data suggests that this bilateral LLA group engaged in greater amount of PA than these unilateral LLA. However, making direct comparison between this studies based on the PAC data is challenging. Theeven et al. (2012) recruited older civilian cohort with mixed aetiology and measured PAC.day using a uniaxial accelerometer. Comparing the recorded PA data to general populations is also challenging. In a NHANES 2003-2006 study, PACs and its relationship to cardiovascular disease was assessed in a general populations, using an older model of the Actigraph (7164 model) worn on the right hip (Luke et al, 2011). In a healthy group of 1,775 male adults (aged 20 to 65 years) the Actigraph accelerometer was worn a mean 14.6 hours.day for ~6 days. A mean 392.7 PAC.min was recorded, equivalent to 344,005 PAC.day (Luke et al, 2011). Using this as a reference value, the daily PAC scores indicate that both LLA group surpass the daily PAC demonstrated by the general population during rehabilitation at DMRC (unilateral LLA = 645,084 PAC.day; bilateral LLA = 492,569 PAC.day: see table 6.4). However, mean daily PAC for the bilateral LLA at home (283,357) did not meet this reference value from the general population. Although it used an age-matched population, one limitation for using this paper as a reference is the use of a uniaxial accelerometer. Uniaxial accelerometers are considered to be less accurate than more modern day triaxial accelerometers which are capable of detecting greater amounts of movement. In a study of 611 males (mean age 68 ± 8 years), Chomistek et al. (2017) used an Actigraph GT3X triaxial monitor for a mean 15.1 hours.day over 6.5 days. The authors measured mean daily PAC to be 602,923 ± 177,199 (Chomistek et al, 2017), considerably greater than the previously mentioned uniaxial-based PA study. This daily PAC value appears to fit between the PAC recorded by unilateral LLA during rehabilitation and home. There are currently no agreed cut-points to determine PAC or PAEE general population norms.

Previously, amputee gait has been reported as less energy efficient than people without amputation, which results in greater physical stress (expressed as HR or oxygen consumption) at a certain speed of walking (Bell et al, 2014; Czerniecki, 1996; Detrembleur et al, 2005; Genin et al, 2008; Ward & Meyers, 1995). Increased EE in amputees could be explained by postural asymmetry induced by compensatory strategies that reduce the production efficiency of muscular work in the amputated limb (Genin et al, 2008). However, it is important to note that UK military personnel with unilateral amputation with access to advanced prosthetic provision and intensive IDT rehabilitation appear to demonstrate comparable gait (temporal spatial parameters) and metabolic profiles to active male controls at self-selected walking speeds (Jarvis et al, 2017). Military personnel with unilateral amputation (transtibial and transfemoral) demonstrated comparable walking speeds, stride length, step length and cadence to uninjured controls. Individuals with unilateral transtibial amputation also demonstrated comparable walking energy costs to uninjured controls. Whereas individuals with bilateral transfemoral amputation walked significantly slower, with reduced cadence and 60% greater energy cost to controls (Jarvis et al, 2017). These findings may provide an explanation for the amount of PA demonstrated between groups in this study. The participants who enrolled in this study could have presented with similar gait profiles to the population described by Jarvis et al. (2017) (participants were exposed to similar durations of the same rehabilitation pathway, were treated by the same clinicians and had access to the same prosthesis provision). Therefore, if similarities in gait and metabolic profiles are known between UK servicemen with unilateral amputation and normative controls, then gait and metabolic profile might not be the primary reason for reductions in PAC and PAEE during habitual free-living at home in this unilateral LLA group. However, gait and metabolic profiles may be a contributing factor for reduced PAC and PAEE demonstrated in the bilateral LLA group. It was beyond the scope of this study to verify this hypothesis by performing kinematic and/or kinetic analyses of amputee gait.

Also pertinent to this military population is the assertion that the strongest predictor of current PA was being physically active before the amputation (Kars et al, 2009). Military personnel are often highly motivated, active young individuals prior to their injuries. Therefore, the PAC and PAEE demonstrated in this study might be higher than commonly attained by civilians with traumatic amputation or individuals with dysvascular-based aetiology. Indeed, physical function of military personnel with amputation has previously been shown to be higher than civilian populations and often comparable to general un-injured population (Doukas et al, 2013; Ladlow et al, 2016; Ladlow et al, 2015). Poorer scores in the 2 minute (Parker et al, 2010) and 6 minute walk tests (Lin et al, 2014) are found to correlate with lower levels of PA. The baseline ambulatory data for individuals with bilateral amputation supports this observation. The significant differences reported between the 3 groups in the 6MWT ($p > 0.001$) are commensurate with the significant difference demonstrated in estimated daily PAC and PAEE between the 2 amputation groups during habitual free-living at home and the normative control group at work ($p < 0.05$). It must be noted that these are cross-sectional observations and therefore it is not possible to determine whether the lack of PA leads to decreased physical ability or the lack of physical ability leads to lower PA levels. It is important to remain mindful that the control group were monitored during a week at work. Their PA behaviours may change significantly if monitored in their home environment.

Reduced levels of PA in lower-limb amputees may be associated with an increased fear of falling (Piazza et al, 2017). This may lead to strategies to overcome mobility restrictions and lead to adaptations in PA behaviour. For example, fewer and shorter walking periods (Klute et al, 2006) and walking slower may compensate for the decreased efficiency and could result in a comparable or even lower HR responses and energy costs than in individuals without amputation (Bussmann et al, 2004). There may also be times when its considered more appropriate for an individual with a LLA (particularly bilateral transfemoral amputation) to use a wheelchair to mobilise as oppose to ambulating in their prosthesis (Sherman et al, 2019). The absence of high mechanical loading through the lower-limb associated with prolonged sitting or wheelchair use may impose additional muscle atrophy and/or increased fat mass to the residual lower-limbs. Prolonged sitting or wheelchair use may also lead to hip and/or knee contractures leading to reduced range of motion at these joints (Esquenazi & DiGiacomo, 2001), which can disrupt prosthetic alignment and further contributes to the functional decline of the individual with amputation and the rehabilitation challenge already associated with their injury. Although not measured during this study, regular changes in residual limb tissue composition and skin damage associated with socket fitting (especially after high intensity exercise or prolonged prosthetic use) are regularly reported by staff and patients at DMRC Headley Court. This may be a leading contributor to decreased PA levels in both of the LLA groups.

Despite new materials and improved socket designs, there are numerous complications associated with prosthetic socket fitting that may negatively impact participation in structured exercise and/or habitual PA during both rehabilitation and home/work environments. Skin exposed to weight-bearing areas of the socket is not always resilient to the pressure and friction caused by the socket during ambulation (Van de Meent et al, 2013). Skin damage associated with prosthetic socket fitting is reported in approximately one third of individuals with transfemoral amputation (Hagberg & Branemark, 2001; Lyon et al, 2000; Meulenbelt et al, 2009). Chronic skin problems in the residual limb can often cause serious limitations in mobility and QoL (Demet et al, 2003; Pezzin et al, 2004). Other considerations related to optimal socket fitting that may negatively influence the function of the residual limb inside the socket include diminished limb sensation (Gottschalk, 1999; Neumann et al, 2005), pain (Behr et al, 2009; Smith et al, 1999) and unfavourable changes in body composition of the thigh (Eckard et al, 2015). Whole body composition changes over 20 weeks of intermittent rehabilitation and recovery at home will be investigated in the following chapter. However, localised body composition changes at the residual limb, such as excess fat mass, may affect the fit of an individual’s prosthesis and may additionally compromise mobility (Haboubi et al, 2001; Kurdibaylo, 1996). Thigh fat percent is significantly greater in the residual limb following transfemoral and transtibial amputations compared with the intact limb (Sherk et al, 2010). Localised lean muscle mass and strength at the thigh may also decrease (Isakov et al, 1996; Jaegers et al, 1995; Raya et al, 2010; Schmalz et al, 2001; Sherk et al, 2010; Tugcu et al, 2009); further complicating the ability to perform functional tasks and engage in daily PA wearing a prosthetic device. As localised thigh composition (adipose tissue, lean muscle tissue) and thigh muscle strength performance is not measured as part of this study it is beyond the scope of this thesis to verify this hypothesis.

6.4.3. Clinical Implications

The PAEE and PAC demonstrated during in-patient rehabilitation could be considered PA ‘capacity’ (i.e. what the participants are capable of) whilst the PAEE and PAC demonstrated at home is PA ‘performance’ (i.e. what they actually do) (Parker et al, 2010). The disparity in PAEE is demonstrated by both amputee groups but is particularly greater in individuals with bilateral amputations. Although it is perhaps unsurprising that PA levels of individuals with bilateral amputations are lower than in those with a unilateral amputation or normative controls, the gap that exists between these populations is significant, particularly as each participant had access to the most technologically advanced prostheses available and prolonged intensive rehabilitation care. The findings also suggest that improvements in technology, research, and clinical care are still needed to restore the PA and functional status of individuals with bilateral amputations to the levels observed in uninjured humans. Greater efforts towards the management and delivery of home-based PA interventions may narrow the gap in PAEE demonstrated between the two environments (rehabilitation versus home) and facilitate the progression of clinical outcomes recorded during in-patient admissions. It is currently unknown whether the amount of PA these amputation groups have demonstrated are enough to protect against cardiometabolic health diseases. Chapter 7 investigates this question in greater detail. However, based on the PAEE demonstrated by individuals with bilateral amputations, this group may be at increased risk of becoming overweight/obese and/or developing cardiometabolic syndrome or diseases.

Where large differences occur in activity levels between in-patient rehabilitation and home, it is a home-based intervention that is likely to contribute to improvements in long-term QoL and functional status of UK military personnel with LLA. By promoting longer-term behaviour change and allowing clinicians to monitor participant compliance the use of objective and validated PA assessment may better inform treatment interventions. An example of this could be when ‘capacity’ for PA (in-patient rehabilitation) is identified and used as a marker against ‘performance’ (free-living conditions). For example, a personalised home-based PA intervention may consider subtracting 10% off the ‘capacity’ value achieved during rehabilitation and use this as a target for daily PA during free-living.

6.4.4. Limitations

A limitation of this study is the relatively small sample size and variations within participants based on the heterogeneity in the severity of lower-limb loss injuries. However, this heterogeneity may be considered beneficial as the range of ambulatory abilities or PA behaviours captured improves the external validity of the study, making them more suitable for the wider UK military LLA population. Although the participants in this study are all serving military personnel, the injuries sustained in the unilateral LLA group are not dissimilar to those that might be expected in a RTA or adventure sports. It would therefore be argued that the results of habitual activity whilst at home are applicable to physically active civilian cohorts with LLA.

There are inherent weaknesses associated with using accelerometers to estimate PA levels during rehabilitation. Accelerometers are unable to register isometric activities, muscular work against an external force, such as lifting or resistance training (Butte et al, 2012). As resistance training forms an integral component of IDT rehabilitation at DMRC and is an activity strongly encouraged whilst at home, it is possible that PA levels were underestimated in this population. Although I used PA prediction models validated in this population, the models were determined during physiological assessments in a controlled laboratory on a treadmill. It is unknown how well this may correlate to ambulatory habitual activity in a variety of location, settings and environments in which the prosthetic user chooses to live, the use of PA diaries or subjective questionnaires may help overcome these pitfalls. The metrics I have used for this study (daily PAC and PAEE) describe the broader differences in PAEE between groups and environments. I have not used more commonly reported duration of time spent being sedentary, performing moderate to vigorous PA or TEE. Validating PA cut-points for this specific population would be required to obtain this information, which is beyond the scope of this study. However, this might be considered a useful follow up investigation to help develop a greater understanding of PA behaviour in this population.

All individuals with amputation receiving rehabilitation at DMRC Headley Court are encouraged to mobilise using their prosthesis during rehabilitation and whilst at home. During 7 days PA monitoring, the GT3X+ was worm on the hip of the shortest residual limb as this had previously found to demonstrate the greatest level of accuracy and least error during ambulatory tasks (Ladlow et al, 2017). Empirical evidence suggests civilians with bilateral amputation are likely to rely largely upon a wheelchair to mobilise in free-living conditions. Anecdotal evidence suggests a small number of military personnel within this bilateral LLA group would sometimes use a wheelchair to mobilise whilst at home (often due to factors associated with their living environment

- quicker and easier to perform some ADL, like going shopping). Time spent mobilising in a wheelchair as oppose to prosthetic use was not controlled for during this study. The algorithms and hip worn device is not sensitive enough to capture wheelchair PAEE, and this may contribute to lower values observe in the bilateral LLA group in this study. As prosthesis use is actively encouraged by rehabilitation therapists it was felt that positioning the GT3X+ on the hip using previously validated prediction models was appropriate to answer the primary research question. However, it is recognised that positioning the GT3X+ positioned at the side of the hip is likely to under-estimate PA levels during wheelchair use in the bilateral LLA group whilst at home. Another important consideration is the timing of 7 days PA monitoring. Exercise-based rehabilitation at DMRC is traditionally very intensive (20 hours per week for 4 weeks). Whilst selecting the first 7 days at home for PA monitoring was practical (I could physically hand over all monitoring equipment, clarify their understanding of my instructions and provide an opportunity for questions), it may have underestimated PA behaviour as participants were likely to have been very tired following their inpatient admission and this could have reduced ambulatory PA and immediate adherence to their personalised exercise programme. In hindsight the second week at home may have provided a closer resemblance to free-living ambulatory behaviour. This approach would have come with its own challenges though (for example, posting all equipment, ensuring they wear the monitor immediately at the right anatomical position, understand all instructions and adhere to the program).

It is not possible to comment on any progress made in PA levels during the 20 week rehabilitation period. The original study design involved monitoring PA during two rehabilitation admissions and two blocks of active recovery at home using the most accurate method of estimating PAEE developed in chapter 5. However, participant enthusiasm for PA monitoring declined and due to unforeseen problems with skin irritation associated with the ECG electrodes for the AHR monitor (senior clinicians requested that the electrodes no longer be worn), meant the use of HR signals in the predictive models to estimate of PAEE could no longer happen. It was later agreed that participants would no longer wear the AHR monitor and the Actigraph would only be worn during one 7 day period in each environment.

6.5. CONCLUSION

The ability to engage in levels of PA comparable to active normative controls is an important factor when discussing return to work status and/or the ability to fully integrate back into society. The ‘capacity’ of UK military personnel with traumatic unilateral LLA was shown to match the PAEE of active normative controls and ambulatory function of the general population. Thereby, maximising the probability of full integration back into society and eliciting the known health benefits associated with regular physical activity.

With considerable time investment in rehabilitation and access to advanced prosthetic provision, UK military personnel with bilateral amputation do not require a wheelchair to mobilise independently and demonstrate a level of ambulatory function similar to older civilian groups with unilateral amputation due to vascular based aetiology. Despite these favourable changes in physical function since their injury, the reduced levels of PAEE and PAC, particularly during free-living conditions at home, may predispose this group to increased risk of obesity and its associated secondary health conditions (cardiovascular disease and diabetes). This will be explored in greater detail in the following chapter.

CHAPTER 7. CHANGES IN FUNCTION, BODY COMPOSITION AND CARDIOMETABOLIC HEALTH, DURING REHABILITATION FROM TRAUMATIC LOWER-LIMB AMPUTATION

Among the key findings from chapter 6 was that individuals with a unilateral amputation demonstrated the capacity for comparable levels of PAEE to active normative controls whilst individuals with bilateral amputations performed significantly reduced levels of PA during in-patient rehabilitation and habitual free-living conditions as an out-patient. The consequences of this lower PA behaviour and associated reductions in EE on changes in physical function, body composition and biomarkers of cardiometabolic health, are currently unclear. Using the same participants and study design outlined in chapter 6, the longitudinal changes in cardiometaboilc health, body composition and function during rehabilitation from traumatic LLA were investigated.

7.1. INTRODUCTION

Individuals with LLA have previously demonstrated increased risk of weight gain / obesity (Bouldin et al, 2016; Gailey et al, 2008; Kurdibaylo, 1996; Robbins et al, 2009), developing cardiovascular disease and type 2 diabetes mellitus (Foote et al, 2015; Robbins et al, 2009; Stewart et al, 2015). The promotion of physical function and reduction of these unfavourable chronic conditions are primary objectives in the short-term rehabilitation and long-term recovery of individuals with traumatic LLA (Ladlow et al, 2016; Ladlow et al, 2015).

