BONE DENSITY AND MACROSTRUCTURE OF THE RADIUS IN PATIENTS AFTER CHRONIC STROKE: RELATIONSHIP TO MUSCLE FUNCTION AND CARDIOVASCULAR HEALTH

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A thesis submitted in partial fulfillment of the requirements for

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CERTIFICATE OF ORIGINALITY

I hereby declare that this thesis is my own work and that, to the best of my knowledge and belief, it produces no material previously published or written, nor material that has been accepted for the award of any other degree or diploma, except where due acknowledgement has been made in the text.

_____(Signed)

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ABSTRACT

Background and purpose: Secondary bone loss in the upper extremity is a common complication after stroke, which would increase the incidence of fractures, especially in the wrist region. However, the extent to which different stroke impairments are associated with bone health status in the paretic upper extremity is not well understood. The objectives of this study were to (1) assess the side-to-side difference in areal bone mineral density (aBMD) of the forearm using dual-energy X-ray absorptiometry (DXA), and densitometric and geometric parameters of the radius diaphysis and epiphysis using peripheral quantitative computed tomography (pQCT) among patients after chronic stroke, (2) compare the side-to-side difference in DXA- and pQCT-derived parameters between individuals with chronic stroke and age-matched healthy control subjects, and (3) identify the determinants of DXA-derived aBMD of the paretic forearm and pQCT-derived bone strength indices of the paretic radius.

Methods: A total of 65 chronic stroke patients, and 34 healthy individuals participated in the study. DXA was used to evaluate the aBMD of the 1/3 region, mid-region, and ultradistal region of the forearm , and pQCT was used to evaluate volumetric BMD (vBMD), bone geometry, and bone strength indices at the radius distal epiphysis and diaphysis on both sides. Each subject was also evaluated for grip strength, spasticity, motor function, and disuse of the paretic upper extremity. Indicators of cardiovascular health including the oxygen consumption rate (VO_2)

during the Six Minute Walk Test, stroke volume index (SI), cardiac output index (CI), large and small artery elasticity index (C_1 and C_2) were also measured.

Results: The results showed that in the stroke group, the DXA-derived aBMD values in different regions of the forearm on the paretic side were significantly lower than those on the non-paretic side, and that the side-to-side differences in aBMD values in these patients were greater than those of their healthy counterparts. The pQCT results also revealed significantly lower vBMD and bone strength index values in the paretic radius epiphysis and diaphysis when compared with the non-paretic side among the patients after stroke, whereas the control group had no significant side-to-side difference in the same parameters. Multiple regression analysis showed that after accounting for age, gender, post-stroke duration and body mass index, grip strength was the most important determinant of the aBMD of the various regions of the paretic forearm, and the bone strength indices of the radius epiphysis and diaphysis in the stroke group.

Conclusions: This study suggests that among the various stroke-related neuromuscular and cardiovascular impairments, muscle weakness is the most important determinant of the DXA-derived aBMD values and pQCT-derived bone strength indices. Promoting muscle strength of the paretic upper extremity may be an important treatment strategy to enhance or maintain bone mass in the paretic upper extremity, and warrants further investigations.

PUBLICATIONS

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TABLE OF CONTENTS

Certificate of originality.....ii

Abstract iii

Publications v

Acknowledgement.....vi

Table of contents.....viii

List of abbreviations..... xvi

List of tables.....xviii

List of figures.....xix

Appendices.....xx

CHAPTER 1: INTRODUCTION

1.1 Epidemiology of Stroke……...23

1.2 Falls in Stroke Patients…….24

1.2.1 Incidence and Characteristics……...24

1.2.2 Risk Factors……..26

1.3 Fractures after Stroke……27

1.3.1 Incidence and Characteristics……...27

1.3.2 Risk Factors……..28

1.4 Bone Changes after Stroke……...30

1.4.1 Changes in Upper Extremity Bone Mineral Density as Measured by DXA……...30

1.4.2 Changes in Bone Mineral Density and Geometry as Measured by pQCT……..31

1.4.3 Comparison with Lower Extremity Bone Sites…….33

1.5 Factors Related to Upper Extremity Bone Changes after Stroke…….35

1.5.1 Neuromuscular Factors…….35

1.5.2 Cardiovascular Factors……..37

1.6 Gap of Knowledge Identified and Potential Contributions of Study……...40

1.7 Objectives of the Study……42

1.8 Research Hypotheses……...42

1.8.1 Hypothesis #1……42

1.8.2 Hypothesis #2……....42

1.8.3 Hypothesis #3……43

CHAPTER 2: METHODOLOGY

2.1 Trial Design……45

2.2 Sample Size Calculation……..46

2.3 Subjects……46

2.3.1 Stroke Group……46

2.3.2 Control Group……...47

2.4 Measurement……47

2.4.1 Demographics……...47

2.4.2 Bone Parameters……48

2.4.2.1 Areal Bone Mineral Density: Dual-energy X-ray Absorptiometry……48

2.4.2.2 Bone Size, Geometry, and Mechanical Properties: pQCT...50

2.4.3 Neuromuscular Function……..55

2.4.3.1 Muscle Strength…….55

2.4.3.2 Motor Skills……...56

2.4.3.3 Disuse……56

2.4.3.4 Spasticity……57

2.4.4 Cardiovascular Function…….....57

2.4.4.1 Vascular Elasticity: Blood Pressure Waveform Analysis….57

2.4.4.2 Impedance Cardiograph…….61

2.4.3.3 Walking Endurance and Oxygen Consumption (VO_{2max}): FitMate™……...64

2.5 Statistical Analysis……...65

CHAPTER 3: RESULTS

3.1 Demographics……..69

3.2 Comparison of Bone Parameters: DXA……..71

3.2.1 Areal Bone Mineral Density…….71

3.3 Comparison of Bone Parameters: pQCT……75

3.3.1 Radius Distal Epiphysis (4% site)…….75

3.3.2 Radius Diaphysis (33% site)…….78

3.5 Comparison of Neuromuscular Parameters…….82

3.6 Comparison of Cardiovascular Parameters……83

3.7 First line of Correlation Analysis: Association of Absolute Bone Parameters with Muscle and Cardiovascular Function in Stroke Group……..……85

3.8 First Set of Regression Analyses: Predicting Absolute Forearm aBMD and Radius Bone Strength Index Values on the Paretic Side……...86

3.8.1 Determinants of Ultradistal Forearm aBMD……87

3.8.2 Determinants of Mid-forearm aBMD……...87

3.8.3 Determinants of 1/3 Forearm aBMD……88

3.8.4 Determinants of Total Forearm aBMD…….88

3.8.5 Determinants of CBSI at 4% Site of Radius…….89

3.8.6 Determinants of p-SSI of the Radius Diaphysis……...89

3.9 Second Line of Correlation Analysis: Association of Percent Side-to-side Difference of Bone Parameters with Neuromuscular and Cardiovascular Function in Stroke Group…….89

3.10 Second Set of Regression Analyses: Predicting Percent Side-to-side Difference in Forearm aBMD and Radius Bone Strength Indices on the Paretic Side ….....91

3.10.1 Determinants of Percent Side-to-side Difference of Ultradistal Forearm aBMD……91

3.10.2 Determinants of Percent Side-to-side Difference in Mid-forearm aBMD…… ……..92

3.10.3 Determinants of Percent Side-to-side Difference in 1/3 Forearm aBMD…… ……...…….92

3.10.4 Determinants of Percent Side-to-side Difference in Total Forearm aBMD…… ……...……...……..93

3.10.5 Determinants of Percent Side-to-side Difference in Compressive Bone Strength Index (CBSI) at the Radius Epiphysis……...……..93

3.10.6 Determinants of Percent Side-to-side Difference in Polar Stress-Strain Index (p-SSI) at the Radius Diaphysis……...….94

3.11 Summary of Results on Regression Analyses……94

CHAPTER 4: DISCUSSION

4.1 Pronounced Side-to-side Difference of Areal Bone Mineral Density Measured by DXA……...98

4.2 Pronounced Side-to-side Difference in Volumetric Bone Mineral Density, Bone Geometry, and Bone Strength Indices of the Radius Measured by pQCT……..…..99