Much of the existing research relating to changes in body composition associated with LLA is based on vascular-related causes of amputation (i.e. non-traumatic), where patients have a broad age spectrum, but primarily comprise of older adults (Kurdibaylo, 1996; Renstrom et al, 1983b; Sherk et al, 2010). The majority of research relating to cardiometabolic disease risk associated with traumatic amputation is now over 20 years old (Hrubec & Ryder, 1980; Modan et al, 1998; Peles et al, 1995; Rose et al, 1987) and pre-dates many recent developments in prosthetic design and rehabilitation. Therefore, relying on these data alone may not translate to a young, previously active military population recovering from traumatic LLA with access to intensive rehabilitation and advance modern-day prosthetic provision. To date, one paper has investigated the impact of traumatic LLA on changes in body composition in young military personnel (mean age 23 ± 3 years) (Eckard et al, 2015). Monitoring for a 12 month period following amputation, Eckard et al.

(2015) reported significant increases in total fat mass and reductions in muscle mass during the first year following unilateral and bilateral amputation in US military personnel receiving rehabilitation. What is unclear is whether the increase in total body fat content (within unilateral and bilateral amputation groups, both were categorised as over-weight after 12 months) continued to increase at same rate over time. Furthermore, there is no indication as to whether there was an increased prevalence of obesity and/or what impact these changes in body composition had on clinical outcomes, including associated risks and incidence of cardiometabolic disease.

A retrospective cohort study of severely injured US military personnel wounded in Iraq and Afghanistan found the severity of combat injury was associated with the subsequent development of hypertension, coronary artery disease, diabetes mellitus, and chronic kidney disease (Stewart et al, 2015). Chapter 4 showed the severity of injury in individuals with bilateral amputation (NISS = 44 ± 12) to be significantly greater than individuals with unilateral amputation (NISS = 28 ± 11, $p < 0.05$). These data indicate that UK military personnel with LLA may be at increased risk of developing cardiometabolic disorders and diseases and that risk appears to be significantly elevated in individuals with bilateral amputations. It is estimated that the total 40-year cost of the UK Afghanistan lower-limb amputee population alone is £288 million; this figure estimates cost of trauma care, rehabilitation, and prosthetic costs. However, these costs do not cover any lifelong care costs for the management and treatment of chronic comorbidities, which may be required given the higher risk and prevalence of disease (Edwards et al, 2015). The functional, body composition and cardiometabolic consequence associated with traumatic LLA in military personnel remains unclear.

The aim of this longitudinal cohort study was to prospectively quantify changes in clinical outcomes, body composition and biomarkers of cardiometabolic health in military personnel during rehabilitation from traumatic LLA. Cohorts of unilateral and bilateral amputees were observed over a 20 week period of rehabilitation, including two 4-week in-patient admissions at DMRC Headley Court and two blocks of 6-weeks active recovery at home. Based on baseline clinical outcome values and each group's requirement to continue with in-patient rehabilitation it was hypothesised that clinical outcomes would improve in both LLA groups at 20-weeks follow-up. Based on PA levels demonstrated during rehabilitation and whist at home (chapter 6), it was hypothesised that individuals with unilateral traumatic LLA would demonstrate more favourable body composition and cardiometabolic component risks than individuals with bilateral LLA but comparable values to normative controls.

7.2. METHODS

The study protocol for chapter 7 has been described in chapter 6 figure 6.1. Baseline clinical outcomes have already been described in chapter 6. In this methodology section, the techniques used to measure body composition and cardiometabolic component risks factors are detailed.

7.2.1. Body Composition

Body composition was determined using a DEXA (Hologic Discovery, Bedford, UK) during each rehabilitation admission. Imaging technologies including DEXA have been identified as accurate methods for determining body composition. DEXA can accurately detect whole-body fat mass (within 2% coefficient of variation) and has the capacity for regional analysis (Heymsfield et al, 1997). A daily quality control scan using a phantom spine was conducted to calibrate the device prior to all scans as per the manufacturer's instructions. Descriptive participant characteristics were then entered into the QDR^{TM} system for Windows software. Participants were scanned wearing light clothing (i.e., shorts) and positioned centrally on the scanning bed with legs spread evenly apart (either side of the mid-line of the body) with hands placed in a mid-prone position to allow a gap between the arms and trunk. This body position allowed regions of interest to be defined by the operator using the software provided by the manufacturer. Participants were instructed to remain as still as possible whilst the principle investigator (trained in ionising radiation (medical exposure) regulations) performed and analysed each scan. Body composition assessments were collected at 3 time points; start of their first, second and third admission (see chapter 6, figure 6.1). DEXA scans were performed within two days of arriving at DMRC for each respective in-patient admission and time matched for the control participants. All scans were recorded between the hours of 07:30 and 08:30 am. As DEXA scans were performed immediately prior to fasted blood sampling at baseline and at end of the study period, to replicate these conditions for the middle scan, participants were asked to attend in a fasted state ≥10 hours. Scans were analysed for total mass, fat mass, lean mass, percentage body fat, android / gynoid fat percentage and VAT area following the guidelines described in the QDR for Windows manual (Hologic, Bedford, UK).

As discussed in chapter 2 (section 2.6) understanding body fat distribution and its clinical implications to health is critical to timely treatment interventions. The android region represents the mid-section of the body (waist). A high distribution of fat in this region leads to an 'apple shaped' body or high central obesity. Central obesity is measured as an increased waist circumference or waist-hip ratio. However, an increase in abdominal circumference may be due to greater amounts of subcutaneous or visceral fat and it is the increase of VAT, which has specifically been linked to several pathological conditions including impaired glucose metabolism, insulin resistance and cardiovascular disease (Tchernof & Despres, 2013). Visceral obesity itself is an independent component of metabolic syndrome and the magnitude of obesity directly relates to the prognosis of this condition (Mathieu, 2008; Ritchie & Connell, 2007) (chapter 2, section 2.7). The quantitative assessment of visceral fat can be estimated with DEXA. Manufacturer developed algorithms estimate visceral fat from the android/abdominal region by quantifying the thickness of subcutaneous fat on each side of the abdominal wall. This measurement is used to extrapolate the amount of subcutaneous fat in the abdominal/android region using appropriate modelling. The gynoid region represents the hip portion of the body. A high distribution of fat in this region leads to a 'pear shaped' body, which tends to demonstrate an accumulation of more subcutaneous fat, and is deemed more cardioprotective in women (Chen et al, 2019). It is recommended to maintain an android:gynoid ratio of under 1 (meaning it is typically a better measure of health if the android region remains a lower relative percentage than the gynoid region (commonly estimated via the anthropometric assessment of waist-to-hip ratio). Typically, changes in lean body mass are primarily driven by increases or decreases in skeletal muscle mass. Consequently, as each amputation group are engaging in an exercise-based rehabilitation program, monitoring changes in lean body mass will be important monitoring tool to assess the successfulness of rehabilitation.

Height was measured and recorded to the nearest centimetre using a stadiometer (Seca 222, Birmingham, UK). Due to the absence of both lower limbs and varying residual limb length, pre-injury body height was used for individuals with bilateral amputation. Waist and hip circumference were taken at baseline only. Waist measurements were taken at the midway point between the lowest rib and the top of the iliac crest, with hip circumference taken as the widest part of the buttocks. The mean of three measurements were used.

7.2.2. Cardiometabolic Health

7.2.2.1. Blood Sampling and the Oral Glucose Tolerance Test (OGTT)

Blood sampling occurred on two occasions during the study, within 3 days of commencing in-patient admission one (baseline) and admission three (20-weeks follow-up - see figure 6.1). All participants reported to the laboratory in the morning (immediately following their DEXA scan) after an overnight fast (≥10 hours). A cannula (BD Venflon Pro, BD, Helsingborg, Sweden) was inserted into the antecubital vein and a 25ml fasted blood sample drawn. All blood samples were collected using syringes (BD, Helsingborg, Sweden). Within 5 minutes of the baseline blood sample being drawn, participants consumed 140g of a carbohydrate supplement (Polycal, Nutricia Advanced Medical Nutrition, Trowbridge, UK) equivalent to 75g of glucose combined with 87 ml of water. Following consumption of this supplement, a further 100 mL of water was added to the same glass to ensure all of the glucose solution had been consumed and secondly, to allow the participant to clear any remaining glucose solution from the mouth. Blood samples were then obtained every 15 minutes for the next 2 hours. Whole blood from each sample was immediately dispensed into two collection tubes (Sarstedt Ltd., Leicester, UK), one containing ethylenediaminetetraacetic acid as the anticoagulant for plasma samples, the other with 'serum separators' for serum samples. The ethylenediaminetetraacetic acidtube was immediately centrifuged at 3500rpm for 10 minutes (IEC Centra CL2 Centrifuge, Basingstoke, UK), the serum separation tube was left to stand at room temperature for 15 minutes before centrifugation. Plasma and serum samples were subsequently dispensed into 0.5 ml aliquots using a pipette and immediately stored at -80ºC (Panasonic MDF-U55-V Ultra-Low Temperature Freezer, Japan). The intravenous cannula was kept patent through periodic flushing with 0.9% Sodium Chloride (NaCl) solution (Braun, UK), with the first 5 ml of each blood draw discarded to avoid contamination.

7.2.2.2. Blood Analysis

At the completion of data collection all blood samples stored at DMRC Headley Court were transported on dry ice to the biochemistry laboratory at the University of Bath for analysis. All blood analyses were performed in duplicate using a batch analyse. Concentrations of total cholesterol, HDL cholesterol, triacylglycerol, NEFA, plasma glucose and C-Reactive Protein (CRP), were conducted on a Daytona (Randox Laboratories, Crumlin, NI) analyser according to manufacturer instructions using commercially available immunoassays (Randox Laboratories, Crumlin, NI). Commercially available enzyme-linked immunosorbent assays were used to measure serum Insulin (Quantikine HS, R&D systems Inc., Abingdon, UK) in accordance with the manufacturer's instructions. A justification for each cardiometabolic health markers is provided in the literature review of this thesis. Absorption was determined using a microplate reader (SPECTROstar Nano, BMG LabTech, Ortenberg, Germany) at the wavelengths specified by the kit manufacturer. LDL cholesterol was estimated using the Friedewald equation (Friedewald et al, 1972):

$$\textit{LDL Cholesterol} = \textit{Total Cholesterol} - \textit{HDL Cholesterol} - (\textit{Triglycerides} / 2.2)$$

Analysis of the OGTT results included calculation of the incremental area under the curve (iAUC) for insulin and glucose concentrations according to the trapezoid rule (Wolever, 2004). Homeostasis model assessment for insulin resistance (HOMA-IR) was calculated (Turner et al, 1979) as:

$$\textit{HOMA-IR} = \textit{Fasting glucose } (mmol{\cdot}l^{-1}) \times \textit{fasting insulin } (mU{\cdot}l^{-1}) / 22.5$$

Homeostasis model assessment for pancreatic β-cell function (HOMA-β) was calculated (Matthews et al, 1985) as:

$$HOMA\text{-}\beta = (\text{Fasting insulin } (mU \cdot l^{-1}) \times 20) / (\text{Fasting glucose } (mmol \cdot l^{-1}) - 3.5)$$

The Insulin Sensitivity Index (ISI Matsuda) was calculated (Matsuda & DeFronzo, 1999) as:

$$ISI_{Matsuda} = 10,000 \sqrt{} [\text{Fasting glucose } (mg \cdot dl^{-1}) \times \text{fasting insulin } (\mu U \cdot ml^{-1})] \times [\text{mean OGTT glucose value } (mg \cdot dl^{-1}) \times \text{mean OGTT insulin value } (\mu U \cdot ml^{-1})]$$

HOMA2-IR, HOMA2-β and HOMA2-S were determined using the calculator downloaded and provided online at https://www.dtu.ox.ac.uk/homacalculator. A justification for using these indices of insulin resistance and sensitivity is provided in the literature review of this thesis.

7.2.3. Statistical Analysis

To test for differences between the 3 groups (unilateral, bilateral, control) at baseline for body composition and indices of cardiometabolic health a one-way ANOVA with Bonferroni *post-hoc* analysis was used. Within and between group responses for most clinical outcomes were analysed using a 2 way (group [unilateral and bilateral] x time [baseline, follow up 1, follow-up 2]) mixed model ANOVA was performed. For 6-MWD and all body composition measures a 2 way (group [unilateral, bilateral, control] x time [baseline, follow up 1, follow-up 2]) mixed model ANOVA was performed. In order to simplify data analysis and facilitate the interpretation of complex data set (Hopkins et al, 2009 and Matthews et al 1990), serial measurements of glucose and insulin responses to the OGTT at baseline and follow-up were converted into simple summary statistics (i.e., fasting and peak concentrations, time to peak, iAUC and estimates of insulin resistance and sensitivity (wolever and Jenkins 1986). Responses within and between groups for these components of cardiometabolic health were analysed using a 2-way mixed model ANOVA (group [unilateral, bilateral, control] x day [baseline, follow-up 2]). The precise time-course of response to glucose loading within and between groups were analysed using a three-way (group [unilateral, bilateral, control] x day [baseline, follow-up 2] x OGTT time point [0, 15, 30, 45, 60, 90, 120] mixed model ANOVA. ANOVA were performed irrespective of any minor deviations from a normal distribution (Maxwell, 2004), but with Greenhouse-Geisser corrections applied to intra-individual contrasts where $\varepsilon < 0.75$ and the Huynh-Feldt corrections applied for less severe asphericity (Atkinson, 2002). Where significant interactions were observed, one-way ANOVAs (for 3 group or time-point comparisons) with Bonferroni *post-hoc* analysis or multiple t-tests were applied to determine the location of variance both between time points within each group relative to baseline. All data are presented in text as means and SD, whereas figures display means with error bars representing standard error of the mean. Descriptive data reported in tables are presented as mean ± SD for continuous variables and prevalence reported as a number and percent in parentheses for categorical variables. Mean and the lower and upper 95% CI of the change (Δ) were calculated using all data points. The prevalence of component risk factors to predict metabolic syndrome in all groups (as detailed in chapter 2, section 2.7.3) is also presented. Statistical significance was set at *a priori* of α < 0.05. All analyses were performed using IBM® SPSS® Statistics 21 for Windows (IBM, Armonk, NY, USA). Standardised effect sizes (Cohens *d*) were also calculated facilitate the interpretation of the substantive significance from baseline to follow-up within these groups, with thresholds of > 0.2 (small), > 0.5 (moderate) and > 0.8 (large) used (Cohen, 1988).

7.3. RESULTS

The injury characteristics of the participants are described in chapter 6 (table 6.1). The results section will first show how functional and clinical measures change with time during the 20 week rehabilitation period, before presenting within and between group body composition and markers of cardiometabolic health responses during the same 20 week study period.

7.3.1 Changes in Clinical Outcomes

7.3.1.1. Physical Function

The level of ambulatory function remained relatively unchanged in the normative control group (705 ± 32 m, 706 ± 25 m and 710 ± 21 m, at baseline, 10 weeks and 20 weeks follow-up respectively) (figure 6.1). The unilateral amputee group demonstrated a non-significant small effect size increase in distance walked between baseline and at 20 weeks follow-up (24 m, *d* = 0.36, p > 0.05). The bilateral amputee group demonstrated a non-statistically significant medium effect size improvement (54 m, *d* = 0.59, p > 0.05), between baseline and follow-up 2 (figure 7.1). The prevalence of individuals with bilateral amputation able to walk independently (no longer requiring an aid or adaptation) increased from 63% (n = 5) to 88% (n = 7) (see figure 7.2A). The prevalence of individuals with unilateral amputation able to run increased from 13% (n = 1) to 63% (n = 5) (figure 7.2C) and able to perform ADL independently increased from 13% (n = 1) to 75% (n = 6) (figure 7.2B). Neither amputee group demonstrated significant AMPQ score changes over time (figure 7.2D).

7.3.1.2. Psychosocial Outcomes

As reported in chapter 6 table 6.3, there were no significant differences at baseline in mean depression or anxiety scores between unilateral and bilateral amputee groups. No time or group x time interaction effect were reported in anxiety of depression scores (p > 0.05). Mean depression and anxiety scores were low and are indicative of 'none / minimal' symptoms in both amputation groups (as reported in chapter 6) and scores remained low throughout the duration of the study. Mean depression (PHQ-9) scores were unchanged (3 ± 3 to 2 ± 2 and 2 ± 2 to 2 ± 1 in the unilateral and bilateral groups respectively) as were mean anxiety (GAD-7) scores (3 ± 3 to 3 ± 3 and 1 ± 1 and 0±1 in the unilateral and bilateral groups, respectively). The only changes in psychosocial health of note were depression scores < 5 increased from 75% to 100% in the unilateral amputee group between baseline and midpoint follow-up (10 weeks) and remained for the duration of the study, and that the prevalence of bilateral amputees reporting no pain increased from 13% to 38% at midpoint follow-up 1 and remained stable at 20 weeks follow-up. The majority (94%) of patients were able to control their pain symptoms throughout the study duration.