4.2.1 Radius Distal Epiphysis……..100

4.2.2 Radius Diaphysis……102

4.3 Comparison between Trabecular and Cortical Bone Sites……104

4.4 Sex-related Differences……105

4.5 Lack of Significant Main Effects of Group……107

4.6 Determinants of DXA-derived Area Bone Mineral Density and pQCT-derived Bone Strength Indices……..108

4.6.1 Demographic factors……108

4.6.2 Neuromuscular Function……108

4.6.2.1 Muscle Weakness……108

4.6.2.2 Motor Function……110

4.6.2.3 Chronic Disuse……111

4.6.2.4 Spasticity...……...112

4.5.3 Cardiovascular Function……113

4.6.3.1 Vascular Elasticity……113

4.6.3.2 Oxygen Consumption Rate, Cardiac Index and Stroke Index……114

4.7 Clinical Implications……..116

4.8 Limitations and Future Research Directions……..118

4.9 Conclusion…….121

References.....123

LIST OF ABBREVIATIONS

6MWT: 6 minute walking test

aBMD: areal bone mineral density

ADL: activities of daily living

AMT: Abbreviated Mental Test

ANOVA: analysis of variance

BBS: Berg balance scale

BMC: bone mineral content

BMD: bone mineral density

BMI: body mass index

BI: Barthel Index

C_1: large artery elasticity index

C_2: small artery elasticity index

CBSI: compressive bone strength index

CHD: congestive heart disease

CI: cardiac index

CV: coefficient of variation

DXA: dual-energy X-ray absorptiometry

FIM: Functional Independence Measure

FMA: Fugl-Meyer motor assessment

ICG: impedance cardiograph

MAL: Motor Activity Log

MAS: Modified Ashworth Scale

MR: magnetic resonance

PASE: Physical Activity Scale for the Elderly

pQCT: peripheral quantitative computed tomography

p-SSI: polar stress-strain index

UD: ultradistal

vBMD: volumetric bone mineral density

VO_2: oxygen consumption

WHO: World Health Organization

ICC: intraclass correlation coefficient

p: significance level

r: Pearson's correlation coefficient

SD: standard deviation

SI: stroke index

LIST OF TABLES

Table 1. Definition and precision of aBMD……..49

Table 2. Definition and precision of pQCT-derived parameters at the radius distal epiphysis……52

Table 3. Definition and precision of pQCT-derived parameters at the radius distal epiphysis……55

Table 4. Key parameters generated by the CardioVascular Profiling System……..59

Table 5. Major parameters generated by impedance cardiograph (ICG)……...63

Table 6. Subject characteristics……70

Table 7. Comparison of DXA parameters……74

Table 8. Comparison of pQCT parameters: 4% site……77

Table 9. Comparison of pQCT parameters: 33% site……...81

Table 10. Comparison of grip strength data……82

Table 11. Other neuromuscular impairments in the stroke group……83

Table 12. Comparison of cardiovascular parameters……84

Table 13. Association of absolute values of bone parameters with neuromuscular and cardiovascular function……...86

Table 14. Correlation between percent side-to-side difference in bone parameters and neuromuscular and cardiovascular function……90

Table 15. The significant determinants identified in regression analyses: DXA…..95

Table 16. The significant determinants identified in regression analyses: pQCT....96

LIST OF FIGURES

Figure 1. A DXA image of the forearm, showing the ultradistal (UD), mid and 1/3 regions……49

Figure 2. An example of a pQCT image at the 4% site of radius…….52

Figure 3. An example of a pQCT image at the 33% site of radius……...53

Figure 4. A schematic illustration of the parameters required for computation of the polar stress-strain index…….54

Figure 5. An electrical analog model of the arterial system…….58

Figure 6. Impedance cardiograph (ICG)……..63

Figure 7. Difference in macrostructure of the distal radius epiphysis between a stroke subject and a control subject……...75

Figure 8. Difference in macrostructure of the radius diaphysis between a stroke subject and a control subject……..78

APPENDICES

Appendix 1. Abbreviated Mental Test (AMT)……...150

Appendix 2. Ethical Approval ……...151

Appendix 3A. Consent Form (English version) …… ……152

Appendix 3B. Consent Form (Chinese version) ……155

Appendix 4. Physical Activity Scale for the Elderly (PASE)……...157

Appendix 5. Fugl-Meyer Motor Assessment (FMA) ……..159

Appendix 6. Motor Activity Log (MAL)……161

Appendix 7. Modified Ashworth Scale (MAS)……..162

Appendix 8. Arterial Elasticity Guideline Tables……163

Appendix 9. Checking multicollinearity: correlation among variables of neuromuscular and cardiovascular function……164

Appendix 10. Multiple regression analyses for predicting areal bone mineral density (aBMD) of the ultradistal forearm…… ……165

Appendix 11. Multiple regression analyses for predicting areal bone mineral density (aBMD) of the mid-forearm…… ……..……166

Appendix 12. Multiple regression analyses for predicting areal bone mineral density (aBMD) of the 1/3 forearm……..……167

Appendix 13. Multiple regression analyses for predicting areal bone mineral density (aBMD) of the total forearm ……168

Appendix 14. Multiple regression analyses for predicting compressive bone strength index (CBSI) of the radius epiphysis……169

Appendix 15. Multiple regression analyses for predicting polar stress-strain index (p-SSI) of the radius diaphysis……170

Appendix 16. Checking multicollinearity: Correlation of percent side-to-side difference of grip strength with other impairment variables……171

Appendix 17. Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (aBMD) of the ultradistal forearm ……172

Appendix 18. Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (aBMD) of the mid-forearm ……173

Appendix 19. Multiple regression analyses for predicting percent side-to-side difference of areal bone mineral density (aBMD) of the 1/3 forearm……174

Appendix 20. Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (aBMD) of the total forearm……175

Appendix 21. Multiple regression analyses for predicting percent side-to-side difference of compressive bone strength index (CBSI) at the radius epiphysis……176

Appendix 22. Multiple regression analyses for predicting percent side-to-side difference of polar stress-strain index (p-SSI) at the radius diaphysis……177

CHAPTER 1

INTRODUCTION

1.1 Epidemiology of Stroke

According to World Health Organization (WHO) criteria, a stroke is defined as "rapidly developing clinical signs of focal (or global) disturbance of cerebral function, with symptoms lasting 24 hours or longer or leading to death, with no apparent cause other than of vascular origin." Approximately 87% of all strokes are ischemic and the other 13% are hemorrhagic in nature (American Heart Association, 2008).

Stroke is one of the most common global health problems. In the United States, there are approximately 780,000 stroke cases each year, of which 120,000 cases are recurrent. In Hong Kong, 26,394 in-patient discharges and deaths were related to stroke in the year of 2008. The incidence of stroke can be influenced by many factors, including demographic factors such as age, sex, ethnicity, socioeconomic status, geographic location (American Heart Association, 2008; Brass LM., 2006; Everson et al., 2001; Feigin et al. 2009; Towfighi et al., 2008; Wong et al., 2001). Stroke is particularly a health concern among the elderly population (Brass LM., 2006). A recent systematic review has shown that the age-adjusted annual incidence of stroke among individuals 75 years of age is approximately 1,030-1,151 per 100,000 populations per year, compared to an annual incidence of only 66-94 per 100,000 populations per year among people younger than 75 years of age (Feigin et al., 2009). Apart from the demographic factors, various medical conditions (e.g., hypertension, diabetes, obesity, preexisting heart disease, coronary artery disease and dyslipidemia) and lifestyle factors (e.g., smoking, heavy alcohol consumption and lower physical activity) are also associated with increasing the risk of stroke (Alberts et al., 2004; Kelly et al., 2007; Sacco et al., 2006).

Stroke is a major cause of mortality among older adults. In the United States, stroke is the third leading cause of death, accounting for one-sixteenth of total deaths (American Heart Association, 2008). A report by the World Health Organization estimated a total of 2.7 million stroke-related deaths in Asia in the year of 2000, including 1.6 million in China alone (Murray et al., 1996). In Hong Kong, stroke is the fourth leading cause of death behind cancer, heart disease and pneumonia (Centre for Health Protection, Department of Health, Hong Kong, 2010), with age-specific mortality rates increasing dramatically from 416 per 100,000 population among individuals aged 45-64 years to 2,962 per 100,000 population among individuals aged 65 years or above (Centre for Health Protection, Department of Health, Hong Kong, 2010).

Stroke is also a leading cause of long-term disability. Spasticity, muscle weakness, pain, balance problems, perceptual deficits, and cognitive dysfunctions are amongst the common impairments resulting from stroke, causing functional limitations, poor community reintegration and secondary complications such as bone loss, falls and fragility fractures (Beaupre et al., 2006; Gresham et al., 1975; Lamb et al., 2003; Poole et al., 2002; Pang et al., 2006; Pang et al., 2007; Pang et al., 2010; Ramnemark et al., 1998). Approximately 15-30% of individuals with stroke are permanently disabled and 20% require institutional care after three months post-stroke (American Heart Association, 2005). Stroke also imposes a tremendous financial burden on the health care system, in view of costs relating to early critical care, long-term medical treatment, ambulatory care, and lifetime cost (Demaerschalk et al., 2010).

1.2 Falls in Stroke Patients

1.2.1 Incidence and characteristics

Falls are the number one medical complication after stroke among stroke survivors (Davenport et al., 1996; Holloway et al., 2007). Fall risk was exaggerated not only in the acute period, but also remained a considerable health concern throughout the post-stroke lifespan (Carter et al., 2007; Truelsen et al., 2006). The incidence of falls after stroke ranged from 8.9-15.9/1000 patients/day in western countries (Foster et al., 1995; Nyberg et al., 1995), and around 5.5/1000 and 3.4/1000 in China and Thailand respectively (Chaiwanichsiri et al., 2006; Sze et al., 2001). Approximately 3.8-22.0% of the patients fell at least once during hospitalization after stroke (Davenport et al., 1996; Holloway et al., 2007; Tutuarima et al., 1997). During the inpatient rehabilitation phase, the rate of falling was 10.5-47%, with 5-27% of these cases being recurrent falls (Weerdesteyn et al., 2008). Among community-dwelling stroke survivors, the reported incidence of falls was approximately 23-34%, 40-73% and 43-70% when measured at 3 to 4-month, 6-month, and 1-year follow up intervals respectively, with 21-57% of patients falling twice or more (Weerdesteyn et al., 2008).