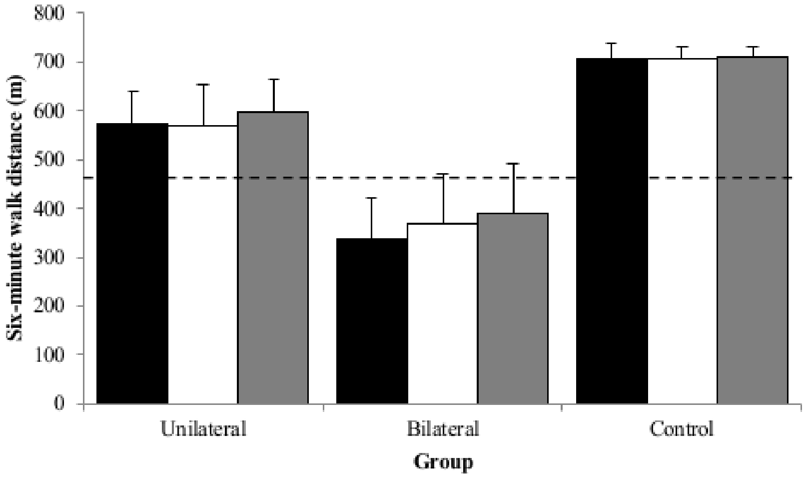


Figure 7.1: Six-minute walk distance (m) of each group over the three time points. Black bars represent baseline, white bars represent 10 weeks follow-up, grey bars represent 20 weeks follow-up. The dotted line represents general population norm (459m) from Chetta et al. (2006). Data are presented as mean and SD.

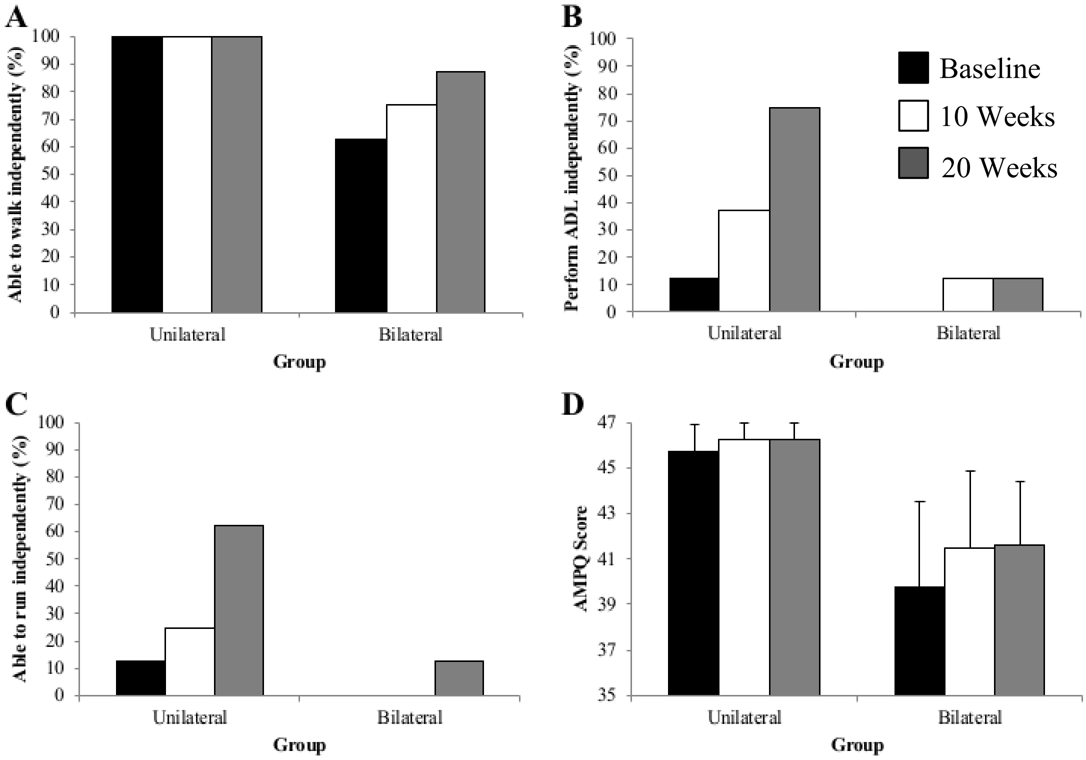


Figure 7.2: Functional Outcomes over time in the unilateral (n = 8) and bilateral (n = 8) amputee groups. A: Prevalence of participants able to walk independently, B: Prevalence of participants able to perform activities of daily living (ADL) independently, C Prevalence of patients able to run independently, D: Amputee Mobility Predictor Questionnaire (AMPQ) scores. Black bars represent baseline, white bars represent 10 weeks follow-up and grey bars represent 20 weeks follow-up. Data are presented as prevalence (%) for figures A, B and C. Data in figure D are presented as mean and SD.

7.3.2. Body Composition

There was a significant main effect of group on waist circumference, waist to hip ratio, lean mass, fat mass, body fat percent, android fat percent, gynoid fat percent and VAT area at baseline (see table 6.1). A *post-hoc* analysis found individuals with bilateral amputation demonstrated significant greater fat mass ($p = 0.005$), percent body fat ($p < 0.001$), android fat percent ($p < 0.001$), gynoid fat percent ($p < 0.001$), VAT area ($p < 0.001$), waist circumference ($p = 0.010$) and waist:hip ratio ($p = 0.001$) and reduced lean mass ($p = 0.026$) compared to normative controls at baseline. Individuals with bilateral amputation also demonstrated significantly larger waist circumference ($p = 0.014$) and VAT area ($p = 0.046$) compared to unilateral amputees. There were no significant differences reported between individuals with unilateral amputation and controls on any measure of body composition at baseline. Despite varying levels (transtibial and transfemoral) and numbers of amputation(s) there were no significant differences in total body mass between groups, see table 7.1 and figure 7.4A. However, table 7.1 and figure 7.4 clearly demonstrate how different human tissues (fat and muscle) are distributed across the body between groups. Figure 7.3 compares the DEXA images of 2 individuals with amputation (one unilateral and one bilateral) the increased distribution of adipose tissue around the abdomen and reductions in lean muscle tissue around the hip can be nicely observed.

Table 7.1: Anthropometric and blood pressure status of unilateral and bilateral LLA and normative control groups at baseline. Data presented as mean ± SD

Anthropometric / Physiological	Unilateral	Bilateral	Control	One-Way ANOVA
Height (cm)	180 ± 9	181 ± 9	180 ± 5	F = 0.226, p = 0.779
Body Mass (kg)	77.9 ± 10.9	83.5 ± 21.3	80.2 ± 8.3	F= 0.357, p = 0.703
Waist Circumference (cm) *†	84 ± 6	101 ± 22	83 ± 5	F = 5.661, p= 0.009
Waist:hip ratio *	0.87 ± 0.04	0.93 ± 0.10	0.83 ± 0.03	F = 8.091, p = 0.002
Lean mass (kg) *	58.8 ± 5.4	56.8 ± 8.5	65.9 ± 6.9	F = 4.607, p = 0.019
Fat mass (kg) *	15.7 ± 5.6	24.0 ± 13.7	11.3 ± 3.3	F = 6.086, p = 0.007
Body fat % #	19.9 ± 5.5	26.9 ± 8.7	14.1 ± 3.4	F = 11.931, p < 0.001
Android fat % #	21.7 ± 7.8	30.2 ± 11.4	14.6 ± 4.1	F = 9.894, p = 0.001
Gynoid fat % #	24.0 ± 6.2	28.9 ± 7.4	17.2 ± 4.4	F = 10.586, p <0.001
Android:gynoid ratio	0.88 ± 0.2	1.02 ± 0.2	0.85 ± 0.1	F = 2.909, p= 0.072
VAT area (cm^2) # †	56.0 ± 20.8	116.7± 47.1	45.6 ± 6.9	F = 12.877, p < 0.001
Systolic BP (mmHg)	125 ± 6	137 ± 8	118 ± 2	F = 29.791, p < 0.001
Diastolic BP (mmHg) *	69 ± 10	78 ± 9	63 ± 7	F = 7.875, p = 0.002

* A significant difference between individuals with bilateral amputation and normative controls ($p < 0.05$).
# A significant difference between individuals with bilateral amputation and normative controls ($p < 0.001$)
† A significant difference between individuals with bilateral amputation and unilateral amputation ($p < 0.05$)

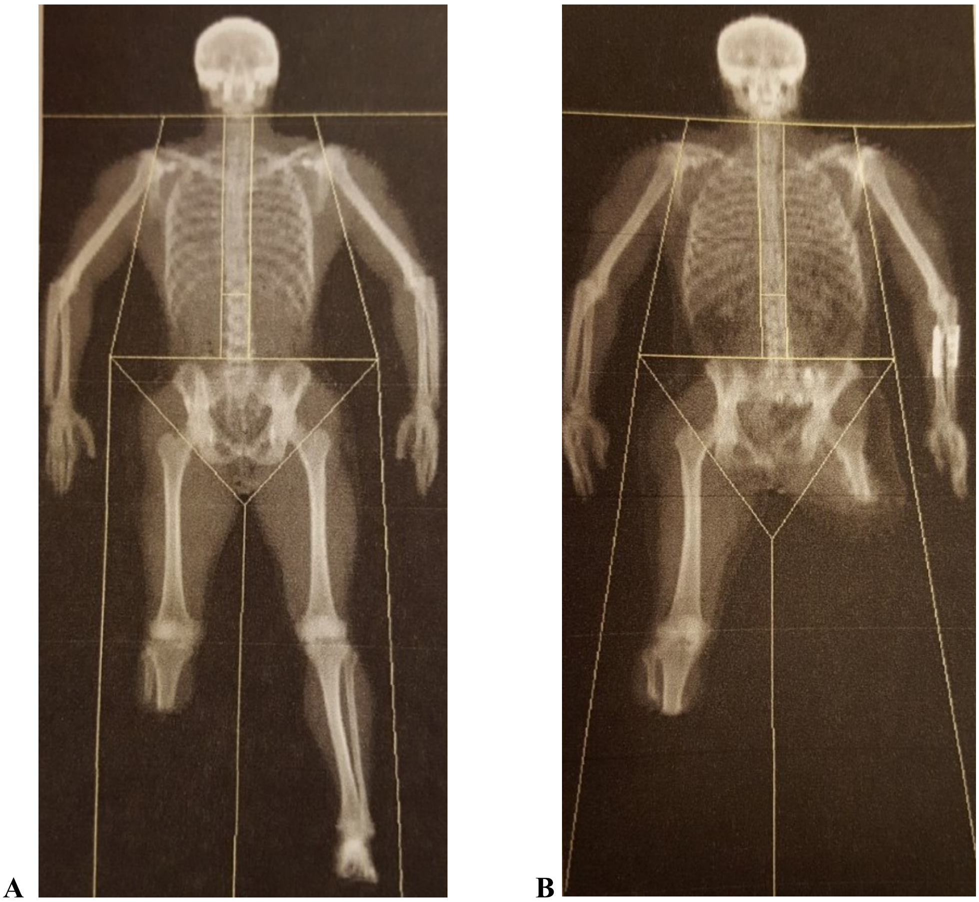


Figure 7.3: DEXA scan image of a UK military serviceman with (A) unilateral transtibial amputation and (B) Bilateral transtibial/transfemoral amputation. The density of different tissue can be identified; human skeleton is coloured white. Lean tissue mass is light grey and adipose tissue (more noticeable around the abdomen of figure B) is a dark grey. Additional metal work (internal fixator) can be seen on the left forearm in figure B and a reduction in muscle tissue around the hip of the shortest residual limb.

There was a significant effect of time (F = 3.413, p = 0.049) and a time x group interaction effect (F = 2.949, p = 0.029) for Android fat percentage and a significant time x group interaction effect in android:gynoid ratio (F = 3.368, p = 0.016). There was a significant difference in Android fat percent change over time between individuals with unilateral amputation and controls (p = 0.015), where android fat percentage reduced over time in the unilateral amputee group. Despite varying levels and numbers of amputation(s) there were no significant differences in total body mass between groups Fig 6.4A. However, figure 6.3 and figure 6.4 (E to H) clearly demonstrates differences in the regional distribution of lean and fat tissues between groups.

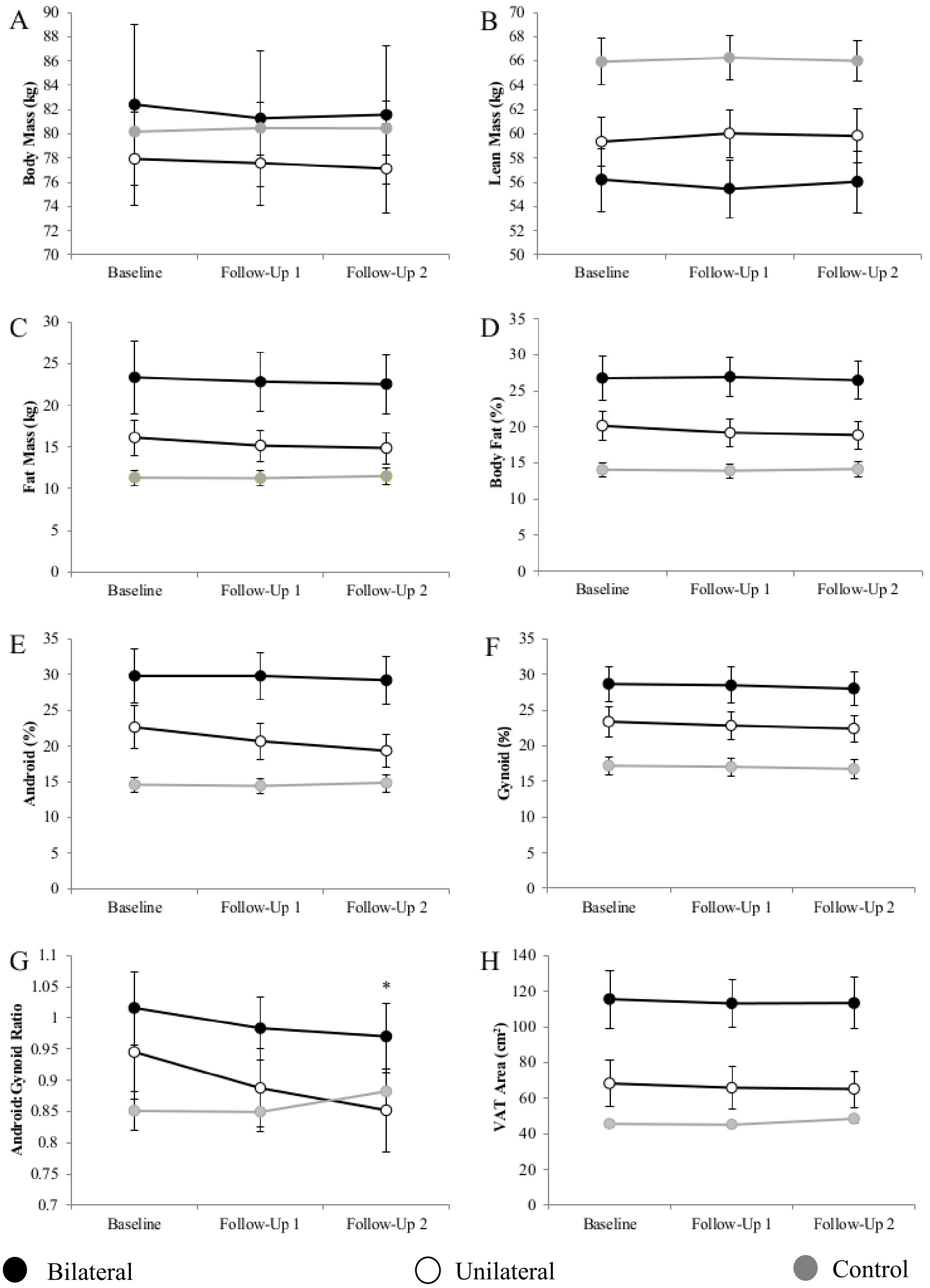


Figure 7.4: Body composition at baseline, 10 weeks and 20 weeks follow-up in unilateral (n = 8) and bilateral (n = 8) amputees and normative controls (n = 13). A, body mass (kg); B, lean muscle mass (kg); C, fat mass (kg); D, percentage body fat; E, percentage android fat; F, percentage gynoid fat; G, android: gynoid ratio; H, visceral adipose tissue (VAT) area (cm^2). Black dots represent bilateral amputees, white dots represent unilateral amputees, grey dots represent normative controls. * A significant difference between individuals with unilateral amputation and normative control ($p < 0.05$). Data are presented as mean and standard error.