A distinct difference in fall circumstances was observed between stroke inpatients and community-dwelling stroke survivors. During the inpatient rehabilitation stage, falls occurred predominantly during daytime hours in the patient's room and lavatory (Suzuki et al., 2005). Transfers were the most common activity leading to a fall (Nyberg et al., 1995; Suzuki et al., 2005). In contrast, among community-dwelling stroke survivors, walking was the most commonly mentioned activity (39%-90%) that precipitated a fall, in addition to transfers. However, falls occurred more often indoors than outdoors (Weerdesteyn et al., 2008). The majority of patients fell toward the affected side, or on their hands and knees (Hyndman et al., 2002). Approximately 24% of falls led to bruises, grazes and lacerations, and 6% resulted in fractures. The frequency and types of injuries were not significantly different between first-time

fallers and repeat fallers (Hyndman et al., 2002). White (1988) investigated the circumstances of falls leading to fracture in a group of 53 stroke patients. The study found that 20% of these falls occurred in the bathroom and 18% during transfer activities.

1.2.2 Risk Factors

Various factors have been associated with an increased fall risk among stroke patients. The most consistently reported risk factor is decreased ability to perform activities of daily living (ADL). However, the deficit that most contributes to the risk of falls cannot be distinguished using common ADL assessment tools such as the Barthel Index (BI) (Forster et al., 1995, Sze et al., 2001) and Functional Independence Measure (FIM) (Suzuki T et al., 2005; Teasell et al., 2002).

Swaying when standing and standing up or sitting down has been associated with an increased risk of falls post-stroke (Cheng et al., 1998; Sackley, 1991). Marigold et al. (2006) suggested that falls could be related to slow postural muscle reflexes often demonstrated by post-stroke patients. Previous studies also found that in reaction to perturbations, the postural reflex response was significantly slower among stroke fallers compared to stroke non-fallers (Marigold et al., 2006).

Among the various physical impairments, poor balance and gait deficits were identified as important risk factors (e.g., Berg Balance Scale [BBS] or Tinetti test) (Forster et al., 1995; Mackintosh et al., 2006; Teasell et al., 2002). Hemineglect may also contribute to increased fall risk in individuals with stroke (Mackintosh et al., 2006). Patients with right cerebral infracts are more likely to fall than those with left cerebral infracts, probably due to visuospatial neglect,

proprioceptive impairments, and attention deficits associated with right cerebral infracts (Ugur et al., 2000). Cognitive deficits (Tutuarima et al., 1997) and depression (Jørgensen et al., 2002; Ugur et al., 2000) have also been identified as risk factors for falls. Patients after stroke tend to fall more frequently when they are required to perform dual task such as walking and talking at the same time, probably due to reduced cognitive control (Andersson et al. 2006; Hyndman et al., 2004). On the other hand, depression, which occurs in 30-54% of stroke victims, may cause poor judgment, which lead to falls (Chau et al., 2010; Kotila et al., 1998; Kouwenhoven et al., 2011; Lenzi et al., 2008; Nidhinandana et al., 2010). Falls in stroke patients have also been associated with the use of antidepressant medications (Ensrud et al., 2003).

1.3 Fractures after stroke

1.3.1 Incidence and Characteristics

Fracture is a serious complication after stroke that has high personal, social, and economic costs for patients, their families, and the wider community (Cooper, 1997). It is well documented that individuals with stroke sustain a much higher risk of bone fracture than the reference population (Dennis et al., 2002; Kanis et al., 2001). Kanis et al. (2001) found that within the first year after hospitalization for stroke, the fracture risk was increased >7-fold when compared with the reference population. In the first year, 4% of individuals suffered a fragility fracture, a figure that increased to 15% at 5 years post-stroke (Ramnemark et al., 1998). A study by Whitson et al. (2006) estimated 2-year fracture rates after stroke to be approximately 4.7-6.1%. Dennis et al. (2002) reported similar results. Approximately 4% of stroke patients had a fracture within 2 years of stroke onset, with an incidence rate of 22 per 1,000 patient-years.

Among all the fracture types in stroke survivors, hip fractures are the most common (30-45%), followed by wrist fractures (14-24%) (Dennis et al., 2002; Ramnemark et al., 1998). There is a significant relationship between the side of hemiparesis and the side of fracture (White, 1988), with the majority of fractures (69-75%) occurring on the paretic side (Dennis et al., 2002; White, 1988). Ramnemark et al. (1998) also found that the median time between the stroke onset and the first fracture was approximately 2 years.

Fractures among stroke patients can lead to serious complications, including reduced survival. Among hip-fractured patients with a history of stroke, the mortality rate (29.3%) was higher than those with no history (16.8%) (Ramnemark et al., 2000). Another undesirable consequence of post-stroke fracture is decreased recovery of mobility function. Of hip-fractured patients who had independent ambulatory function pre-admission, only 38% of the patients with a stroke history could regain independence in ambulatory function at discharge, compared with 69% of patients with no stroke history (Ramnemark et al., 2000). Prolonged hospital stay is also a concern among stroke patients with hip fracture (Di Monaco et al., 2003). Di Monaco et al. (2003) showed that hospital stay was significantly longer among those hip-fractured patients with a history of neurological disorder (including stroke) than their peers with no such history.

1.3.2 Risk Factors

Previous studies have identified a number of risk factors of post-stroke fragility fractures. Age is also an important determinant of fragility fractures. Older stroke patients over 80 years of age have a higher rate of fragility fractures than their younger counterparts (Dennis et al., 2002; Kanis et al., 2001). Female gender has been associated with increased risk of fragility fractures

after stroke, probably due to the superimposed effects of postmenopausal bone loss (Dennis et al., 2002; Kanis et al., 2001). Aside from demographic factors, various stroke-related physical impairments were also identified as important risk factors of fragility fractures in patients after stroke. Triceps weakness, leading to reduced ability of the affected arm to outstretch and cushion the impact of the fall, was also identified as an independent risk factor of hip fracture (Chiu et al., 1992). Other notable risk factors included functional limitation of the paretic lower limb (Kanis et al., 2001; Ramnemark et al., 2000), cognitive deficits (Dennis et al., 2002; Whitson et al., 2006), poor trunk control, and visual impairment (Grisso et al., 1991a&b).

The hospitalization time since the first stroke is another determinant of fragility fractures. Increased risk of a fragility fracture following a stroke is associated with a longer duration of stay in the hospital among men, but with a shorter stay among women (Kanis et al., 2001). However, the reason behind this observation is unclear. Interestingly, Melton III et al. (2001) found that fracture risk in patients after stroke increased among patients after stroke with moderate functional impairment, but not those with severe functional impairment. Similarly, Whitson et al. (2006) found that those with intermediate functional disability suffered a higher fracture risk than those with mild or severe functional disability, concluding that those patients with intermediate physical impairment might have more exposure to fall-inducing situations due to their relatively better mobility level. In contrast, the individuals with more severe impairment might have less exposure to situations that may put them at risk of fall (Melton III et al., 2001).

Post-stroke bone loss is another risk factor that is often overlooked. Browner et al. (1993) found that for every two standard deviations decrease in femoral neck bone mineral density (BMD), the risk of a hip fracture in people who had previously sustained a stroke was increased by 7-fold.

1.4 Bone Changes after Stroke

1.4.1 Changes in Upper Extremity Bone Mineral Density as Measured by Dual-energy X-ray Absorptiometry (DXA)

Bone loss is an important contributing factor of fragility fractures in the upper extremities (Poole et al., 2002). Dual-energy X-ray absorptiometry (DXA) has been used as the gold standard to measure BMD in various studies. DXA provides the values of bone mineral content (BMC) in grams, the area of the scanned region in cm^2, and areal bone mineral density (aBMD) in g/cm^2. Areal BMD (aBMD) as measured by DXA provides a good estimate of bone strength and is used to diagnose osteoporosis (Kanis & Gluer, 2000). The results can be interpreted using the WHO's T-score definition of osteoporosis, which is a useful predictor of fracture risk (Blake & Fogelman, 2007). According to the WHO published report, osteoporosis was defined as a BMD T-score $\leq$-2.5 at the spine, hip, or forearm, indicating that the measured aBMD value was at or more than 2.5 standard deviations below the mean value of the young reference population (World Health Organization, 1994).

Secondary bone loss in the paretic upper extremity is common following stroke (Ashe et al., 2006; Hamdy et al., 1993; Hamdy et al., 1995; Jørgensen & Jacobsen, 2001; Ramnemark et al., 1999; Yavuzer et al., 2002). In a cross-sectional study, Hamdy et al. (1993) used DXA to compare the BMC and aBMD between the paretic and non-paretic arms in a sample of 30 patients after stroke and found that the mean percent side-to-side differences in bone mineral content and aBMD of the total arm are 13.80% and 7.95%, respectively. The figures were much more prominent than those found between the dominant and non-dominant side in non-disabled older individuals ($\leq$1.5%), clearly indicating the detrimental effect of stroke on bone health in the

paretic upper extremity (Taaffe et al., 1994). Few prospective studies have examined progressive bone loss after the onset of stroke. Ramnemark et al. (1999) used DXA to measure aBMD in 21 acute stroke patients and found that the aBMD in the ultradistal radius site on the paretic side decreased by 9% in the 1-year follow-up period. In contrast, significant increase (6%) of aBMD on the corresponding skeletal site on the non-paretic side was observed. Another DXA study monitored the proximal humerus aBMD change for 1 year after the onset of stroke (Jørgensen & Jacobsen, 2001) and found significant aBMD decrease (11-27%) in the proximal humerus on the paretic side, but no significant change on the non-paretic side.