7.3.3. Cardiometabolic Health

7.3.3.1. Lipid Profiles, Markers of Inflammation and Blood Pressure

When comparing baseline cardiovascular risk, there was a significant main effect of group on total cholesterol:HDL cholesterol (TC:HDL) ratio (F(2,26) = 12.018, p < 0.001), triacylglycerol (F(2,26) = 10.250, p = 0.001), NEFA (F(2,26) = 4.223, p = 0.026), CRP (F(2,26) = 9.185, p = 0.001), systolic BP (F(2,26) = 29.791, p < 0.001) and diastolic BP (F(2,26) = 7.875, p = 0.002) (see table 6.2). A trend towards a significant main effect of group was demonstrated in HDL cholesterol (F(2,26) = 2.781, p = 0.080). Compared to active normative controls, individuals with bilateral amputation demonstrated significantly increased TC:HDL ratio (p < 0.001), triacylglycerol (p = 0.001) and CRP (p = 0.002), systolic BP (p < 0.001) and diastolic BP (p = 0.002). *Post-hoc* analysis also revealed significant differences between individuals with unilateral and bilateral amputation for TC:HDL ratio (p = 0.002), triacylglycerol (p = 0.003), NEFA (p = 0.030) and CRP (p = 0.004), systolic BP (p = 0.001) (see table 1). No significant differences in lipid profiles or inflammation were reported between individuals with unilateral LLA and active normative controls, there was however a significantly greater systolic BP (p = 0.020) demonstrated in the unilateral LLA group.

There was a significant main effect of time on total cholesterol (F(1,26) = 5.400, p = 0.028), HDL cholesterol (F(1,26) = 4.366, p = 0.047) and LDL cholesterol (F(1,26) = 10.068, p = 0.004). Significant time x group interaction effects were demonstrated in systolic BP (F(2,26) = 5.371, p = 0.011). No significant time x group interaction effects were demonstrated in any lipid profile or inflammatory health marker, however a trend towards an interaction effect was reported with LDL cholesterol (F(2,26) = 3.224, p = 0.056). *Post-hoc* analyses revealed significant large effect reductions in total cholesterol (*d* = 1.11, p = 0.029), HDL cholesterol (*d* = 1.26, p = 0.032) and LDL cholesterol (*d* = 1.05, p = 0.030) between baseline and 20 weeks follow-up in the bilateral LLA group only. The unilateral amputation group demonstrated a significant large effect size reduction in systolic BP (*d* = 1.0, p = 0.005) over the study duration.

Table 7.2: Cardiovascular health values at baseline and reported change after 20 weeks in individuals with unilateral and bilateral amputation and normative uninjured controls. Data presented as mean ± SD and mean change Δ (95%CI)

Marker	Unilateral (n = 8)		Bilateral (n = 8)		Control (n = 13)	
	Baseline	**Δ (95% CI)**	**Baseline**	**Δ (95% CI)**	**Baseline**	**Δ (95% CI)**
Total Cholesterol, mmol·L^{-1} *	3.1 ± 1.1	-0.1 (-0.8 to 0.6)	3.6 ± 0.4	-0.7 (-1.3 to -0.1)	3.2 ± 0.9	-0.2 (-0.5 to 0.2)
HDL Cholesterol, mmol·L^{-1} *	0.9 ± 0.3	-0.0 (-0.3 to 0.2)	0.8 ± 0.1	-0.1 (-0.3 to 0.0)	1.1 ± 0.4	-0.1 (-0.2 to 0.1)
LDL Cholesterol, mmol·L^{-1} *	1.9 ± 1.0	-0.1 (-0.6 to 0.4)	2.6 ± 0.4	-0.6 (-1.0 to -0.2)	2.0 ± 0.6	-0.2 (0.4 to 0.1)
TC:HDL ratio ¶ †	3.3 ± 0.6	0.1 (-0.1 to 0.4)	4.8 ± 0.9	-0.1 (-0.4 to 0.2)	3.2 ± 0.8	0.0 (-0.3 to 0.3)
Triacylglycerol, mmol·L^{-1} ¶ †	0.7 ± 0.3	0.2 (0.0 to 0.3)	1.8 ± 1.0	-0.1 (-0.8 to 0.5)	0.7 ± 0.3	0.2 (0.0 to 0.4)
NEFA, mmol·L^{-1} ¶	0.3 ± 0.2	0.1 (-0.1 to 0.3)	0.6 ± 0.3	-0.1 (-0.4 to 0.1)	0.4 ± 0.1	0.1 (-0.2 to 0.1)
C-Reactive Protein, mg.L^{-1} ¶†	0.40 ± 0.28	0.3 (-0.2 to 0.7)	2.85 ± 2.56	-0.4 (-2.8 to 2.1)	0.40 ± 0.48	0.2 (-0.4 to 0.8)
Systolic BP (mm Hg) #¶	125 ± 6	-7 (-11 to -3)	137 ± 8	-4 (-8 to 0)	118 ± 2	-1 (-2 to 0)
Diastolic BP (mm Hg)	69 ± 10	-3 (-11 to 6)	78 ± 9	-2 (-8 to 5)	63 ± 7	1 (-2 to -2)

Abbreviations: HDL = high-density lipoprotein, LDL = low density lipoprotein, TC:HDL total cholesterol : high-density lipoprotein, NEFA = nonesterified fatty acid, BP = blood pressure

* Significant difference between baseline and follow-up in individuals with bilateral amputation ($p < 0.05$)
# Significant difference between baseline and follow-up in individuals with unilateral amputation ($p < 0.05$)
† Significant difference between bilateral amputation and normative controls ($p < 0.05$)
¶ Significant difference between bilateral amputation and unilateral amputation ($p < 0.05$)

7.3.3.2. Insulin and Glucose Response to OGTT

Serum Insulin

No significant main effect of group was demonstrated for levels of fasted serum insulin concentrations at baseline ($p > 0.05$) (see table 6.3). However, a significant main effect in insulin concentrations response following a 75g OGTT challenge were demonstrated between groups in peak insulin ($F(2,26) = 5.139$, $p = 0.013$) and iAUC ($F(2,26) = 8.316$, $p = 0.002$). There was a main effect of group on insulin concentration at the following time-points; 45minutes ($F = 4.463$, $p = 0.022$), 60 minutes ($F = 4.967$, $p = 0.015$), 90 minutes ($F = 14.877$, $p < 0.000$), 120 minutes ($F = 8.011$, $p = 0.002$). A trend towards a significant main effect was demonstrated at 30 minutes ($F = 3.276$, $p = 0.054$). *Post-hoc* analyses reveal significantly greater insulinemic response in individuals with bilateral amputation compared to the control group at 45minutes ($p = 0.024$), 60 minutes ($p = 0.017$), 90 minutes ($p < 0.001$) 120 minutes ($p = 0.001$) after a glucose load with greater peak insulin levels ($p = 0.015$), and iAUC ($p = 0.002$) (figure 6.5). The mean time to peak insulin concentrations were 10 minutes longer in the bilateral amputations group compared to normative controls, but this difference was not statistically significant ($p > 0.05$). Individuals with bilateral amputations demonstrate significantly greater iAUC ($p = 0.017$) and a trend toward significantly greater peak insulin ($p = 0.061$) compared to individuals with unilateral amputation. No significant differences in insulinemic response to glucose loading were demonstrated between individuals with unilateral amputation and active normative controls ($p > 0.05$).

There was no time or time x group interaction effect in serum insulin response during the OGTT. Although the mean iAUC values did not change statistically within any group ($p > 0.05$) the shape of the AUC did change for the bilateral amputations group and this may be clinically significant (see figure 6.5C and 6.5D). There was a medium effect size reduction of 7.5 ± 21 minutes in time to peak insulin values ($d = 0.62$). A medium ($d = 0.50$) effect size increase in serum insulin concentration were recorded in the 30 minutes post-glucose solution ingestion, followed by a medium ($d = 0.62$) and small ($d = 0.039$) effect size decreases in serum insulin at 60 and 120 minutes post-glucose ingestion, respectively. Peak insulin levels remained unchanged. Changes in insulinemic response (iAUC) to glucose loading did not occur to this magnitude in the unilateral amputation group or normative controls.

Plasma Glucose

No significant main effect of group was demonstrated for fasting levels of plasma glucose concentrations ($p > 0.05$) (see table 7.3). Following a 75g OGTT challenge there was a significant main effect of group in the time to peak glucose concentration ($F(2,26) = 4.859$, $p = 0.016$) and plasma glucose concentrations at 60 minutes ($F = 5.418$, $p = 0.011$). A trend towards a significant main effect of group was demonstrated at 30 minutes post-glucose ingestion ($F = 3.276$, $p = 0.054$)

and for glycaemic response (AUC) (F(2,26) = 3.032, p = 0.066). *Post-hoc* analyses found significantly greater plasma glucose concentrations in individuals with bilateral LLA at 60 minutes post-glucose ingestion (p = 0.010) and a significantly greater mean time to peak glucose (p = 0.016) compared to normative controls (see figure 7.5).

No time or time x group interaction effect in plasma glucose response to the OGTT was found during the study ($p > 0.05$). Despite mean glycemic response (AUC) not changing statistically over the 20 week period, the shape of the glucose AUC appears to change in a similar way to serum insulin concentration (see figure 6.5). Peak plasma glucose concentrations in the bilateral amputation group did not change, however a large effect size reduction ($d = 0.96$) in time to peak glucose concentration (9 ± 16 minutes) was demonstrated. A medium ($d = 0.50$) and small ($d = 0.43$) effect size increase in plasma glucose values were recorded in the first 15 minutes and 30 minutes post-glucose ingestion, respectively. Followed by medium and small effect decreases in glucose concentration at 60 minutes ($d = 0.56$) and 120 minutes ($d = 0.48$), respectively. While the plasma glycemic response (AUC) to a 75g OGTT challenge in the bilateral amputations group were not statistically significant, it may still be clinically relevant. No change in glycaemic response to the 75g OGTT challenge occurred over 20 weeks in the unilateral amputation group of normative controls.

Table 7.3: Metabolic health values at baseline and reported change after 20 weeks in individuals with unilateral and bilateral amputation and normative uninjured controls. Data presented as mean ± SD and mean change Δ (95% CI)

Marker	**Unilateral (n = 8)**		**Bilateral (n = 8)**		**Control (n = 13)**	
	Baseline	**Δ (95% CI)**	**Baseline**	**Δ (95% CI)**	**Baseline**	**Δ (95% CI)**
Fasted Glucose, mmol·L^{-1}	5.7 ± 0.4	-0.1 (-0.4 to 0.2)	5.5 ± 0.6	0.1 (-0.2 to 0.5)	5.5 ± 0.4	0.0 (-0.2 to 0.3)
Peak Glucose, mmol·L^{-1}	9.9 ± 1.5	-0.6 (-1.8 to 0.6)	10.1 ± 1.5	-0.1 (-2 to 1.9)	9.4 ± 1.4	0.1 (-1.0 to 1.2)
Time to peak Glucose (minutes) #	35.6 ± 11.2	1.9 (-12.2 to 16.0)	45.0 ± 8.1	-9.4 (-22.7 to 4.0)	33.5 ± 6.6	0.0 (-7.4 to 7.4)
Glucose AUC, mmol·L·120min^{-1}	352.3 ± 90.5	8.0 (-105.0 to 120.9)	450.4 ± 103.5	-36.9 (-191.4 to 117.5)	337 ± 115.3	-23.2 (-128.8 to 82.5)
Fasted Insulin, pmol·L^{-1}	30.5 ± 25.4	2.1 (-4.4 to 8.7)	49.2 ± 36.3	-7.5 (-30.9 to 15.9)	22.4 ± 9.4	0.7 (-6.1 to 7.5)
Peak Insulin, pmol·L^{-1} #	282.2 ± 99.3	-27.7 (-84.3 to 29.0)	482.5 ± 254.2	-38.3 (-133.6 to 56.9)	258.7 ± 116.1	3.6 (-49.4 to 56.5)
Time to Peak Insulin (minutes)	43.1 ± 12.5	0.0 (-11.6 to 11.6)	48.8 ±13.3	-7.5 (-25.2 to 10.2)	39.2 ± 9.8	-2.3 (-8.6 to 3.9)
Insulin AUC, pmol·L·120min^{-1} # †	2769 ± 1110	-147 (-803 to 508)	4859 ± 1750	-229 (-1645 to 1188)	2389 ±1292	-84 (-494 to 327)
HOMA-IR	1.11 ± 0.91	0.0 (-0.2 to 0.3)	1.80 ± 1.46	-0.3 (-1.3 to 0.6)	0.78 ± 0.32	0.0 (-0.2 to 0.3)
HOMA-β % #	40.5 ± 34.7	6.5 (-2.1 to 15.1)	69.4 ± 36.1	-11.1 (-28.4 to 6.2)	33.8 ± 17.3	-1.5 (11.5 to 8.5)
HOMA2-IR	0.63 ± 0.45	0.0 (-0.0 to 0.1)	0.96 ± 0.70	-0.1 (-0.6 to 0.3)	0.47 ± 0.14	0.0 (-0.1 to 0.1)
HOMA2-β % #	51.1 ± 23.9	-0.9 (-14.6 to 12.8)	70.5 ± 23.9	-6.5 (-18.9 to 6.0)	46.3 ± 12.4	-0.6 (-8.3 to 7.2)
HOMA2-S	201.5 ± 72.5	-17.6 (-51.4 to 16.2)	150.8 ± 80.9	-12.9 (-65.0 to 39.2)	223 ± 42.9	-7.0 (-43.5 to 29.5)
ISI $_{\text{Matsuda index}}$	10 .8 ± 4.3	1.4 (-2.4 to 5.2)	6.9 ± 3.4	-0.6 (-3.0 to 1.8)	12.9 ± 3.8	-0.3 (-2.8 to 2.2)

Abbreviations: AUC = area under the curve, HOMA-IR = homeostasis model assessment for insulin resistance, HOMA-β homeostasis model assessment for β-cell function, HOMA-S = homeostasis model assessment for insulin sensitivity, ISI = Insulin Sensitivity Index

Significant difference between bilateral amputation and normative controls ($p < 0.05$)
† Significant difference between bilateral amputation and unilateral amputation ($p < 0.05$)

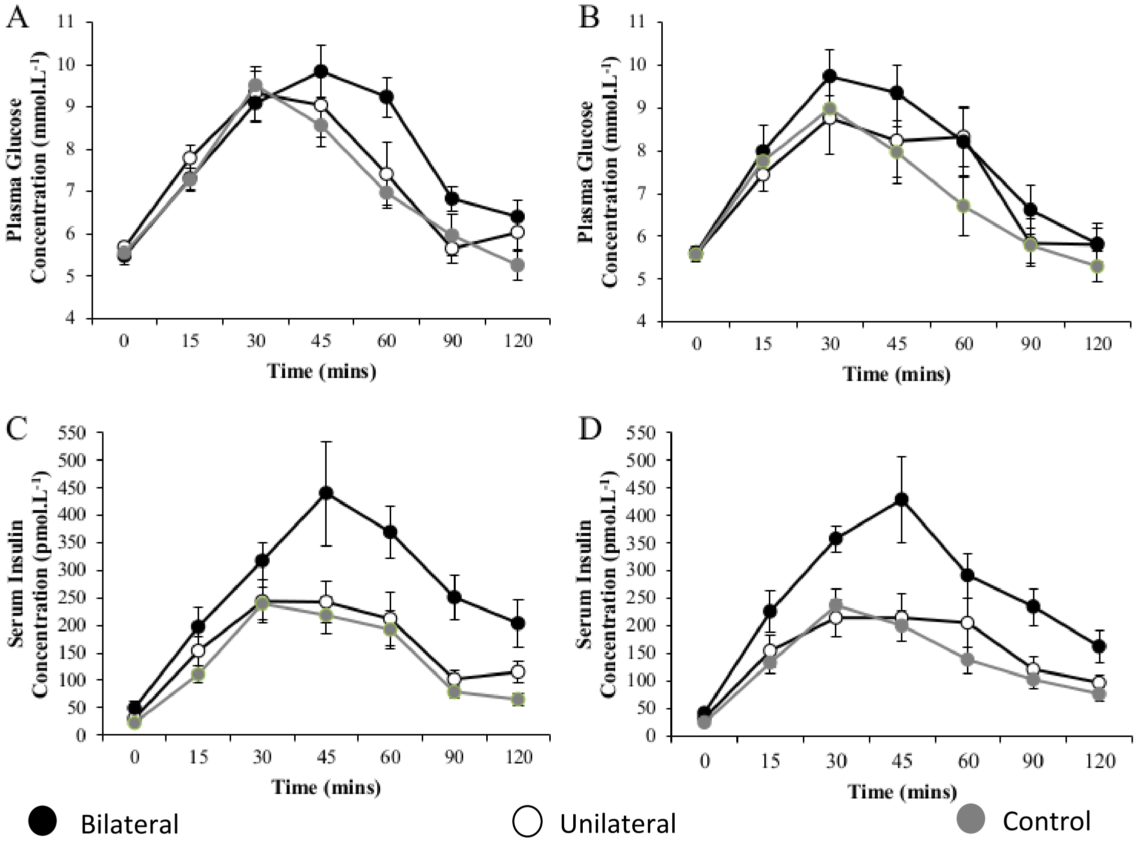


Figure 7.5: Plasma glucose and serum insulin response during a modified oral glucose tolerance test (OGTT). Graph A and B are the plasma glucose concentration responses, C and D are the serum insulin concentration responses. Graph A and C are baseline values, B and D are 20 week follow-up values. Black dots represent bilateral amputation, white dots represent unilateral amputation and grey dots are normative controls. Data are presented as mean and standard error.

7.3.3.3. Indices of Insulin Sensitivity/Resistance

When comparing baseline biomarkers metabolic health risk, a main effect of group for HOMA-β (F(2,26) = 4.022, p = 0.030), HOMA2-β (F(2,26) = 4.026, p = 0.030) and ISI Matsuda (F(2,26) = 6.128, p = 0.007). A trend towards a significant main effect of group was demonstrated in HOMA-IR (F(2,26) = 3.027, p = 0.066), HOMA2-IR (F(2,26) = 2.962, p = 0.069) and HOMA2-S (F(2,26) = 3.245, p = 0.055). *Post-hoc* analyses confirmed that individuals with bilateral amputation demonstrated significantly higher fasted estimates of pancreatic β-cell function (HOMA-β) (p = 0.030) and HOMA2-β (p = 0.030), and significantly lower ISI-Matsuda (p = 0.005) compared to normative controls (see table 7.3). No significant differences were reported between individuals with unilateral amputation and normative controls. No significant time or time x group interaction effect was demonstrated for any indices of insulin sensitivity/resistance (table 7.3).

7.3.3.4. Metabolic Syndrome and Cardiovascular Disease Risk Factors

Elevated levels of fasted plasma glucose concentration and reduced HDL cholesterol were the most common risk factors present across all groups (table 6.4). However, the bilateral amputation group presented the highest number of cardiovascular disease and metabolic syndrome risk factors, with metabolic syndrome being present in 63%.

Table 7.4: Component factors used to predict metabolic syndrome risk factors. Data presented as number of participants and prevalence (%)

Metabolic Syndrome Risk Factor	Unilateral LLA		Bilateral LLA		Control	
	Baseline	**20 weeks**	**Baseline**	**20 weeks**	**Baseline**	**20 weeks**
Waist circumference $\geq$ 94 cm	0	-	6 (75%)	-	0	-
Hypertriglyceridemia: $\geq$ 1.7 mmol·L^{-1}	0	0	3 (38%)	3 (38%)	0	0
Hyperglycaemia: FPG $\geq$ 5.6 mmol·L^{-1}	4 (50%)	3 (38%)	4 (50%)	4 (50%)	6 (46%)	6 (46%)
Hypertension: Systolic BP $\geq$ 130 mm Hg	0	0	4 (50%)	4 (50%)	0	0
Hypertension: Diastolic BP $\geq$90 mm Hg	1 (13%)	0	2 (25%)	1 (13%)	0	0
HDL cholesterol: < 1.03 mmol·L^{-1}	5 (63%)	6 (75%)	8 (100%)	8 (100%)	5 (38%)	8 (62%)
Metabolic Syndrome ($\geq$ 3 component risk factors)	0	0	5 (63%)	5 (63%)	0	0

Cut-points are determined using the International Diabetes Foundation published criteria for metabolic syndrome (Zimmet et al, 2005).

Abbreviations: BP = blood pressure, HDL = high density lipoprotein

7.4. DISCUSSION

This study investigated the impact of 20 weeks of the UK DMS rehabilitation paradigm for complex trauma casualties with LLA at DMRC, Headley Court. Changes in clinical outcomes, body composition and cardiometabolic health were assessed before and after two 4-week in-patient admissions of intensive rehabilitation followed by 6 weeks of habitual recovery at home, clinical outcomes and body composition were also assessed at the 10-week mid-point (see figure 6.1 - study design). With regards to the primary hypothesis, these results demonstrate that individuals with unilateral LLA have a comparable body composition and cardiometabolic disease risk as active age-matched normative controls and levels of ambulatory function comparable to the general population. Individuals with bilateral LLA demonstrate unfavourable body composition commensurate with being classified as overweight / obese. These body composition values were accompanied by increases in numerous indices of cardiovascular disease risk factors and compromised metabolic health. There were no significant differences between baseline values and 20 weeks follow-up in any body composition, or cardiometabolic health markers except for reductions in cholesterol concentrations (particularly atherogenic lipids) reported in individuals with bilateral LLA. However, clinically significant improvements in function were demonstrated over 20 weeks follow-up in both unilateral and bilateral amputation groups.

7.4.1. Changes in Clinical Outcomes

7.4.1.1. Function

Despite mean 14 ± 8 months and 39 ± 15 months of IDT rehabilitation in the unilateral and bilateral LLA groups, respectively, both groups continue to demonstrate clinically significant improvements in their physical function. The greatest functional improvements demonstrated over 20 weeks by the unilateral amputation group were increases in the prevalence ‘able to perform ADL’ and ‘run independently’. All members of the unilateral LLA group could walk independently and comparable distances to general population at baseline (see chapter 6, table 6.3), they also appear to have reached a plateau in distance walked in 6 minutes and scored near maximal scores in the AMPQ assessment. Over the 20 weeks, the bilateral LLA group attained minimal clinically significant improvement (> 45 metres (Resnik & Borgia, 2011)) in the 6MWT of 54 metres and an increase in the prevalence ‘able to walk independently’ during the 20 week monitoring period. All individuals with bilateral LLA were able to ‘perform ADL with an aid or adaptation’ by the end of the 20 weeks and none were dependent on a wheelchair to mobilise.

Previous research has found limitations to using the AMPQ due to a commonly reported ceiling effect in high functioning patients (Gailey et al, 2013a). The results from the unilateral LLA cohort support this finding with the majority scoring near maximum (mean 46 ± 1 out of max score of 47), thus questioning the practical utility of this assessment in high functioning military personnel with unilateral LLA. Halfway through data collection, the complex trauma department at DMRC began using an alternative functional assessment that accommodates higher level of function. These data are not reported as part of this thesis as there were no baseline data available for comparison. When I compare the clinical outcomes recorded at the end of this 20 week observation study to the larger cohort attending their last admission at DMRC in chapter 4, I found comparable levels of objective ambulatory performance. Individuals with unilateral and bilateral LLA in this study (chapter 6 and 7) could walk 598 ± 68m and 391 ± 99m in 6 minutes respectively, compared to 544 ± 99m and 445 ± 104m, in the larger cohort collected in chapter 4 (Ladlow et al, 2015). The individuals with bilateral LLA also score similarly on the AMPQ (42 ± 3 in this study compared to 42 ± 5 demonstrated in chapter 4), indicative of a level of function comparable to community ambulators with LLA (Gailey et al, 2002).

7.4.1.2. Psychosocial Health

Mean psychosocial health outcomes remained unchanged throughout the duration of the study and, as discussed in chapter 6, similar depression (mean PHQ-9 = 2 ± 1) and anxiety scores (GAD-7: unilateral = 2 ± 1, bilateral = 0 ± 1) were found at the end of rehabilitation to those demonstrated in chapter 4 (PHQ-9 = 3 ± 5, GAD-7 = 3 ± 4), with all mean scores indicative of no symptoms. There are numerous explanations why mental health outcomes may remain low during rehabilitation at DMRC, but are likely to be patient specific. The acceptance of a changed body image over time, higher levels of active coping, an optimistic personality disposition, increased levels of social support, greater satisfaction with the prosthesis, and decreased pain levels all contribute to better mental health outcomes over time have all been suggested (Horgan & MacLachlan, 2004). In chapter 4, it was proposed that a goal-oriented exercise rehabilitation program will improve function and ADL over time in amputees, thereby promoting a closer identification with their pre-injured self (Ladlow et al, 2015).

7.4.2. Baseline Body Composition

Despite losing two lower-limbs, the total body mass of the bilateral LLA group recorded at baseline is comparable to the normative control group, yet the amount of body fat mass stored is 2-fold greater (24 kg versus 11 kg, respectively). The location of this increased fat mass appears to be around the abdomen (waist circumference: 101 ± 22cm, android fat percentage: 30 ± 11%, and VAT area 117 ± 47cm^2) which is known to be a risk factor for future cardiovascular and metabolic related disorders. Body composition changes in the first year following traumatic amputation have previously been demonstrated in US military servicemen (Eckard et al, 2015). Eckard et al. (2015)

found significant increase in fat mass occurring in the first 9 months following unilateral amputation. They also found a clinically relevant, but not statistically significant, weight gain of 14.5 kg from baseline to 12 months in individuals with bilateral LLA with both groups exceeded their pre-injury BMI by 6 months post-amputation. This is particularly relevant to this study population, as the US and UK militaries both invest heavily in rehabilitation services and advanced prosthetic provision, yet these predominantly young, physically and mentally determined servicemen with traumatic LLA are not immune from significant (unfavourable) weight gain following traumatic injury. Older studies have also demonstrated increases in body fat mass during the first year after amputation and increases in body fat mass were directly related to the level of LLA (Kurdibaylo, 1996). Kurdibaylo et al. (1996) found the larger increases in obesity progression in individuals with bilateral transfemoral or transfemoral/transtibial amputation with both groups averaging 25.9% body fat after one year, which is comparable to this bilateral LLA group (26.9 ± 8.7%). There were no statistically significant differences in body composition between the unilateral LLA group and active normative controls. One explanation for this may be the capacity of UK military personnel with unilateral LLA to perform comparable levels of PAEE to active normative controls, as demonstrated in chapter 6. The access to advanced prosthetic technology in combination with regular in-patient IDT rehabilitation admissions at DMRC are also likely to have facilitated these desirable outcomes which include an reasonably high prevalence (63%) able to run independently whilst wearing their prosthesis.

Significant muscle atrophy and diminished muscle strength following amputation can complicate the ability to perform ADL (Moirenfeld et al, 2000) and may negatively influence the function of the residual limb and prosthesis (Renstrom et al, 1983b). The level of amputation and length of the residual limb are significant factors in the severity of muscle atrophy (Isakov et al, 1996; Jaegers et al, 1995). Although site specific lean muscle tissue mass was not measured (i.e. quadriceps cross-sectional area or volume) the bilateral amputation group did demonstrated significantly reduced total lean body mass (56.8 ± 8.5 kg) compared to active normative controls (65.9 ± 6.9 kg) and when considered as a percentage of relative total body mass this equates to a 14% reduction in (mostly metabolically active) lean muscle tissue. The quantity and quality of skeletal muscle is known to have a significant role to play in the development of insulin resistance (see chapter 2, section 2.7.2). The reduced amount of whole body lean muscle tissue demonstrated by individuals with bilateral LLA may have been a precursor to their increased risk of insulin resistance; however increases in insulin resistance may also cause an individual to store more adipose tissue.

7.4.3. Changes in Body Composition during the 20 Weeks Rehabilitation Period

There were no significant differences between baseline, 10 weeks and 20 weeks follow-up in any body composition markers ($p > 0.05$). The unchanged body composition (and cardiometabolic health markers) during this 20 week study period both relate to energy balance. Due to the impact of VAT storage and lean muscle mass on metabolic regulation, should body composition not change throughout the duration of this study, it is not surprising that cardiometabolic health outcomes did not change either.

7.4.4. Baseline Cardiovascular and Metabolic Health

7.4.4.1. Indices of Cardiovascular Disease Risk

Cardiovascular risk factors such as age, sex, smoking, diabetes, hypertension, elevated triglycerides, CRP, LDL cholesterol and low HDL cholesterol levels have all been widely established as independent cardiovascular risk factors in able bodied adults (Millan et al, 2009). Total cholesterol concentration, especially LDL cholesterol, is considered an atherogenic lipid marker. Mean concentration levels of these isolated lipid markers are low in all three groups with no individuals demonstrating elevated risks for hypercholesterolemia ($\geq$ 5.2 mmol$\cdot$L^{-1}) or high LDL cholesterol ($\geq$ 3.5 mmol$\cdot$L^{-1}) (table 7.2). However, mean baseline levels of HDL cholesterol were below 1 mmol$\cdot$L^{-1} in both amputation groups. Lipoprotein ratios based on two values (total and HDL cholesterol), indicate a relationship or proportion between the atherogenic and anti-atherogenic lipid fractions. These ratios have greater predictive power for cardiovascular disease risk superior to isolated lipid parameters alone, particularly LDL cholesterol (Millan et al, 2009). The TC:HDL cholesterol ratio in this study are within normal healthy range in the unilateral and normative control groups, but elevated in individuals with bilateral LLA (mean 4.8 ± 0.9 mmol$\cdot$L^{-1}) with 50% demonstrating a ratio > 5, indicative of moderate atherogenic risk (Millan et al, 2009). When TC:HDL cholesterol is used in combination with other baseline markers of cardiovascular disease risk [elevated mean triglyceride concentrations, waist circumference, VAT area, systolic BP, inflammatory markers (CRP) and reduced PAEE] the findings indicate that UK military personnel with bilateral LLA are at an increased risk of developing cardiovascular disease.

Previous research indicates, individuals with bilateral LLA appear to have two times greater risk of developing metabolic disorders including obesity, hypertension, hyperlipidemia, and hyperinsulinemia (Ejtahed et al, 2017; Magalhaes et al, 2011; Naschitz & Lenger, 2008). Ninety-five percent of injured Iranian male servicemen with combat-related traumatic bilateral LLA presented with at least one modifiable cardiovascular disease risk factor, significantly higher than population norms (Shahriar et al, 2009). In short, the prevalence of risk factors found in Shahriar's study included: hyperglycemia 13.1%, systolic hypertension 18.9%, diastolic hypertension 25.6%, abdominal obesity 82.5%, high total cholesterol 36.7%, low HDL 25.9%, high LDL 24.7% and high triglycerides 32.1%. In this study, when using the component risk factors for developing metabolic syndrome, hyperglycaemia and reduced HDL cholesterol where the most common components presented across all groups (see table 7.4). However, the bilateral amputations group presented with the most components of metabolic syndrome risk, with obesity (75%) the most prominent component, which facilitated the accumulation of 63% of this bilateral LLA cohort presenting with metabolic syndrome. This elevated prevalence of metabolic syndrome demonstrated in the bilateral LLA group is comparable to Ejtahed et al. (2017) who reported metabolic syndrome in 62% of Iranian military veterans with combat-related bilateral LLA. Greater muscle strength and lean muscle tissue is associated with lower prevalence of abnormal metabolic syndrome components (Roberts et al, 2013), with each of the five components of metabolic syndrome inversely associated with muscular strength (Jurca et al, 2004). The complex task of generating lean muscle tissue in the lower limbs of bilateral LLA is well documented (see chapter 2 section 2.4 and chapter 3 section 3.4).

Stewart et al. (2015) performed a retrospective analysis of 3846 US injured military personnel wounded between 2002 and 2011 during the recent conflicts in Iraq and Afghanistan. They predicted each 5-point increment in the ISS was associated with a 6% and 13% increase in incidence rates of hypertension and coronary artery disease, respectively. They estimated incidence rates of hypertension and coronary artery diseases for the most severely injured patients (ISS > 25) were 2.5 to 4-fold higher than published rates for the overall US military population, respectively (Crum-Cianflone et al, 2014; Granado et al, 2009). Chapter 4 reported mean NISS of 44 ± 12 and 28 ± 11 for UK military personnel with bilateral and unilateral LLA, respectively (Ladlow et al, 2015). The data in this chapter supports the theory that increases in injury severity are associated with increased risk of developing cardiovascular disease. These projected outcomes provided by Stewart et al. (2015) much like the ones reported here, were observed after a relatively short period of 1 to 3 years post-injury which may have profound implications for both those wounded during combat and for government departments responsible for funding long-term care of veterans. The unilateral LLA group demonstrate a low risk of developing cardiovascular-related diseases following their injury, similar to the active normative control group.