There is evidence that bone loss occurs faster in the acute and subacute phases (<6 months) than during the chronic phase of stroke recovery (>6 months). In a 1-year prospective study, the patients with severe paresis in the upper limb sustained an impressive 23% decrease of aBMD in the proximal humerus during the first 7 months after stroke, with only an additional 4% reduction of the same variable when measured at 1-year post-stroke (Jørgensen & Jacobsen, 2001). Moreover, the degree of bone loss is more severe in the paretic upper extremity than the lower extremity (Hamdy et al., 1993; Jørgensen et al., 2000; Jørgensen & Jacobsen, 2001; Pang et al., 2007; Pang et al., 2008). For example, Hamdy et al. (1993) found a 12.4% side-to-side difference in total arm aBMD, but only a 4.3% side-to-side difference in total leg aBMD in a sample of patients after chronic stroke (Hamdy et al., 1993).

1.4.2 Changes in Upper Extremity Bone mineral Density and Geometry as Measured by Peripheral Quantitative Computed Tomography (pQCT)

While DXA is the gold standard for diagnosis of osteoporosis, it provides a 2-dimensional assessment of the 3-dimensional structure of bone (Järvinen et al., 1999; Melton III, 2001). Although aBMD is correlated with bone strength and is often used to predict fractures, bone strength can also be affected by its geometry (Burr & Turner, 2003; Frost, 2003). For example, adding bone to the periosteal surface would increase bone bending or torsional strength even when the absolute bone mass and BMD remain constant, due to the increase in cross-section moment of inertia (Burr & Turner, 2003). In skeletal sites where bones are mainly cortical (e.g. radius and tibial diaphysis), geometry may even be a predominant factor in determining bone strength (Kiratli et al., 2000). Indeed, including geometry in evaluation of bone status has been shown to improve the prediction of fracture risk (Genant et al., 1996; Peacock et al., 1995). DXA, due to its planar nature, is incapable of evaluating bone geometry. In contrast, peripheral quantitative computed tomography (pQCT), a bone imaging technique now widely used in research, yields volumetric BMD (mg/cm^3) measurements and can analyze cortical and trabecular bone separately. It could serve as a useful tool to evaluate bone geometry and mechanical properties such as bone strength indices in bending, compression, and torsion. Incorporating pQCT measurements could thus provide a more comprehensive bone assessment.

Two recent studies have assessed long bone macrostructure in individuals with chronic stroke (onset >1 year) by using pQCT (Ashe et al. 2006, Pang et al 2007). In a cross-sectional study, Pang et al. (2007) examined bone density and the geometric properties of the radius diaphysis (primarily cortical bone) in 47 older adults with chronic stroke. The results showed that the cortical bone mineral content (BMC), cortical vBMD, cortical thickness, and polar stress-strain index (p-SSI, a torsional bone strength index) on the paretic side were significantly lower than the non-paretic side. In another cross-sectional study, Ashe et al. (2006) reported that

the vBMD and BMC measured at the distal radius epiphysis (primarily trabecular bone and a common site of wrist fracture) on the paretic side were lower than their corresponding values on the non-paretic side by 15% and 11%, respectively.

Only one study has used pQCT to examine the longitudinal changes in vBMD and bone strength index in stroke patients (Lazoura et al., 2008). In this study, vBMD and p-SSI in the radius epiphysis and diaphysis were evaluated at 3-month, 6-month and 12-month intervals post-stroke. During the follow-up period, trabecular vBMD and p-SSI reduced by 14.0% and 28.6% respectively in the radius distal epiphysis among male stroke survivors. The corresponding changes in these bone parameters were 9.29% and 19.17% respectively in the female group. In the radius diaphysis, the changes in bone density and strength were less dramatic. Specifically, the cortical vBMD and p-SSI decreased by 4.0% and 7.4% respectively among men, and by 2.6% and 7.0% among women (Lazoura et al. 2008). It is intriguing that the extent of decline in bone vBMD and bone strength index was less among female stroke survivors. One potential explanation was that the male group might be more physically disabled than the female group, which may contribute to more pronounced bone loss. However, this is speculation, because the gender difference in physical impairments was not well documented in the study. Moreover, changes in bone geometry were not reported. For example, it was not known whether the bone loss occurred on the periosteal surface or/and endosteal surface of the bone.

1.4.3 Comparison with Lower Extremity Bone Sites

There are a few differences between the upper extremity and lower extremity sites in terms of bone loss post-stroke. First, the extent of bone loss in lower extremity skeletal sites tend

to be less pronounced than that in the upper extremity sites (Hamdy et al., 1993; Iversen et al., 1989; Jorgensen et al., 2000; Jorgensen & Jacobsen, 2001; Ramnemark et al., 1999; Yavuzer et al., 2002). In a cross-sectional study, Hamdy et al. (1993) demonstrated that the percent side-to-side difference in total arm aBMD, was 3.5% and 12.4% for patients with recent strokes (onset <1 year) and remote strokes (onset > 1 year), respectively. The corresponding percent difference in total leg aBMD between the paretic and non-paretic sides was only at 2.6% and 4.3%, respectively. This less pronounced side-to-side difference in aBMD between the paretic and non-paretic lower limbs could be due to a combination of factors. First, the non-paretic lower extremity also suffers from a certain degree of bone loss, probably due to reduced participation in weight-bearing activities in general (Jorgensen et al., 2000). Second, while learned non-use can be common in the paretic upper extremity, the paretic lower limb is subject to a certain degree of mechanical loading during activities such as standing, transfers and walking.

The paretic lower extremity long bone sites also undergo changes in bone geometry. A recent study by Pang et al. (2010) examined the distal tibial epiphysis in patients after chronic stroke and found that the total vBMD, trabecular vBMD and bone strength index values on the paretic side were all significantly lower than the non-paretic side, but the total area had no significant between-group difference. In another cross-sectional study using a sample of 55 patients after chronic stroke, Pang et al. (2008) showed that at the tibial diaphysis, the cortical bone mineral content was lower on the paretic side than the non-paretic side in both men and women. However, for men, the marrow cavity area on the paretic side was significantly greater than that on non-paretic side but the total area was similar between the two sides. On the other hand, women demonstrated different bone geometric properties in that the total area on the paretic side was significantly smaller than the non-paretic side but the marrow cavity area was

similar between the two sides. These findings suggest the possibility of endosteal resorption in men and periosteal resorption in women post-stroke. The exact causes of the observed sex-specific differences in geometry at the tibial diaphysis remain uncertain.

1.5 Factors Related to Upper Extremity Bone Changes after Stroke

1.5.1 Neuromuscular Factors

The mechanisms underlying alterations in bone structure following stroke are not well understood. Previous studies have attempted to identify the factors associated with bone changes in stroke patients. One of the factors related to upper extremity bone loss post-stroke is impaired motor recovery. Reduced functional ability of the affected upper limb is a common manifestation in stroke patients. Previous studies found that 32% and 37% of patients had severe and mild arm paresis, respectively, upon admission for stroke (Nakayama et al., 1994). In 13% of post-stroke patients, the affected arm remained entirely non-functional, despite the efforts of a comprehensive rehabilitation program (Nakayama et al., 1994). In the upper extremity, more severe muscle weakness/functional limitations have been shown to be significantly associated with lower bone mineral levels (Iwamoto et al., 2001; Pang & Eng, 2005a; Sato et al., 1998; Yavuzer et al., 2002). In a cross-sectional study using DXA, Pang et al. (2005a) reported that muscle strength was a significant predictor of the paretic arm BMC. Initial motor deficits are particularly important in determining subsequent bone loss. In a longitudinal study, Jørgensen and Jacobsen (2001) showed that those with severe initial upper limb motor impairments (as measured by the Scandinavian Stroke Scale) had significantly more bone loss (27%) in the

proximal humerus after 1 year of stroke onset, compared with those who had mild (4%) or moderate (11%) motor impairments initially.

Hand dominance and side of paresis may also influence the degree of bone loss in the upper extremity in individuals with stroke. Naftchi et al. (1975) found that bone loss was more severe among those stroke patients whose non-dominant side was affected by stroke, compared with those whose paretic side was the dominant side. This phenomenon could be related to the fact that individuals with non-dominant upper extremity affected by stroke tend to have more severe impairment such as muscle weakness, pain and stiffness than those with the dominant upper extremity affected (Harris et al., 2006). It was speculated that individuals with the dominant upper extremity affected by stroke may be more inclined to use their dominant hand during the rehabilitation process, because they wish to perform functional tasks in the same way as prior to the onset of stroke. In contrast, if the non-dominant upper extremity happened to be the paretic side, patients may be less motivated to use it in daily activities, because they were not used to using it for these activities before the stroke event (Harris et al., 2006). The less frequent use of the non-dominant paretic upper limb may initiate a vicious cycle of further muscle weakness, loss of range of motion, and other complications such as contractures and subluxation that may lead to further decline in bone mass (Harris et al., 2006).