7.4.4.2. Insulin Resistance / Sensitivity

Two older studies report impaired metabolic health in individuals with traumatic amputation (Modan et al, 1998; Peles et al, 1995). Both Peles et al. (1995) and Modan et al. (1998) investigated an older traumatic LLA population. Fasted glucose values were comparable to my study (Peles et al: 6.3 ± 1.6 mmol·L^{-1}, Modan et al: 6.0 ± 1.6 mmol·L^{-1}). However, hyperinsulinemia was evident in this older cohort with traumatic amputation (Peles et al: 128 ± 67.4 pmol·L^{-1}, Modan et al: 123 ± 59 pmol·L^{-1}) with fasted values considerably greater than the unilateral (30.5 ± 25.4 pmol·L^{-1}) and bilateral (49.2 ± 36.3 pmol·L^{-1}) LLA groups who participated in this study. However, the bilateral

LLA group did have a fasted insulin concentration 120% greater than the active normative control group.

Recently it has become more common to provide additional surrogate indices to predict whole body insulin sensitivity (ISI Matsuda Index (Matsuda & DeFronzo, 1999)) and insulin resistance (HOMA-IR; (Turner et al, 1979)) and β-cell function (HOMA- β; (Matthews et al, 1985) and the HOMA2 calculator. To my knowledge this is the first time HOMA/HOMA2 and Matsuda Index have been used to estimate insulin resistance/sensitivity in individuals with traumatic LLA and certainly military personnel injured during the Iraq/Afghanistan conflicts. The HOMA2 data must be interpreted with caution though because 3 individuals with unilateral amputation, 1 with bilateral LLA and 6 normative controls demonstrated fasted insulin values that were below the concentration required by the HOMA2 calculator to derive fasting estimates of β-cell function and insulin resistance accurately. In these scenarios, insulin concentrations had to be artificially elevated to 20 pmol·L^{-1} so that a value could be provided. This indicates a heightened level of sensitivity to blood glucose in the unilateral LLA group as only very small amounts of insulin secretion are required to regulate blood glucose. Insulin sensitivity data measured using the ISI (Matsuda) reveal a mean value for bilateral LLA group (6.9 ± 3.4), half the value (47 %) of normative controls (12.9 ± 3.8), but > 5.0 which is considered the cut-off point for insulin resistance (Radikova et al, 2006). When I compare the mean values reported in individuals with bilateral amputation (where a greater proportion had fasting values of insulin that could be incorporated into the HOMA2 calculator) with other disabled groups, I note similarities in fasting glucose, fasting insulin, HOMA2-IR, HOMA2-β % and ISI Matsuda profiles as individuals with spinal cord injury (Nightingale et al, 2017). Frequent wheelchair dependency to mobilise in the community and home environment may have a large role to play in the similarities observed in metabolic health between these two disabled populations. The current study found UK military personnel with unilateral amputation demonstrate comparable metabolic health profiles to active normative controls.

An additional mechanism behind an increased risk of insulin resistance in the bilateral LLA group could be attributed towards their elevated injury severity scores. Stewart et al. (2015) estimated incidence rates of diabetes mellitus for the most severely injured patients from Iraq/Afghanistan (ISS > 25) to be 2.6 fold higher than published rates for the overall US military population (Boyko et al, 2010). Much like the cardiovascular disease risk mentioned previously, a particularly compelling finding considering the short length of time (< 3 years) since their injuries occurred. Individuals with bilateral LLA also demonstrated increased VAT area, reduced lean muscle tissue and cardiorespiratory fitness. It is well documented that muscle atrophy, diminished muscle strength and increased abdominal fat mass commonly occur after amputation (Eckard et al, 2015;

Isakov et al, 1996; Jaegers et al, 1995; Kurdibaylo, 1996; Renstrom et al, 1983b; Schmalz et al, 2001; Shahriar et al, 2009; Sherk et al, 2010) and skeletal muscle is a critical glucose disposal site during postprandial conditions (Booth et al, 2017). Physical inactivity promotes insulin resistance through reduced utilization of glucose in skeletal muscle, but it may also promote the increased storage of glucose and free fatty acids into adipose tissue. As such, adiposity and insulin resistance may develop at the same time through these processes (Booth et al, 2017). Insulin resistance is associated with reduced quadriceps muscle strength (Kalyani et al, 2013; Leenders et al, 2013), muscle mass (Leenders et al, 2013) and power (Kalyani et al, 2013) which are all known to occur following traumatic LLA (chapter 2, section 2.6.2). It is not obvious how much muscle mass each participant lost following bilateral LLA group. However, should the control group be used as an indicator; the bilateral LLA lost a mean 9kg (or 14%) of metabolically active tissue. This is likely to have considerable long-term implications for metabolic health. Unfortunately, even the most effective of exercise interventions are unlikely to restore this quantity of muscle tissue in individuals with complex primary and secondary injuries/conditions common to bilateral LLA (refer to chapter 2 section 2.3 and chapter 3 for greater detail). Investigating strategies that mitigate for muscle atrophy and weakness in military personnel with bilateral LLA is certainly an area worthy of future scientific exploration

7.4.5. Changes in Cardiovascular and Metabolic Health Over 20-weeks Rehabilitation

There is a distinct lack of research looking at the effect of IDT rehabilitation on the response to a standardised glucose load in individuals with traumatic LLA. There were no significant differences between baseline and 20 weeks follow-up in any cardiometabolic health markers except for reductions in cholesterol values reported in individuals within the bilateral LLA group. As cardiometabolic health is regulated by lean muscle and adipose tissue, and changes were not observed in body composition (see figure 7.4), this finding is perhaps not surprising.

It is difficult to determine which component of the bilateral LLA group rehabilitation or lifestyle at home is responsible for the significant large effect size reductions in atherogenic lipid cholesterol concentrations. However, in uninjured individuals, greater improvements in lipid profiles have been observed following high-intensity exercise (O'Donovan et al, 2005) or with higher amounts of weekly exercise (Kraus et al, 2002). As ambulatory and physical function continue to improve throughout the duration of the study, this extra capacity to engage in more intense forms of exercise may have a role to play in the positive changes in cholesterol observed. There were no significant differences in serum CRP measured in the study. The inflammatory cells (i.e., macrophages) that infiltrate into adipose tissue are responsible for the secretion of inflammatory cytokines (Thompson et al, 2012), which trigger the acute phase response, whereby proteins, such as CRP, are released from the liver (Moshage, 1997). Weight loss is therefore associated with a decline in CRP (Selvin et al, 2007). A lack of significant changes in total body mass and fat mass is likely responsible for no changes in serum CRP. It remains to be seen whether the combination of exercise and dietary restrictions are the most effective way of improving lipid profiles in persons with traumatic LLA

No statistically significant changes in either amputation group were observed for plasma glycaemic or serum insulinemic responses (AUC) to IDT rehabilitation. However, some moderate effect size changes in insulin concentrations throughout the OGTT were demonstrated altering the shape of the AUC in the bilateral LLA group over the duration on the study. Early glucose response during an OGTT can be considered an index of hepatic insulin resistance, while the drop in glucose levels from peak to nadir estimates peripheral insulin resistance predominantly of skeletal muscle with a smaller contribution from adipose tissue (Abdul-Ghani et al, 2007). The acute response to glucose load (OGTT) in the bilateral LLA group appears to accelerate the hepatic insulin response with time to peak insulin values being recorded a mean 7.5 minutes earlier and the prolonged response (values between 60 to 120 minutes) demonstrates reduced peripheral insulin secretion. This may indicate a trend in the bilateral amputation group demonstrating a greater ability to regulate metabolism in response to glucose loading over the 20 weeks.

7.4.6. Strengths and Limitations

A potential limitation of this study is the within group variations in the severity of lower-limb injures in an all-male military participant cohort. However, this heterogeneity of injury may be considered beneficial as the range of functional abilities improves the external validity of the findings, making them more suitable for the wider UK military LLA population. The participants recruited into this observational study present similar clinical outcomes, years of age and length of rehabilitation to those reported in chapter 4. They therefore provide a useful representative sample of the wider UK military LLA population who attended DMRC, Headley Court at the end of the rehabilitation pathway. To my knowledge this is the first study to collectively assess the clinical outcomes, body composition and cardiometabolic risks associated with traumatic LLA in military personnel injured during the time of the Iraq and Afghanistan conflicts. It provides insight into the health and wellbeing of a severely injured population. Very few studies have been completed in individuals with traumatic LLA during rehabilitation and none using UK military personnel.

An unavoidable limitation of the study design was the inability to standardise the length of time since injury or the amount of rehabilitation exposure prior to participating in the trial. For example, it would have been very insightful to investigate the changes in function, body composition, PA and cardiometabolic health within a known period of time (i.e., the first 6 to 12 months of rehabilitation following LLA). This was not possible due to the UK military withdrawal from Afghanistan in 2014 and data collection for chapters 6 and 7 starting in the autumn of 2014. My way around this inconsistency was to work closely with the clinical team and identify eligible patients who had 3 admissions remaining prior to their discharge from DMRC, Headley Court. This provided an opportunity for us to comment on the health and well-being of severely injured military personnel at the end of the DMRC complex trauma rehabilitation care pathway. In the context of measuring the impact of rehabilitation following traumatic LLA on objective markers of health, the current study is one of the largest, however the sample size of individuals with amputation remains relatively small (n = 16). Despite the small sample size, significant differences were demonstrated between groups and moderate effect size changes within groups. The inability to detect more significant effects or differences is likely a reflection of the small sample size and large variability in responses.

The importance of energy intake on energy balance and its role in body composition and cardiometabolic health is recognised and the lack of energy intake data is a limitation of this thesis. We therefore recognise the lack of energy intake data as a limitation in this thesis. Efforts were made at the start of the study to capture food intake via weighing scales and a food diary. However, there was very low acceptance and adherence from the participants to engage and accurately track food intake during the study. At DMRC, weighing of food in the canteen meant other patients had to cue and wait longer for their food leading to frustration and unwanted attention, sometimes leading to embarrassment to the participant. This unwanted attention in the canteen also led to some eligible patients refusing to volunteer to take part in the trial as they saw the weighing of food an unnecessary/unwanted burden. Whilst at home, patients would often forget to engage in the process of recording a food diary/weighing their food, stating this was not compatible with their lifestyle. In an effort to encourage participant recruitment and adherence to the trial, a decision was taken 6 months into data collection to exclude energy intake from the analyses.

7.4.7. Clinical Implications

One important message to policy makers highlighted by this thesis is clinically significant improvements in function are still demonstrated 18 months to three years following injury. These participants (especially those with bilateral LLA) are among the most severely injured adults studied in the available literature. Significantly less traumatic injuries are commonly treated in civilian sectors yet the functional targets for the final stage of treatment is also significantly less (see chapter 3, section 3.6). For example, civilians with unilateral amputation should be able to stand and sit from a chair and mobilise with aids or adaptations sufficiently around their home, many military personnel with unilateral amputation expect and are capable of running independently and demonstrating PA, body composition and cardiometabolic health status that closer resembles their pre-injured self and are comparable to active age-matched controls. Most individuals with bilateral amputations in civilian settings will be dependent on a wheelchair to mobilise; the majority of UK military personnel with bilateral LLA expect and are capable of walking and performing ADL independently with an aid or adaptation. My findings therefore have significant implications for the life-long cardiovascular, musculoskeletal, mental health and vocational benefits of rehabilitation. The favourable functional responses to rehabilitation is likely to have major economic benefits both for the individual, and for the nation as a whole, as severely wounded individuals are rehabilitated back into independent, actively contributing members of society, generating tax revenue through meaningful employment. The outcomes presented here are also highly relevant to the UK civilian population. To quote Dr John Etherington, President of the Faculty of Sport and Exercise Medicine UK, former Director for Defence Rehabilitation at DMRC Headley Court and National Clinical Director for Rehabilitation for NHS England:

> *"The lack of investment in trauma-based rehabilitation, and the dis-investment in a transformational rehabilitation programmes in the NHS has severe implications for the economy of the country and the health and wellbeing of individual patients. This is particularly important at the time of increased terrorist threat, when trauma services and their rehabilitation counterparts may be significantly challenged."*

7.5. CONCLUSION

To my knowledge this is the first time individuals with traumatic unilateral LLA have demonstrated mean body composition, PA and cardiometabolic health outcomes comparable to active age-matched controls. Individuals with bilateral LLA injured during the Iraq and Afghanistan conflicts are among the most severely injured adults living, yet they present with physical function and psychosocial status that is fully compatible with integration back into society. While these findings are reassuring for the patient, their family members and clinicians, the bilateral amputations group did demonstrate unfavourable body composition accompanied by an increased risk of cardiovascular disease and compromised metabolic health. When these findings are considered alongside reduced ambulatory PAEE demonstrated in chapter 6, future efforts to promote individualised home-based exercise/PA interventions in UK military personnel with bilateral LLA should become a priority if more favourable changes in body composition and cardiometabolic health are to be realised.

CHAPTER 8. GENERAL DISCUSSION

8.1. OVERVIEW

The first retrospective chapter of this thesis (**Chapter 4**) assessed the functional and mental health status of UK military personnel with traumatic LLA at the end of their rehabilitation pathway. Specifically, humans had not previously survived the types of blast and blunt trauma injuries sustained in the conflicts of Iraq and Afghanistan. It was unknown what impact these life-altering injures had on their quality of life, level of independence and their ability to demonstrate levels of physical function that would facilitate integration back into society. This study sought to demonstrate the impact of in-patient IDT rehabilitation on UK servicemen with unilateral, bilateral and triple amputations

The first of three experimental chapters of this thesis (**Chapter 5**) was designed to assess the accuracy of a tri-axial accelerometer and multi-sensor methods in the prediction of PAEE in individuals with unilateral and bilateral LLA. This study sought to identify the most appropriate anatomical location to wear the GT3X+ device around the pelvis, in order to minimise measurement error in the prediction of PAEE in individuals with LLA. The study also aimed to determine the validity of using multi-sensor methods, which incorporate HR and acceleration signals (AHR versus GT3X+ and Polar HR), to estimate ambulatory PAEE in military personnel with traumatic LLA. In essence a comparison between a research-grade device with PAEE algorithms intrinsic to the device (AHR) compared to a population specific multi-sensor algorithm (GT3X+HR). The performance of these devices was assessed across a wide range of walking velocities and gradients on a treadmill in a controlled laboratory setting. PAEE prediction models were developed using corresponding criterion data (from indirect calorimetry) and outputs from each device from each walking intensity/gradient, using linear regression analysis. Error statistics were then determined using a leave-one-out cross validation analysis.

The second and third experimental chapters (**Chapters 6 and 7** of this thesis) were part of a longitudinal observational study monitoring a group of unilateral and bilateral amputees over the duration of two 4-week in-patient rehabilitation admissions at DMRC Headley Court and two extended periods of active recovery at home. A control group of physically active, age-matched, non-injured, males were recruited as a normative dataset. This control group provided the opportunity to comment on the impact that traumatic LLA has on previously physically active military personnel and enable comparisons to their peers. See figure 6.1 for a schematic of the study design but in brief, **Chapter 6** also aimed to assess and compare estimated daily PAC and PAEE during structured in-patient rehabilitation at DMRC compared to free-living habitual activity at home. Daily PAEE (kcal.day^{-1}) was estimated using daily PAC derived from a GT3X+ worn on the hip of the shortest residual limb (from **chapter 5**). Alongside these PA measures, this chapter also determined the functional status of each group at baseline using a range of clinical outcome measures routinely collected at DMRC. Using the same participants as **chapter 6**, **chapter 7** aimed to assess the impact of two in-patient admissions and two blocks of active recovery at home on clinical outcomes, body composition and cardiometabolic health. Upon commencing the start of a new 4-week in-patient admission, following a DEXA scan, fasted baseline blood samples were taken and an OGTT performed. Routine clinical outcomes and a DEXA scan were performed at the mid-point (10 weeks) at the start of their second admission. Upon returning to DMRC after their second period of active recovery at home (20 weeks), clinical outcomes were performed again alongside fasted blood samples and an OGTT.

8.2. SUMMARY OF KEY FINDINGS

8.2.1. Chapter 4: Functional and Mental Health Status of UK Military Post-Rehabilitation

- The bilateral LLA and triple amputee participants described in this study are among the most severely injured humans (based on NISS) reported in the available literature.
- At their last admission to DMRC, military personnel with unilateral LLA demonstrated levels of ambulatory function considered superior to the general population. Military personnel with bilateral LLA demonstrated a level of function comparable to age-matched general population.
- UK military personnel with LLA demonstrate none/minimal levels of depression and anxiety at the end of their rehabilitation pathway indicative of preparedness for full integration back into society.

8.2.2. Chapter 5: Estimating Ambulatory Energy Expenditure in UK military Personnel with Traumatic Lower-Limb Amputation

- Of the three anatomical locations around the pelvis considered, PAC signals from the GT3X+ worn on the shortest residual limb provide the strongest association, smallest LoA and least error in the prediction of PAEE in individuals with LLA during an incremental walking protocol in laboratory conditions.
- Across all walking speeds, PAEE was 1.4 to 1.7 and HR was 1.3 to 1.5 times greater in military personnel with unilateral LLA compared to normative controls.