Aside from muscle weakness, another neuromotor impairment commonly observed after stroke is spasticity. Interestingly, the available DXA studies did not show a significant relationship between spasticity and bone loss. A cross-sectional study found no significant relationship between spasticity and hip aBMD in a group of chronic stroke patients (Pang & Eng, 2005b). In a 1-year longitudinal study, Jørgensen & Jacobsen (2001) did not identify an association between 1-year decline in total arm BMC and spasticity level. The relationship

between bone loss and spasticity may be a complex one. On one hand, spasticity may adversely influence upper extremity function, which leads to disuse (Harris & Eng, 2007), and hence, bone loss. On the other hand, the tonic muscle activity related to spasticity may have a protective effect on bone. Another possible explanation of the non-significant finding is related to the measurement tool used to evaluate spasticity. Both Pang & Eng (2005b) and Jørgensen and Jacobsen (2001) used the Modified Ashworth Scale to assess spasticity (Bohannan & Smith, 1987), which is only a gross measure of resistance to passive movement. Further study is required to study the relationship between spasticity and bone loss in patients after stroke.

Only two studies have examined the relationship between bone parameters and stroke-related impairments using pQCT. In a study using a sample of 15 community-dwelling individuals with stroke, Ashe et al. (2006) demonstrated significant association of bone strength index (Stress-Strain index) in the radius diaphysis with composite muscle strength score (R^2 = 0.72), and Fugl-Meyer motor score (R^2 = 0.38) on the paretic side. In another study, Pang et al. (2007) found that the percentage side-to-side difference in cortical BMC and cortical thickness in the radius diaphysis was independently associated with the percentage side-to-side difference in upper limb extremity muscle strength, spasticity and chronic disuse (measured by the Motor Activity Log) after accounting for demographics such as age, sex, Body Mass Index (BMI) and post-stroke duration (R^2= 0.15- 0.23) (Pang et al., 2007). These studies seem to suggest the importance of recovery of neuromotor function in maintaining the integrity of bone tissue in the paretic upper extremity.

1.5.2 Cardiovascular Factors

One overlooked factor underlying bone health is the integrity of the cardiovascular system. Contrary to popular belief, mounting evidence suggests that cardiovascular disease and osteoporosis are separate, but related, entities (Hak et al., 2000; Kado et al., 2000; Kiel et al., 2001; Rodriguez-Garcia et al., 2005; Whitney et al., 2004). First, the two diseases may share common etiologies (e.g., physical inactivity) (Evenson et al., 1999; Håheim et al., 1993; Garrett et al., 2004; Nguyen et al., 2000; Uusi-Rasi et al., 2001; Lau et al., 2001). Second, the endothelium is an active organ involved in the pathogenesis of various cardiovascular conditions (Whitney et al., 2004). Indeed, endothelial dysfunction is associated with multiple cardiovascular risk factors (Celermajer et al., 1992). Bone is a highly vascularized structure, particularly the trabecular bone, which interacts with endothelial tissue in bone marrow. Hence, impaired blood flow could negatively impact on bone properties and vice versa (Rodriguez-Garcia et al., 2005; Whitney et al., 2004). The structural and function of the blood vessel wall are influenced by various factors including age, environment, and genetic makeup. Older adults are particularly susceptible to decreased vascular elasticity and elevated arterial stiffness (Laurent et al., 2002; Nichols et al., 1990; Safar et al., 1989).

A good number of studies have provided evidence of an association between low BMD and cardiovascular disease (Farhat et al., 2007) or subclinical atherosclerosis (Farhat et al., 2006; Schulz et al., 2004; Vogt et al., 1997). For example, Collins et al. (2009) found that peripheral arterial disease is associated with high rates of hip bone loss in older men (Collins et al., 2009). Barengolts et al. (1998) found a negative correlation between total coronary calcium score and hip aBMD in women with osteoporosis. Increased risk of stroke mortality and cardiovascular death was also associated with reduced bone mass among postmenopausal women (Browner et al., 1991; Hak et al., 2000). Longitudinal studies also showed that progression of aortic

calcification was associated with the rate of decline in lumbar spine BMD and osteoporotic fractures in older adults (Kiel et al., 2001; Naves et al., 2008). Kado et al. (2000) studied osteoporotic fractures in women and found that with each standard deviation decrease in bone mass, there was a1.3 and 1.2-fold risk of dying from congestive heart disease (CHD) or atherosclerosis, respectively. Using magnetic resonance imaging (MRI) technology, it was found that bone marrow perfusion was positively correlated with hip aBMD in patients with osteoporosis and osteopenia (Griffith et al., 2008).

Considering the link between cardiovascular functioning and bone health in other populations, it is possible that cardiovascular mechanisms may also play a role in bone loss among post-stroke survivors given the impaired cardiovascular system of this population. First, most post-stroke patients have a history of cardiovascular disease (Roth et al., 1993) or suboptimally controlled cardiovascular risk factors (Kopunek et al., 2007). Roth et al. (1993) reported that approximately 75% of stroke patients have some form of cardiovascular disease (e.g., arrhythmia, hypertension, congestive heart failure, etc.). In a study of 364 post-stroke patients, Kopunek et al. (2007) found that 99% of the participants had at least one suboptimally controlled cardiovascular risk factor (e.g., hypertension, obesity, impaired fasting glucose, suboptimal low-density lipoprotein, etc.) and 91% had two or more concurrent risk factors inadequately treated.

Second, carotid atherosclerosis and high arterial stiffness are both related to risk factors associated with the occurrence of stroke (Agabiti & Muiesan, 2007; Kannel et al., 1981). It is known that more severe central arterial stiffness has been associated with lower aerobic fitness levels (Cameron et al., 1999; Eugene et al., 1986; Feske et al., 1988). It is thus not surprising that poor cardiovascular fitness, (as indicated by peak oxygen consumption or VO_2) is so prevalent

among stroke survivors (MacKay & Makrides, 2002, 2004; Pang et al. 2005b). Peak VO_2 has been found to be as low as 50% of the age and gender-matched reference values in chronic stroke patients (Eng et al., 2004; Pang & Eng, 2005a, Talbot et al., 2000).

Finally, arterial inflow and endothelium-dependent dilation of vasculature are often impaired in the paretic extremities among stroke survivors (Wang et al., 2002). Although the resting arterial inflow and venous outflow in the paretic upper limb did not significantly differ from those in the non-paretic upper limb, the hyperemic arterial inflow on the paretic side was significantly lower than on the non-paretic side. Moreover, acetycholine-induced cutaneous perfusion was also much lower in the paretic upper limb, when compared with its non-paretic counterpart, indicating impairment in microcirculatory function post-stroke (Wang et al., 2002). Such compromised microcirculatory function was even more pronounced when the paretic upper limb was edematous (Wang et al., 2002).

To date, no study has examined the association between cardiovascular factors and upper extremity bone health in patients after stroke. Previous published studies in chronic stroke have shown that cardiovascular fitness (peak VO_2) is strongly associated with hip aBMD (Pang & Eng, 2005b) and tibial bone strength index (Pang et al., 2008). Whether cardiovascular health is related to paretic upper limb bone health remains uncertain.

1.6 Gap of Knowledge Identified and Potential Contributions of Study

Few studies have examined the relationship between bone health, neuromuscular and cardiovascular factors in chronic stroke patients, and these are not without their limitations. First, although Pang & Eng (2005b) identified a positive relationship between peak VO_2 and hip

aBMD among patients after chronic stroke, it is not known whether a similar relationship exists for the paretic upper extremity. Moreover, peak VO_2, a product of the cardiac output (central component) and arterio-venous oxygen difference (peripheral component), is only an overall measure of cardiovascular function, and does not provide specific information on vascular health. More specific assessment tools are required to evaluate the vascular health component. Second, While Pang et al. (2007) examined the relationship between pQCT-generated bone parameters in the radius diaphysis and various neuromuscular factors (muscle strength, motor function, spasticity, disuse) in patients after chronic stroke, the radius distal epiphysis, which is a common site of fracture in patients after stroke (Dennis et al., 2002), was not measured. Finally, the regression models used in Pang et al. (2007) only accounted for 14.9-25.9% of the variance in pQCT-derived bone parameters in patients after chronic stroke. Certain important factors such as cardiovascular health were not taken into account.

This study is designed to fill the knowledge gaps in this area. It is the first study worldwide to investigate the relative contributions of neuromuscular and cardiovascular impairments to bone health indicators in the upper extremity following stroke. By examining relationship among bone status, muscle function and cardiovascular parameters in patients after chronic stroke, it is possible to identify the key mechanisms involved in bone loss following stroke. Such information can be useful in the design of appropriate intervention strategies for the prevention and treatment of secondary bone loss among stroke survivors. The results may thus have significant impact on the clinical management of post-stroke patients and provide important data for future clinical trials to investigate the efficacy of different rehabilitative strategies in enhancing upper extremity bone health in the stroke population.

1.7 Objectives of the Study

The objectives of this study were:

(1) To assess the difference in DXA-derived aBMD of the forearm and pQCT-derived densitometric and geometric parameters of the radius diaphysis and epiphysis between the paretic and non-paretic sides among patients after chronic stroke.

(2) To compare the side-to-side difference in DXA- and pQCT-derived parameters between individuals with chronic stroke and age-matched non-disabled control subjects.