- At all gradients and treadmill speeds ≤ 0.89 $m.s^{-1}$, PAEE was 2 to 2.6 and HR was 1.5 to 1.7 times greater in military personnel with bilateral LLA compared to normative controls.
- Including length of rehabilitation (time since amputation) and level of amputation for unilateral LLA and waist circumference for the bilateral LLA prediction models provided a small but significant improvement in variance for PAEE.
- The AHR multi-sensor device, which uses proprietary group calibrations intrinsic to the device, explains less of the variance and produced greater error in the estimation of PAEE than HR alone.
- The inclusion of a physiological variable (HR) with acceleration signals (GT3X+HR) created a more accurate population-specific predictive model for determining PAEE than outputs from the GT3X+, AHR or HR used in isolation.
- The GT3X+HR developed prediction models explained 85%, 87% and 83% of the variance in PAEE for the unilateral, bilateral and control group, respectively.

8.2.3. Chapter 6: Influence of Lower-Limb Amputation Severity and Recovery Environments on Indices of Physical Activity and Functional Recovery in UK Military Personnel

- Baseline 6-MWD and AMPQ values in the bilateral LLA group were comparable to civilians with unilateral LLA due to peripheral vascular disease and comparable to US military personnel with bilateral LLA.
- Baseline 6-MWD and AMPQ values in the unilateral LLA group were comparable to the general population and already indicative of a functional status necessary for community integration. However, 6-MWD values were significantly less than active normative controls.
- Mean scores for depression and anxiety indicated ‘none/minimal’ symptoms for both LLA groups.
- During in-patient rehabilitation, bilateral LLA demonstrated significantly reduced daily PAEE compared to unilateral LLA.
- The daily PAEE and PAC demonstrated by military personnel with unilateral LLA during rehabilitation were comparable to active normative controls during work.
- Whilst at home, both LLA groups demonstrated significantly reduced PAC and PAEE compared to active normative controls during work. Bilateral LLA demonstrated 47% reduced PAC and 70% less estimated PAEE than unilateral LLA in this environment.
- Both amputation groups recorded greater amounts of daily PAC and PAEE during in-patient rehabilitation compared to habitual living at home.

8.2.4. Chapter 7: Baseline differences between groups for Body Composition and Cardiometabolic Health in military personnel with traumatic LLA

- Despite similar total body mass, military personnel with bilateral LLA demonstrate significantly greater fat mass, percentage fat mass, android fat mass, gynoid fat mass, VAT area, waist circumference, waist-to-hip ratio and reduced lean mass compared to active normative controls.
- Bilateral LLA demonstrated significantly greater waist circumference and VAT area compared to unilateral LLA.
- No significant differences in body composition or cardiometabolic health outcomes were demonstrated between unilateral LLA and normative controls.
- The bilateral LLA group demonstrated significantly increased TC:HDL ratio, triacylglycerol, CRP, time to peak glucose, insulin AUC, peak insulin, higher fasting estimates of β-cell function and lower insulin sensitivity compared to active normative controls.
- Bilateral LLA group demonstrated significantly greater TC:HDL, triacylglycerol, NEFA, CRP, and insulin AUC compared to the unilateral LLA group
- Metabolic Syndrome was present in 63% of military personnel with bilateral LLA, but not present in any individuals with unilateral LLA.
- Bilateral LLA are at an increased risk of obesity, cardiovascular disease and compromised metabolic health.

8.2.5 Chapter 7: Interaction effects and pre to post differences within groups for function, body composition and cardiometabolic health during Rehabilitation from Traumatic LLA

- Over the 20 week monitoring period unilateral LLA improved their 6MWD by 24 metres and bilateral LLA by 54 metres. Neither group demonstrated statistically significant improvements in distances walked. However, by the end of the study the bilateral LLA group did achieve minimal clinically significant difference (>45 metres) and the majority (88%) were capable of walking independently without an aid or adaption.
- The prevalence of unilateral LLA able to run independently over the study duration increased from 13% to 63% and the ability to perform ADL also increased from 13% to 75%.
- No significant time x group interaction effects were reported in any cardiometabolic health measure. And body composition measures remained stable across all groups

8.3. GENERAL DISCUSSION POINTS

This section provides an opportunity to collectively summarise the general findings from each observational/experimental chapter. For a detailed discussion and interpretation of the findings, the reader is encouraged to read each individual chapter of the thesis.

8.3.1. Rehabilitation of UK military personnel with unilateral and bilateral LLA

Clinical outcomes data (presented in **chapters 4, 6 and 7**) from military personnel with traumatic LLA, recorded towards the end of the DMS combat casualty care pathway (**chapter 3**), demonstrate a level of functional performance and psychosocial health indicative of full preparedness for community integration. Specifically, the unilateral LLA group demonstrate a 6-MWD comparable to general population norms (Chetta et al, 2006), a prosthetic ambulatory ability that can exhibit high impact, stress and energy levels typical of the prosthetic demands of an active adult or athlete (Gailey et al, 2002). All operationally injured unilateral LLA reported on within this thesis are able to walk independently without an aid or adaption and many are capable of running. This is quite a contrast to the end stage of rehabilitation care typically demonstrated within civilian based rehabilitation settings, where the aspiration for individuals with unilateral LLA is to ensure they have the ability to sit and stand from a chair and mobilise with aids or adaptations around their home (section 3.6). These superior outcomes demonstrated in UK military personnel are also reflected by their PA capacity, body composition and cardiometabolic health status being comparable to active normative controls (discussed in more detail in the next couple of sections).

Consistent with previous literature (Doukas et al, 2013), UK military personnel with unilateral LLA demonstrate levels of function superior to individuals with traumatic bilateral LLA. However, UK military personnel with traumatic bilateral LLA can walk comparable distances in six minutes to US military personnel with bilateral LLA and comparable distances to civilians with unilateral amputation due to peripheral vascular disease (Gailey et al, 2012). At the end of the 20 week longitudinal study (**chapter 7**) all could perform ADL independently with an aid or adaptation. Considering the severity of injures typical of blast-based bilateral amputation (see chapter 3), and the likely functional status of civilians with bilateral amputation almost certainly being dependent upon a wheelchair to mobilise (section 3.6) it can be argued that this group of participants present with a high level of physical functioning. One important message to policy makers highlighted by this thesis is that clinically significant improvements in function are still demonstrated 18 months to three years following injury. The functional outcomes presented in both amputation groups nicely captures what can be achieved when health care organisations invest in rehabilitation services for people with complex traumatic injuries such as LLA.

Symptoms of depression and anxiety are frequently reported in individuals with traumatic LLA (Caddick et al, 2019; Epstein et al, 2010; Foote et al, 2015). However, data from this thesis consistently demonstrate that UK military personnel with unilateral and bilateral LLA who are nearing the end of the DMS complex trauma rehabilitation care pathway indicate '*none or minimal*' symptoms. As discussed in section 2.3.8, an intense emotional response is common following traumatic LLA and forms part of the psychological adjustment to injury. Due to the cross-sectional nature and timing of data collection, it is not possible to comment on any fluctuations in psychosocial health that may have occurred during the first year following injury. However, these favourable psychosocial health outcomes when combined with the functional performances indicated in this thesis should help facilitate full integration back into society. Unfortunately, it was beyond the scope of this thesis to confirm whether these clinical outcomes were maintained long-term following discharge from the military.

8.3.2. Validating and using population specific models of estimating PAEE in unilateral and bilateral LLA

Attempts to accurately predict PAEE in individuals with traumatic LLA had not been performed prior to the two publications in PLoS One (Ladlow et al, 2017; Ladlow et al, 2019), which stem from chapter 5 of this thesis. PAEE has proven inherently difficult to measure, even in humans without mobility related physical impairments. The challenges are heightened with heterogeneous groups of individuals where the level of amputation results in varying loss of muscle mass and sensory/motor function of the lower limb (Czerniecki & Morgenroth, 2015). Of the three anatomical locations considered in chapter 5, wearing the Actigraph GT3X+ accelerometer on the side of the shortest residual limb provides the most accurate prediction on PAEE in LLA. The manually derived model integrating additional covariates including physiological variables (HR) with triaxial acceleration data has been shown to provide greater validity for estimating ambulatory-related PAEE in individuals with traumatic LLA. The newly developed population specific algorithms substantially outperformed the proprietary group algorithm intrinsic to the research grade multi-sensor device (AHR). Due to the poorer predictive validity of the AHR device, I recommend using the GT3X+ together with the algorithms presented in this article, which demonstrate a lower estimated error.

The original preference was to use the GT3X+HR prediction models developed in experiment two (**chapter 5**) to estimate PAEE during rehabilitation and whilst at home. Unfortunately, this was not possible due to the significant problems encountered with participants wearing ECG electrodes that have previously been explained. I therefore, used the GT3X+ predictive model validated in experiment one of **chapter 5** to estimate daily PAC and PAEE during waking hours in two environments (in-patient rehabilitation and whilst at home) in **chapter 6**. The newly developed, population-specific prediction models revealed that military personnel with unilateral LLA have the 'capacity' to perform similar amounts of PAEE to active normative controls. A finding not previously demonstrated in the available literature. A significant difference in PAEE between environments was demonstrated for both LLA groups, but was particularly greater in individuals with bilateral amputations. Although it is perhaps unsurprising that PA levels of individuals with bilateral amputations are lower than in those with a unilateral amputation or normative controls, the gap that exists between these populations is significant, particularly as all individuals with LLA who participated in this study had access to the most technologically advanced prostheses available and prolonged intensive rehabilitation care. Where large differences occur in activity levels between in-patient rehabilitation and at home, it is possible that a home-based intervention is likely to lead to substantial improvements in long-term QoL and functional status of UK military personnel with LLA (study design proposed in section 8.5). The findings suggest that improvements in technology and clinical care are still needed to restore the PA and functional status of individuals with bilateral LLA. By promoting longer-term behaviour change and allowing clinicians to monitor participant compliance the use of objective and validated PA assessment may better inform treatment interventions in the future.

8.3.3. The impact of injury severity on body composition and components of cardiometabolic health in unilateral and bilateral LLA

Blast related trauma (i.e., IEDs) can result in catastrophic injuries to the entire body (as described in **chapter 3**) resulting in extended bed rest in a hospital environment which leads to significant reductions in physical function to both the upper and lower extremities. This is followed by prolonged amounts of time with a reduced level of PA. During these early months of rehabilitation, many UK military personnel rely on wheelchairs to mobilise, require aids/adaptations to perform ADL and in some cases require a full or part-time carer during this early phase of treatment. This prolonged period of reduced capacity to engage in PA or conventional exercise rehabilitation is likely to have a profound impact on body composition earlier in the rehabilitation care pathway. Progress with physical fitness is only likely to occur after sufficient wound healing from multiple revision surgeries and socket fittings. Based on the available evidence, individuals with traumatic LLA (particularly bilateral LLA) are predisposed to considerable muscle atrophy, strength deficits and increases in fat mass early in their rehabilitation care pathway (**chapter 2**, section 2.6.2). The unfavourable body composition demonstrated by the bilateral LLA group is likely a reflection of their earlier journey as a patient recovering from their injuries and the challenges to engage in higher volumes and intensities of PA and exercise.

To my knowledge this is the first time individuals with traumatic unilateral LLA have demonstrated mean body composition and cardiometabolic health outcomes comparable to active age-matched controls. One explanation previously described is this groups capacity to engage in daily PA at comparable levels to active normative controls. The unfavourable body composition (particularly the distribution of adipose tissue around the abdominal area) demonstrated by the bilateral LLA group was also accompanied by an increased risk of cardiovascular disease and compromised metabolic health, with 63% of participants being classified with metabolic syndrome.

Rehabilitation and exercise prescription within DMS are not typically based on interventions to specifically target improvements in body composition or cardiometabolic health and these measurements do not form part of routine clinical practice at DMRC Headley Court. Clinicians were not privy to any of the PA, body composition or cardiometabolic health data collected about their patients during the data collection period. Had they been, this may have altered aspects of exercise prescription and delivery. This study clearly demonstrates an increased cardiometabolic risk associated with the most severely injured military personnel (i.e. bilateral LLA). It also provides a justification for greater health/PA education and use of holistic monitoring procedures (for example, the use of cardiometabolic biomarkers, body composition and habitual PA monitoring) in patients most at risk, to better inform clinical decision making and practice. A recent article reported that globally physiotherapists lack knowledge on PA guidelines with the main barriers to them promoting PA being lack of time, lack of skills and counselling for behaviour change, and a belief that promoting PA would not change patient behaviour (Yona et al, 2019). Previously, it has been reported that only 16% of physiotherapist (n = 514) correctly answered questions about the content of PA guidelines (Lowe et al, 2017). I did not assess DMRC clinician's knowledge on PA guidelines or their understanding of how to promote or encourage PA. However, it is clearly evident that PA promotion is an important component of any rehabilitation pathway and essential for the health of all UK military personnel. Therefore, health care professionals like physiotherapists and ERIs are ideally placed to provide LLA patients with the skills to overcome barriers to engage in health promoting behaviour at home and lead PA interventions on behalf of DMS. Such practices may also provide the patient with greater accountability / awareness of the impact of negative lifestyle behaviours (i.e. low PAEE) whilst recovering at home or back in civilian life. Proposed future research investigating the use of home-based exercise/PA intervention strategies to elicit long-term favourable cardiometabolic health outcomes, particularly in individuals with bilateral LLA are described in section 8.5.

8.4. CONSIDERATIONS

Specific considerations for each study have been discussed within the individual chapters. However, for the benefit of this general discussion the limitations for each chapter will be briefly described.

Whilst outside my control, perhaps the most consistent limitation of every chapter was the timing of data collection and therefore the recruitment pool of eligible participants. Data collection for the experimental chapters commenced when the overwhelming majority of military personnel with LLA had already been discharged from DMRC Headley Court. Whilst there was still a sizeable number of LLA still receiving rehabilitation care when data collection started, the majority of these patients only had one in-patient admissions remaining, and therefore were not eligible for the longitudinal study (chapters 6 and 7). Consequently the sample size for each experimental chapter was smaller than originally planned. The timing of data collection did however provide a useful consistency throughout the thesis, as all participants recruited were at the end of their rehabilitation pathway, providing a unique opportunity to comment of the impact of the DMS rehabilitation care pathway on this unique population.

8.4.1. Limitations of reporting the functional and mental health outcomes of UK military personnel with LLA at their last admission to DMRC (chapter 4)

While this study (published in Archives of Physical Medicine and Rehabilitation) was the first published account of the functional and mental health outcomes of UK military personnel with LLA from the Iraq / Afghanistan conflict, it did have some limitations. The retrospective cross-sectional nature of the study design does not reflect the inevitable fluctuations that occur in clinical outcomes throughout a prolonged period of rehabilitation. The length of rehabilitation was reported, however, an association between the reported clinical outcomes and the content of the rehabilitation program to provide evidence of a causal link is lacking due to the cross-sectional study design. The primary reason for not reporting this journey of rehabilitation is because the electronic storage of medical records using Defence Medical Information Compatibility Program commenced in 2012 and by this time the majority of LLA from the conflicts in Iraq / Afghanistan had already occurred making retrieval of earlier clinical outcome measures not possible. The only consistent reporting time period that outcomes could be extracted was the final in-patient admission at DMRC Headley Court (i.e., patients discharged from 2012 onwards). This reduced the available sample size for analysis and restricted my ability to perform a retrospective analysis of clinical outcomes performed at each admission since their injury / first in-patient admission to DMRC.

8.4.2. Developing validated methods to estimate ambulatory energy expenditure in military personnel with unilateral and bilateral LLA

Despite the small sample size, the inclusion of a diverse range of participants is in accordance with best practice recommendations for PA validation studies (Bassett et al, 2012). The requirements for large and representative samples present unique challenges, focusing around cost or time involvement, and this is even more problematic when considering the difficulties associated with recruiting from various disabled populations. It remains to be seen whether specific predictive algorithms developed using military personnel with traumatic LLA, will offer improvements in the prediction of PAEE for the wider civilian population. In order to achieve this, I encourage research groups to work together to foster progress in the development of objective monitoring tools to be used in this population. The ability to capture raw acceleration data now permits more sophisticated methods of predicting PAEE and allows researchers to detect the types of activity a person is performing (Bonomi et al, 2009a). This might be highly relevant to populations that perform atypical movement patterns, like individuals with traumatic bilateral LLA. It is possible that predicting PAEE from linear regression equations, as in Chapter 5, may be too simple an approach to examine complex movements or behaviours (Strath et al, 2012). Whilst linear regression models performed well enough to quantify PAEE across various treadmill speeds and gradients, it cannot be concluded whether this is the case for other activities, such as walking over ground, up and down stairs or resistance training protocols that use multiple limbs. Diverse ranges of ADL were not included in the validation protocols. It is advised that the shortest possible epoch (1 second) should be selected for activity monitor data collection, to ensure that as much information as possible regarding the original PA related biosignal is retained (Heil et al, 2012). This then permits the use of new data analysis methodologies, including hidden Markov models (Pober et al, 2006), artificial neural networks (Staudenmayer et al, 2009; Trost et al, 2012) and classification trees (Bonomi et al, 2009b), which use the rich information to classify certain activities and derive a more accurate estimate of PAEE (Bassett et al, 2012). Future research should investigate these techniques across diverse activity protocols, which may include light-intensity ADLs and high intensity multi-joint exercise rehabilitation regimes.