(3) To examine the associations of neuromuscular and cardiovascular impairments with DXA-derived aBMD of the paretic forearm and pQCT-derived bone strength indices of the paretic radius.

1.8Research Hypotheses

1.8.1 Hypothesis #1

Numerous studies have demonstrated compromised bone mass in other skeletal sites in the paretic upper extremity (Ashe et al., 2006; Jørgensen & Jacobsen, 2000; Pang et al., 2005a; Pang et al., 2007; Prince et al., 1988; Ramnemark et al., 1999a). Therefore, it was hypothesized that the DXA- and pQCT-derived bone parameters of interest measured in the paretic forearm would be significantly different from those in the non-paretic forearm.

1.8.2 Hypothesis #2

In light of the reported accelerated bone loss post-stroke in prospective studies (Ramnemark et al., 1999a; Jørgensen et al., 2001), it was hypothesized that the percent side-to-side difference in

DXA- and pQCT-derived bone parameters of interest in the stroke group would be significantly greater than the corresponding values in the age-matched non-disabled control group.

1.8.3 Hypothesis #3

Previous studies have found various neuromuscular impairments to be associated with aBMD measured in other upper extremity skeletal sites in various post-stroke populations (Ashe et al., 2006; Pang et al., 2007; Pang et al, 2005a; Jørgensen & Jacobsen, 2000). Pang et al. (2007) also found a significant relationship between various neuromuscular deficits and percentage side-to-side difference in cortical thickness in the radius diaphysis. Peak VO_2 has also been identified as a significant determinant of hip aBMD post-stroke (Pang & Eng. 2005b). Based on these previous findings, it was hypothesized that the neuromuscular and cardiovascular impairments would be significant determinants of the DXA- and pQCT-derived bone parameters of interest.

CHAPTER 2

METHODOLOGY

2.1 Trial Design

This was a cross-sectional, exploratory study.

2.2 Sample Size Calculation

All sample size calculations were performed using the online software program Statistics Calculators (version 2.0). A number of studies have shown that the bone mass in various upper extremity skeletal sites in stroke patients demonstrates a significant side-to-side difference (Pang et al., 2007). In patients after chronic stroke, it was found that the percent side-to-side difference in total arm bone mineral content (BMC) was 13.8-22.8% (Pang et al. 2005a; Ramnemark et al., 1999a), with a medium to large effect size of 0.46-0.75. A more recent study has also demonstrated a significant 7.6% side-to-side difference in cortical BMC at the radius diaphysis among chronic stroke patients, with a medium to large effect size of 0.65 (Pang et al., 2007). Therefore, for the within-subject comparisons of bone parameters between the paretic and non-paretic sides in the stroke group (hypothesis 1), a medium to large effect size (0.7) was expected. Based on an analysis using a two-tailed t-test, with an effect size of 0.7, alpha at 5%, and power at 80%, the estimated sample size required to detect a significant side-to-side difference in bone parameters was 34 subjects in the stroke group.

For between-group comparisons of bone parameters (stroke group Vs control group) (hypothesis 2), we expected a similar effect size (0.7), since previous research had demonstrated that the side-to-side difference in bone parameters among control subjects was largely unremarkable (Taaffe et al., 1994). Therefore, the target sample size of control subjects was also 34.

Another objective of the study was to identify the significant determinants of bone parameters in the stroke group. Based on a peripheral quantitative computed tomography (pQCT) study by Pang et al. (2007) in a group of 47 patients after chronic stroke, spasticity and muscle strength were shown to account for 20.2-22.9% of the variance of side-to-side difference in cortical BMC, after accounting for age, sex, body mass index, and post-stroke duration (R^2=0.294-0.321). Thus, a similar effect size was estimated for this study. Based on hierarchical regression analysis (alpha=5%, power=80%, effect size=0.25, 8 predictors), a minimum sample size of 57 stroke subjects was required to detect a significant association of four stroke-impairment variables (i.e., muscle strength, spasticity, disuse and cardiovascular health) with the key bone parameters, after accounting for age, sex, body mass index, and post-stroke duration (hypotheses 3).

2.3 Subjects

2.3.1 Stroke Group

Stroke subjects were recruited from the stroke self-help groups in the community and the existing database of stroke patients who had participated in previous studies of the research team. Relevant information (e.g., demographics, medical history) was obtained by medical records and subject interview. Each subject had to fulfill the following inclusion criteria: (1) a diagnosis of stroke (onset of 6 months or more); (2) aged 18 years or more; (3) medically stable; (4) of Chinese origin; and (5) able to understand simple verbal commands with an Abbreviated Mental Test (AMT) score of 7 or higher (Jitapunkul et al., 1991; Sahadevan et al., 2000) (Appendix 1). The exclusion criteria were: (1) recurrent stroke; (2) other neurological conditions (e.g., Parkinson's disease, multiple sclerosis); (3) significant musculoskeletal conditions (e.g.,

amputations); (4) metal implants in the upper extremity; (5) previous fracture of the upper extremity; (6) were taking drugs for osteoporosis before or after stroke (e.g., bisphosphonates); and (7) other serious illnesses that precluded participation in the study. Ethical approval was obtained from the Hong Kong Polytechnic University (Appendix 2). Each subject gave his or her informed written consent before participating in the study (Appendix 3). All experimental procedures were conducted in accordance with the Declaration of Helsinki.

2.3.2 Control Group

Age- and gender-matched non-disabled control subjects were recruited from community elderly centers, and an existing database of healthy subjects who had participated in previous studies of the research team. To attempt to recruit more eligible healthy subjects, promotional posters containing the information of the study were also posted in the University. The eligibility criteria were the same as those for the stroke group, except that the subjects should not have a history of stroke.

2.4 Measurement

2.4.1 Demographics

Height (in centimeters) and weight (in kilograms) were measured using the Health o meter® (Continental Scale Corp., Bridgeview, IL, USA). Relevant information (e.g., demographics, medical history) was obtained by medical records and subject interview. The Abbreviated Mental Test (AMT), a 10-item cognitive screening instrument with well-established reliability and validity (Jitapunkul et al., 1991; Sahadevan et al., 2000), was used to evaluate the cognitive status of the subjects. Those who had an AMT score of lower than 7 (i.e., significant

cognitive deficits) were excluded from the study. The Physical Activity Scale for the Elderly (PASE) questionnaire was administered by the researcher to assess the physical activity level of each subject (Appendix 4) (Washburn et al., 1993). PASE scores were calculated based on the recall of the amount of participation in various types of activities (e.g., leisure time activity, household activities, and work-related activities) in the past 7 days. PASE has been shown to be a valid and reliable instrument in assessing physical activity in older people (Washburn et al., 1993).

2.4.2 Bone parameters

2.4.2.1 Areal Bone Mineral Density: Dual-energy X-ray Absorptiometry

Dual-energy X-ray absorptiometry (DXA; Hologic Inc., Bedford, MA, USA) was used to scan the forearm on each side. Areal bone mineral density (aBMD, in g/cm^2) of the 1/3 region, mid-region, and ultradistal region of the forearm and the total forearm was determined using the region of interest program (Fig. 1). The definition of each region is presented in Table 1.

The precision of the DXA scanner was determined by measuring 30 healthy subjects twice (28 men and 2 women, mean±SD age = 72.3±5.1 years), with repositioning after the first scan, according to the recommendations of the International Society for Clinical Densitometry (Baim et al., 2005). The coefficient of variation, CV (%) value for each DXA variable is listed in Table 1.

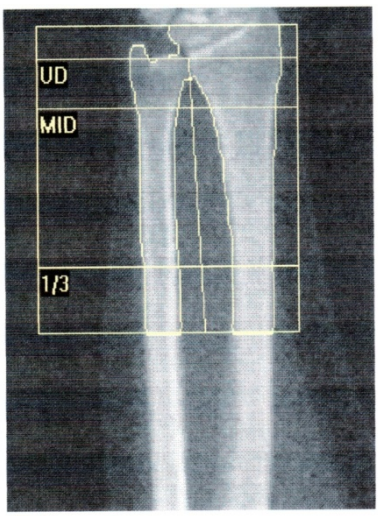

Figure 1. A DXA image of the forearm, showing the ultradistal (UD), mid and 1/3 regions. The aBMD values of the different subregions of the forearm were determined.