8.4.3. Indices of Physical Activity and function during rehabilitation from traumatic LLA

The original plan for this chapter was to use the most accurate PA prediction method validated in chapter 5. This incorporated the use of acceleration signals from the GT3X+ and HR signals. These HR signals were originally obtained from the AHR PA monitor. Due to unforeseen problems with skin irritation with the electrodes (requiring nurse intervention) a decision was made by senior clinical staff prohibiting the use of the AHR device in complex trauma patients, thereby excluding HR as a physiological variable to improve the accuracy of the prediction equations to estimate

PAEE in the 20 week longitudinal study. Therefore, only the use of the Actigraph GT3X+ was possible. Fortunately, we had already validated the use of this method for estimating ambulatory EE (chapter 5: experiment one) and this was used to quantify PAC and thus estimate PAEE during in-patient rehabilitation and whilst at home. Unfortunately, there are known limitations to using accelerometers in isolation that could not be avoided. Accelerometers are unable to register isometric activities, muscular work against an external force, such as lifting or resistance training (Butte et al, 2012). As resistance training is an integral component of any rehabilitation program and actively encouraged whilst at home, it is possible that PA levels were underestimated in this study population. It is unknown how well predictive algorithms developed in a controlled laboratory on a treadmill translate to free-living ambulatory EE in other locations (i.e. during rehabilitation sessions or habitual activity at home). Another important consideration detailed in section 6.4.4 was the anecdotal reports from some participants during the study that they reduce the amount of time they wear their prosthesis whilst at home, instead favouring a wheelchair to mobilise. The anatomical location of the hip is not the preferred anatomical location to detect activity in wheelchair users (Nightingale et al, 2014) and this may have underestimated PA levels whilst at home, perhaps explaining the more pronounced reduction in PAC and PAEE observed in this environment in the bilateral LLA group. Nevertheless, these data still suggest a difference in PA behaviour in this specific population and highlight that ambulation is likely reduced in the home environment in the bilateral LLA.

8.4.4. Changes in function, body composition and cardiometabolic health during rehabilitation from traumatic LLA

This is the first study to collectively assess the clinical outcomes, body composition and component risk factors for cardiometabolic disease in military personnel with traumatic LLA injured during Iraq / Afghanistan. It provides insight into the health and well-being of some of the most severely injured adult populations alive today. As mentioned earlier the timing of data collection meant that all participants were towards the end of their rehabilitation care pathway. Clinically this can often indicate participants starting to reach a plateau in their clinical status (i.e. functional performance and psychosocial health). This is likely to be one of the leading contributing factors for the lack of change over time (20 weeks) recorded in body composition and cardiometabolic health within the two LLA populations. However, the findings of this chapter clearly show differences between groups and it is likely that the increased severity of injuries sustained by the bilateral LLA group had a large role to play in these differences. Had I been able to start data collection during earlier stages of rehabilitation care, I may have been able to detect changes in body composition and cardiometabolic health within unilateral and bilateral LLA groups over time.

Attempts to monitor energy intake through methods of weighed food diaries were terminated approximately 6 months into data collection due to low compliance and the impact this had on recruitment into the trial (see section 7.4.6). I recognise the importance of energy intake on energy balance and the knock-on consequences on body composition and cardiometabolic health. The inability to control for energy intake is considered a weakness of the chapter. However, weighing and rigorously recording of food intake is notorious for under-reporting and the accuracy / usability of these types of data are deemed questionable by the wider scientific community (Shim et al, 2014). The challenges associated with accurately recording energy intake are therefore not unique to this study. Until more sophisticated and less burdensome methods to capture energy intake are developed, researchers are always going to experience this dilemma.

8.5. RECOMMENDATIONS FOR FUTURE RESEARCH

As the UK is no longer engaged in military conflicts, like the ones witnessed in Iraq and Afghanistan, the number of traumatic LLA has decreased considerably. Only a few patients with LLA currently receive rehabilitation at DMRC, and the majority who do are elective unilateral LLA with very different rehabilitation needs compared to the participants reported in this thesis. DMRC do not typically provide in-patient rehabilitation to veterans, making follow-up rehabilitation-based studies similar to the one presented in this thesis challenging. However, a 20 year follow-up study called The Armed Services Trauma Rehabilitation Outcome (ADVANCE) Study is being delivered at DMRC Stanford Hall (formerly DMRC Headley Court) and could provide a suitable cohort of participants to perform relevant follow-up research. The ADVANCE Study is investigating the long-term physical and psychosocial outcomes of battlefield casualties from the UK Armed Forces following deployment to Iraq or Afghanistan between 2003 and 2014. These participants are now veterans and receive their healthcare needs from the civilian sector (i.e., the NHS), but return to DMRC ever 3 to 5 years for follow-up assessments. Among the many outcome measures used as part of the ADVANCE study, the fasting cardiometabolic health measures, DEXA-based body composition and functional outcomes (6-MWD) are identical to those measured in this thesis. However, the ADVANCE study does not collect objective PA outcomes.

One of the main findings from this thesis was a high prevalence (63%) of UK military personnel with traumatic bilateral LLA being classified with metabolic syndrome, despite a relatively short duration since injury (39 ± 15 months) and continued provision for ongoing IDT rehabilitation. When I refer back to the literature review (section 2.7.3), uninjured adults with metabolic syndrome demonstrate a 2-fold increase in developing cardiovascular disease over the following 5 to 10 years (Alberti et al, 2009). Consequently, individuals with metabolic syndrome have increase mortality and a shortened lifespan (Guize et al, 2007). One the most important observations made within this thesis is the significantly reduced PAEE demonstrated at home in the bilateral LLA group. Evidence based exercise / PA interventions that can be successfully integrated and performed in a home environment (away from DMRC) are crucial if the promotion of long-term health and well-being of the most severely injured UK military personnel are to be realised.

Remotely based telemedicine / telehealth interventions have been successfully delivered to promote health outcomes and QoL in range of clinical populations such as spinal cord injury (Irgens et al, 2018) type 2 diabetes mellitus (Lee et al, 2018) heart failure, asthma, COPD and cancer (Hanlon et al, 2017). Telehealth interventions are considered particularly useful for individuals who live in rural areas, have limited mobility, time constraints due family/life demands, are socially isolated, or fear meeting new people (Banbury et al, 2018). It is currently unknown whether this type of exercise / PA intervention would be successful in UK veterans with traumatic bilateral LLA. However, among the reasons why these veterans might benefit from the use of telemedicine-based services (if delivered by DMRC) is due to the lack of specialists outside of DMS who are qualified to care for individuals who have experience such high severities of traumatic injury. Should a telehealth home-based exercise intervention be delivered by experienced clinicians (i.e., physiotherapists and ERIs) from within complex trauma at DMRC Stanford Hall, the potential for optimising longer-term health outcomes could be realised. Also, the retention of clinical knowledge and expertise in delivering rehabilitation care to this unique population by DMRC clinicians is considered vitally important for DMS. Should a similar conflict to the one realised in Afghanistan occur, where IEDs were the preferred weapon of choice (leading to devastating injuries like traumatic bilateral LLA), this unique clinical expertise will be urgently required. Providing clinicians with the opportunity to provide remote access care (via video conference calls) of previous patients highlighted as being most at risk of developing unfavourable long-term health conditions would be welcomed, especially as this group of patients are now rarely seen or treated at DMRC. I therefore propose the following research trial as a logical follow-up study to data collected and presented as part of this thesis.

Research Question: Can a 12 month telehealth home-based exercise intervention reduced the component risk factors for metabolic syndrome and increase long-term physical function in military personnel with bilateral LLA?

See figure 8.1 for a schematic of the proposed study design. Following an ADVANCE Study clinic, military personnel with bilateral LLA who have been classified with having metabolic syndrome (based on the criteria set by the International Diabetes Foundation: Zimmet et al, (2005)) will be invited to participate in a RCT. One group will receive a telehealth home-based exercise intervention; the other will not receive an intervention. Both groups will be provided with an Actigraph GT3X+ accelerometer to wear on the hip of their shortest residual limb, asked to wear it during waking hours for 7 days and return the accelerometer back to DMRC in the post. The telehealth intervention will consist of multiple video conference calls between an experienced complex trauma physiotherapist or ERI based at DMRC and the veteran with bilateral LLA. The intervention group will receive a 12 month home-based progressive exercise program unique to their movement capabilities and their home living environment. For the first 6 months, participants will receive one 60 minute video conference call, once a month. For the remaining 6 months this will decrease to one video call every 2 months, resulting in a total of 9 video calls over the 12 month duration. The aim of each conversation is for the therapist to prescribe and promote adherence to home-based exercises and for both parties to discuss ways to overcome any potential barriers to PA engagement that the veteran may have, thereby hopefully facilitating greater independence around their home / local community and reduce the component risk factors of metabolic syndrome. After 12 months all participants will return to DMRC Stanford Hall for a follow-up where they will be asked to provide a fasted blood sample for components of cardiometabolic health, have their blood pressure taken, a whole body DEXA scan along with a waist circumference measure and asked to perform a 6-MWT functional assessment. They will also be provided with a GT3X+ accelerometer for a second 7 day monitoring period. An anticipated outcome is that the telehealth intervention group will perform more favourably on each outcome measure compared to the group who did not receive any intervention. The real test of the telehealth intervention however will be whether this remote personalised approach to rehabilitation care provides lasting health benefits. Therefore, when individuals return to DMRC Stanford Hall for their planned 3 year ADVANCE Study follow-up, the outcomes measures used at 12 months will be extracted from the vast amounts of data recorded during these ADVANCE study clinics and differences between and within groups can be analysed. If successful, the benefits of this proposed RCT will provide DMS with the provision for an evidence-based intervention to support the long-term health and wellbeing of the UK militaries most severely injured veterans, a provision not currently provided to military veterans with LLA.

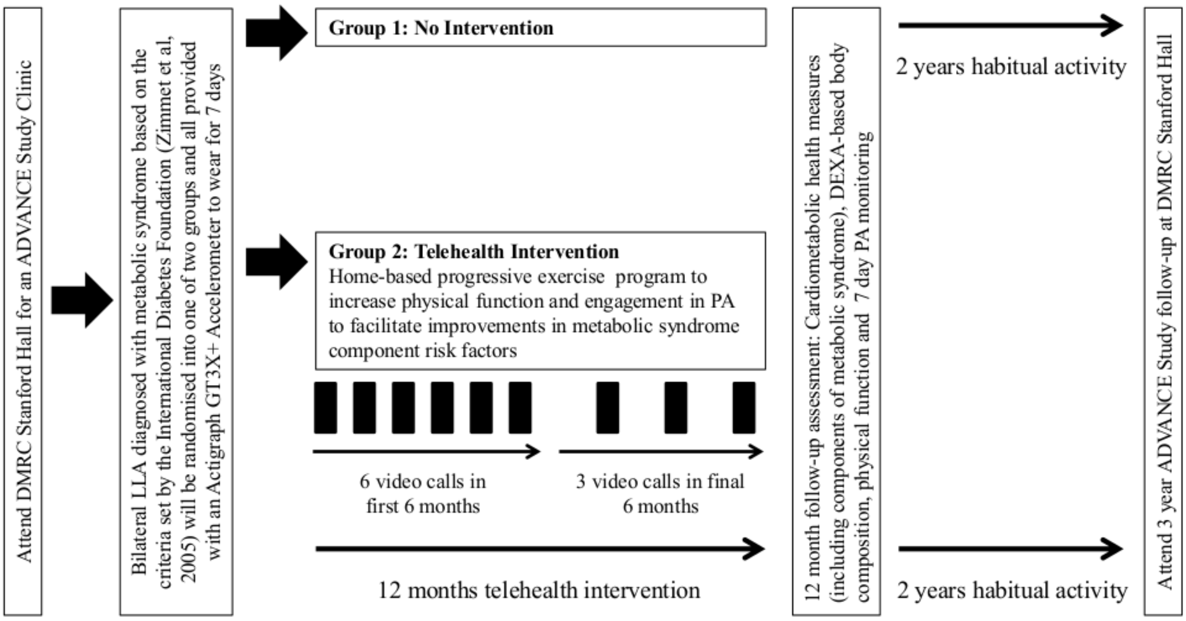


Figure 8.1: Schematic of the study design for a telehealth RCT for bilateral LLA with metabolic syndrome

8.6. IMPACT OF RESEARCH

8.6.1. National and International Impact

The outcomes presented in **Chapter 4** have been presented and discussed at a NATO summit. The Surgeon General, Medical Director and two Commanding Officers at DMRC have used this data when discussing the impact of rehabilitation care provided by DMS to international partners and government officials. The former National Clinical Director for Rehabilitation in NHS England has used this data to support greater investment into rehabilitation services across NHS England. The National Rehabilitation Centre (a facility proposed to be built on the same estate as DMRC Stanford Hall) will be specialising in neurological and complex trauma rehabilitation. One of the primary objectives of this National site is for the NHS to share and learn from the experiences of the UK military. Over the past few years I have presented the findings from Chapter 4 to regional/national NHS managers, executives and orthopaedic surgeons to help support the building of this facility as the data presented throughout the thesis showcases what can be achieved when organisations invest in rehabilitation services.

8.6.2. Local Impact: Changes in Clinical Practice

The ceiling effect of the AMPQ outcome measures demonstrated in chapter 4 facilitated a review of clinical outcome measures and actually informed clinical practice. This led to complex trauma disbanding this outcome measure in in favour of alternative functional outcome measures to record function in LLA at DMRC.

In Chapter 5, the inclusion of waist circumference was found to significantly improve the estimation of PAEE in bilateral LLA. This was not a measure routinely recorded within complex trauma, but has since been included for all complex trauma patients considered at risk of unfavourable weight gain and referred to the unit dietician. In Chapter 6 and 7 I used measurements techniques not conventionally utilised during rehabilitation within complex trauma (i.e., DEXA to measure body composition and sampling blood biomarkers to measure components of cardiometabolic health). Findings from this thesis demonstrate an increased risk of obesity and unfavourable changes in cardiometabolic health in the most severely injured military cohorts. Consequently, conversations are now on-going surrounding the use of more objective physiological assessments to guide clinical decision making where clinicians and patients could be held more accountable to changes in health following traumatic injury.

8.7. CONCLUSIONS

This thesis determined the functional and psychosocial outcomes of UK military personnel at the end of the UK DMS rehabilitation pathway at DMRC Headley Court. Followed by systematically developing and evaluating wearable PA monitoring devices to predict ambulatory EE during in-patient rehabilitation and habitual activity at home in military personnel with traumatic LLA. The findings suggest wearing an Actigraph GT3X+ triaxial accelerometer on the hip of the shortest residual limb and using a predictive model incorporating heart rate offers the most accurate prediction of PAEE in traumatic lower-limb amputees. This thesis includes the first study to estimate ambulatory EE using an objective triaxial accelerometer during in-patient rehabilitation and active recovery at home. Findings reveal UK military personnel with unilateral LLA have a similar capacity to engage in PA as active normative controls. Unilateral LLA also report comparable body composition and cardiometabolic health outcomes to active normative controls, which is to my knowledge the first time demonstrated in the available literature.

Despite demonstrating levels of function and psychosocial health indicative of integration back into society, estimated daily PAC and PAEE in bilateral LLA demonstrate large discrepancies between the rehabilitation and home environments, with mean PA considerably less than unilateral LLA and normative controls. The significant reductions in mean daily estimated ambulatory EE demonstrated whilst at home and reduced physical function (recorded during rehabilitation) are likely to be a primary mechanism leading to the unfavourable distributions of fat and components of cardiometabolic health when compared to unilateral LLA and normative controls. To support and manage the long-term health and well-being of military personnel with bilateral LLA, future research should aim to investigate strategies that promote regular engagement in PAEE and/or structured exercise whilst at their home environment.

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