Table 1. Definition and precision of aBMD

Parameter	Definition	CV (%)
Ultradistal region aBMD	An area of predominately trabecular bone extending 15mm proximally from the cortical endplate of the radius.	1.55
Mid-region aBMD	The mean density of the mineral content within the area between the ultradistal and one-third region.	1.16
1/3 region aBMD	The mean density of the mineral content within the area centered 1/3 of the length of the forearm below the ulnar styloid.	1.24
Total forearm aBMD	The mean density of the mineral content within the total forearm area scanned (sum of the mid-region, 1/3 region and ultradistal region).	0.93

The definition and precision of DXA-derived parameters are provided (aBMD = areal bone mineral density)

2.4.2.2 Bone Size, Geometry, and Mechanical Properties: pQCT

Peripheral quantitative computed tomography (XCT 3000, Stratec Medizintechnik GmbH; Pforzheim, Germany) was used to generate three-dimensional cross-section scans of the radius on each side. This technique has been used in other studies to measure bone geometry of various populations, including stroke patients (Haapasalo et al., 2000; Kontulainen et al., 2002; Liu et al., 2004; Pang et al., 2006, 2007). After proper positioning, a scout view was obtained and the anatomical reference line was placed at the cortical end plate of the distal radius. A voxel size of 0.4 mm and scan speed of 25 mm/sec was used. One-millimeter-thick scans were obtained at two different sites of the radius: (1) radius epiphysis (at 4% of the total bone length proximal to the distal endplate of the radius; mainly trabecular bone), and (2) radius diaphysis (at 33% of the total bone length proximal to the distal endplate of the radius; mainly cortical bone). Studying the distal epiphysis of the radius was clinically relevant, as it is a common site of fracture in individuals with stroke (i.e., Colles fracture) (Dennis et al., 2002).The radius diaphysis was also selected for measurement due to its anatomical proximity to the origin/insertion of many important muscle groups (e.g., extensor pollicis brevis, pronator teres) . Thus, motor impairment, paresis, and spasticity may potentially have more influence on this site. All the analyses were performed using customized software (Stratec software, Version 6.0). Different density thresholds were used to identify different tissues within each scan. For image analysis of the 4% site, CALCB Contour (outer edge-detection) Mode 2 (iterative contour detection) and Peel Mode 2 were used with outer threshold/inner threshold of 169/400 mg/cm^3. The outer edge of the bone was detected using a threshold of 169 mg/cm^3. A threshold of 400 mg/cm^3 was used to separate the trabecular from the subcortical bone. For image analysis of the 33% site, cortical bone analysis was performed by using CORTBD (Mode 1) with threshold of 710 mg/cm^3. These

thresholds were chosen based on previous studies in stroke (Ashe et al., 2006; Pang et al., 2006, 2007).

For the distal epiphysis of the radius (4% site), the variables of interest were total bone mineral content (total BMC, mg/mm), total area (mm^2), total volumetric bone mineral density (total vBMD, mg/cm^3), and trabecular vBMD (mg/cm^3) (Table 2). The above parameters generated by pQCT were used to compute a compressive bone strength index (CBSI, g^2/cm^4). This CBSI has been used in previous studies to indicate the strength of the bone segment against compressive forces in distal end of long bones (Kontulainen et al., 2002; MacDonald et al., 2006). This is appropriate, considering that long bone epiphysis is primarily subjected to axial compression (Hayes & Myers, 1997). Indeed, it has been shown in a human cadaver study that at the distal tibial epiphysis, the CBSI is highly associated with the failure load, accounting for 85% of the variance (Kontulainen et al., 2008). An example of a pQCT image of the radius distal epiphysis is shown in Figure 2.

Table 2. Definition and precision of pQCT-derived parameters at the radius distal epiphysis

Parameter	Definition	CV (%)
Radius distal epiphysis (4% site)		
Total BMC (mg/mm)	The mean content of the bone material within a 1 mm slice	3.79
Total vBMD (mg/cm^3)	The mean density of the bone material within a 1 mm slice	2.90
Trabecular vBMD (mg/cm^3)	Mean density of the pure trabecular bone within a 1 mm slice	1.76
Total area (mm^2)	Cross-sectional area of the bone after the soft tissue has been peeled off	3.02
CBSI (g^2/cm^4)	A strength index indicating the resistance against compressive forces in distal end of long bones	5.30

The definition and precision of the pQCT-derived parameters at the radius epiphysis are provided (total BMC = total bone mineral content; total vBMD = total volumetric bone mineral density; trabecular vBMD = trabecular volumetric bone mineral density; CBSI = compute a compressive bone strength index).

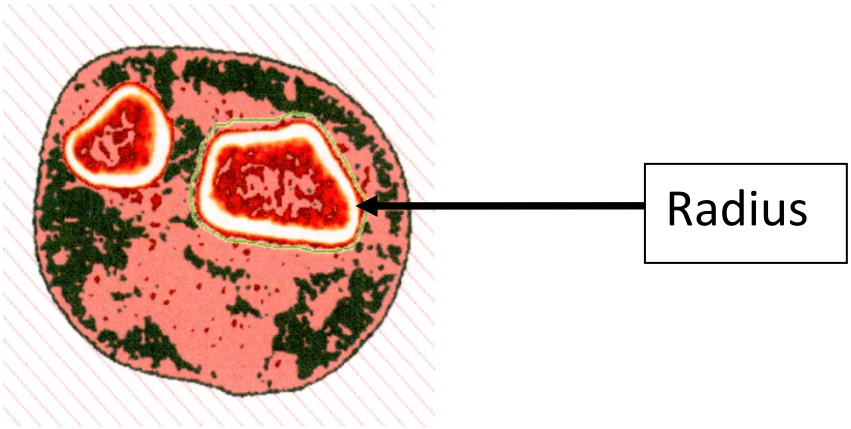

Figure 2. An example of a pQCT image at the 4% site of radius.
The trabecular bone (red area) is surrounded by a cortical bone shell (white area).

For the radius diaphysis (33% site), the variables of interest were total area (mm^2), cortical bone area (mm^2), cortical BMC (mg/mm), cortical vBMD (mg/cm^3), and cortical

thickness (mm). Marrow cavity area (mm^2) was derived from subtracting cortical bone area from the total area. An example of a pQCT image of the radius 33% site is shown in Figure 3.

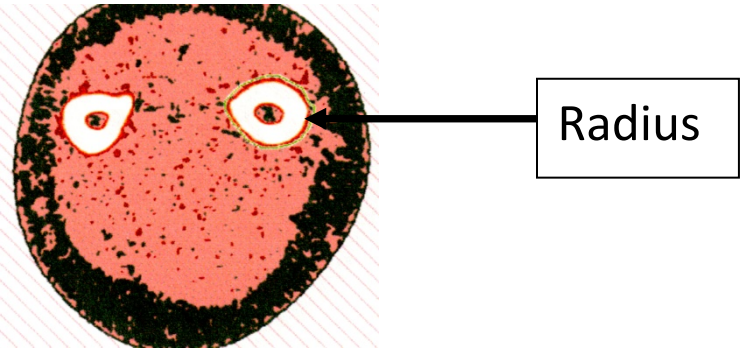

Figure 3. An example of a pQCT image at the 33% site of radius.
The cortical bone shell is much thicker, which surrounds the bone marrow.

Additionally, a polar stress-strain index (p-SSI, mm^3) was computed automatically by the pQCT system using the following formula (Haapasalo et al., 2000; Kontulainen et al., 2002; Leonard & Shore, 2003; Schiessl et al., 1996) (Fig. 4):

$$p\text{-}SSI = \frac{\sum [(A_z \times d_z^2)(BMD_{cort}/ND)]}{d_{max}}$$

where A_z represents the area of each pixel, d_z is the distance between the pixel and the corresponding torsion (z) axis, ND is the normal physiological bone density (1,200 mg/cm^3), and d_{max} is the maximum distance to the center of gravity (Ferretti et al., 2002; Kontulainen et al., 2002). The p-SSI reflects the torsional rigidity of the long bone shaft (Kontulainen et al., 2002).

The p-SSI in the radial diaphysis has been found to be highly correlated to the failure load when the bone is loaded in 3-point bending and is thus considered a valid indicator of bone strength (Lochmüller et al., 2002; Wilhelm et al., 1999).

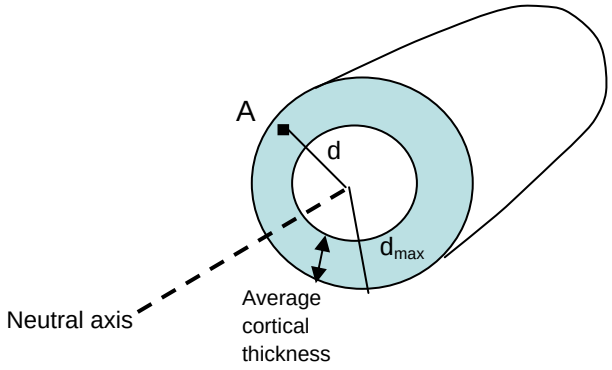

Figure 4. A schematic illustration of the parameters required for computation of the polar stress-strain index.
If bone material is distributed further away from the center, a higher polar stress-strain index will be obtained. A represents the area of each pixel, d is the distance between the pixel and the corresponding torsion axis, and d_{max} is the maximum distance to the center of gravity.

Similar to the DXA, the precision of the pQCT scanner was established by testing 30 healthy subjects twice, with repositioning after the first scan. The CV% value for each variable of interest is displayed in Table 3.

Table 3. Definition and precision of pQCT-derived parameters at the radius distal epiphysis

Parameter	Definition	CV (%)
Radius diaphysis (33% site)		
Cortical BMC (mg/mm)	The mineral content of the pure cortical bone within a 1 mm slice.	0.69
Cortical vBMD (mg/cm^3)	The mean density of the pure cortical bone within a 1 mm slice	0.54
Total area (mm^2)	Cross-sectional area of the bone after the soft tissue has been peeled off	1.97
Cortical area (mm^2)	The area assigned to be pure cortical	1.01
Marrow cavity area (mm^2)	The cortical area subtracted from the total area	6.35
Cortical thickness (mm)	Difference between the outer and the inner radius of the cortical shell	2.05
p-SSI (mm^3)	Bone strength index indicating the resistance against torsional loads at cortical bone sites	2.36

Definition and precision of pQCT-derived parameters at the radius distal epiphysis are provided (cortical BMC = cortical bone mineral content; cortical vBMD = cortical volumetric bone mineral density; p-SSI = polar stress-strain index).

2.4.3 Neuromuscular Function

2.4.3.1 Muscle Strength

Hand grip strength (kg) was measured with a Jamar dynamometer (Sammons Preston, Mississauga, Ontario, Canada). Subjects were asked to seat on a chair; the test position was standardized with shoulder placed at neutral, 0°flexion, elbow in 90° flexion, and wrist in neutral position. Subjects were then instructed to squeeze the dynamometer as hard as possible for 5 sec, with a rest period of about 1 minute in between to avoid fatigue. A total of three trials were performed to obtain the mean hand grip strength on both sides. Based on the data obtained from these three trials, the test-retest reliability was evaluated. The reliability was found to be high for the paretic side ($ICC_{3.1} = 0.984$) and non-paretic side ($ICC_{3.1} = 0.966$) in the stroke group, and the dominant side ($ICC_{3.1} = 0.943$) and non-dominant ($ICC_{3.1} = 0.958$) side in the control group.

2.4.3.2 Motor Skills

The Fugl-Meyer Motor Assessment (FMA) (Fugl et al., 1975) was used to evaluate the severity of impairment in the upper extremity (Appendix 5). It was based on the performance of 33 tasks, which assessed the quality of movements and coordination. A score from 0 to 2 was given to each task, with a higher score indicating better performance (0 = no performance; 2 = complete performance). The maximum upper limb motor score was 66. FMA has shown excellent intra- (Pearson's correlation coefficient (r) = 0.995-0.996) and inter-rater reliability (r = 0.89-0.95) (Gladstone et al., 2002; Sanford et al., 1993).

2.4.3.3 Disuse

To evaluate how much the person used the paretic upper extremity in daily activities, the 30-item Motor Activity Log (MAL) was used (Page et al., 2004) (Appendix 6). Each item in the MAL is related to a functional task (e.g., opening a drawer, brushing teeth, etc.) The questionnaire was administered by the researcher and the subjects were asked to indicate how much they used the paretic arm in each task with a score from 0 to 5 (0: paretic arm not used; 1: occasionally tried to use the weaker arm; 2: sometimes used the affect arm, but did most of the activity with the non-affect arm; 3: used the weaker arm about half as much as before the stroke; 4: used the weaker arm almost as much as before the stroke; 5: used the weaker arm as much as before stroke). The scores for the 30 items were averaged to obtain a mean MAL score. The Motor Activity Log has high internal consistency and reasonable construct validity (van der Lee et al., 2004).

2.4.3.4 Spasticity

The Modified Ashworth Scale (MAS) was used to assess resistance to passive movements in the elbow on the paretic side (Bohannon & Smith, 1987)(Appendix 7). A higher score on the scale indicates more severe spasticity. The MAS has demonstrated acceptable reliability (Kendall's tau correlation =0.847) (Pandyan et al., 1999).

2.4.4 Cardiovascular Function

2.4.4.1 Vascular Elasticity: Blood Pressure Waveform Analysis

The HDI/PulseWave CR-2000 Research CardioVascular Profiling System (Hypertension Diagnostics Inc., Eagan, MN, USA) was used to measure the integrity of the vascular system (Qin et al., 2002). The system is a reliable, valid, and non-invasive tool to measure arterial compliance (Zimlichman et al., 2005), and has been extensively used in cardiovascular research worldwide (Cohn et al., 1995; Duprez et al., 2000; McVeigth et al., 1993).

An electrical analog model (a modified Windkessel model) (Hypertension Diagnostics, Inc., 2005) was used to generate the hemodynamics results (Fig. 5). In this model, the vasculature is represented as consisting of a capacitative compliance element (large artery elasticity index), an oscillatory or reflective compliance element (small artery elasticity index), an inductance, and a resistance (systemic vascular resistance), during the diastolic decay portion of the cardiac cycle. The following figure represents the modified Windkessel model, where P_1 is the proximal arterial pressure, P_2 is the distal arterial pressure, C_1 is the large artery elasticity index, C_2 is the small artery elasticity index, L is the inertance of the blood, R is the systemic vascular resistance, and I_{in} is the flow into the arterial system during systole.

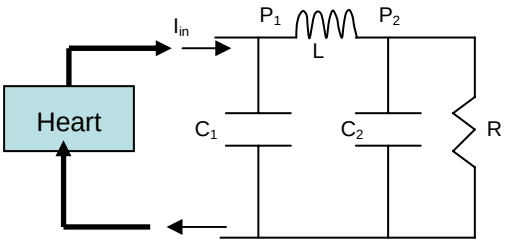

Figure 5. An electrical analog model of the arterial system
The modified Windkessel model is used by the CardioVascular Profiling System to generate the hemodynamic results.

The decay of the voltage in this model could be represented as the solution to a third order differential equation of the circuit, with 6 unknown "A" elements (Hypertension Diagnostics Inc., 2005).

$$\mathbf{P_2(t) = A_{1e}^{-A}2^t + A_3e^{-A}4^t \cos (A_5t + A_6)}$$

The "A" parameters were determined by a non-linear curve fitting approach to match the shape of the above third order equation to that of the diastolic decay. Mathematical relationships existed which related the model values of C_1 and C_2, to the "A" parameters and R. R was calculated as the mean arterial pressure (mmHg) divided by the estimated cardiac output (liters per minute) (Hypertension Diagnostics Inc., 2005).

$$\mathbf{C_1 = 2A_4 [(A_2 + A_4)^2 + A_5^2]/RA_2 (2A_4 + A_2) (A_4^2 + A_5^2)}$$

$$\mathbf{C_2 = 1/R (2A_4 + A_2)}$$

The main outcome parameters generated by the system are summarized in Table 4.

Table 4. Key parameters generated by the CardioVascular Profiling System

Elasticity Index	Explanation
C_1	Large artery elasticity index (capacitative arterial compliance) (mL/mmHg ×10)
C_2	Small artery elasticity index (oscillatory or reflective arterial compliance) (mL/mmHg ×100)

The outcome measures generated by the CardioVascular Profiling System are large artery elasticity index (C_1) and small artery elasticity index (C_2).

To measure C_1 and C_2, the subject was asked to rest in a supine position, and a blood pressure cuff was placed on the left upper-arm and a rigid plastic wrist stabilizer was placed on the right wrist to minimize movement of the radial artery during the measurement. With the right forearm resting in a supine position, a piezoelectric-based, acoustical sensor was placed over the radial artery adjacent to the styloid process of the radius by the wrist. The sensor was adjusted to the highest relative signal strength. Blood pressure was measured by a linear dynamic deflation method. When the waveform was stable and satisfactory, the radial arterial blood pressure waveform data over a 30-second period was acquired for compliance analysis (Hypertension Diagnostics Inc., 2005). Four trials were performed, and the two trials that showed the closest readings were averaged to yield the mean value of the large (C_1) and small (C_2) artery elasticity index. A 1-minute rest period was provided between trials.

The C_1 and C_2 provided measurements of function and structure of the large and very small arteries, respectively. Generally, the higher the value, the more elastic and healthy the arteries are. C_1 is a measure of the elasticity of the aorta and large arteries. With age or in the presence of atherosclerotic disease, the walls of these large arteries will demonstrate increasing stiffness (Duprez et al., 2001). On the other hand, C_2 has been correlated with flow-mediated

vasodilatation, a well-established measure of endothelial function (Wilson et al., 2004). This dysfunction of the endothelium occurs in the entire arterial system, but it can be more easily detected in very small arteries and arterioles. In fact, a reduction in C_2 is often the first sign of a developing atherosclerosis (Syeda et al., 2003). A previous study has also shown that for every two units of decrease in C_2, there is a 50% increase in cardiac events (Grey et al., 2003). Young people normally have higher C_1 and C_2 than older people, and men tend to have higher C_1 and C_2 than women (Winer et al., 2001) (Appendix 8). The subjects were categorized to have abnormal or borderline or normal C_1 and C_2 values according to the reference values provided by the manufacturer (Hypertension Diagnostics Inc., 2005).

To establish the test-retest reliability of the system to measure C_1 and C_2, the measurement procedures described above were repeated after a brief rest period. The test-retest reliability was found to be high, regardless of whether the acoustic sensor was placed on the paretic side (intraclass correlation coefficient $ICC_{3,1} = 0.954$ for C_1, $ICC_{3,1} = 0.948$ for C_2) or non-paretic side ($ICC_{3,1} = 0.934$ for C_1, $ICC_{3,1} = 0.954$ for C_2) among stroke subjects, or the dominant side ($ICC_{3,1} = 0.897$ for C_1, $ICC_{3,1} = 0.946$ for C_2) or non-dominant side ($ICC_{3,1} = 0.911$ for C_1, $ICC_{3,1} = 0.918$ for C_2) among control subjects. In addition, no significant difference was found in C_1 and C_2 values within the same subjects when the sensor was placed on the paretic or the non-paretic sides in the stroke group (C_1: $p=0.249$, C_2: $p=0.629$) or the dominant side or non-dominant side in the control group (C_1: $p=0.549$, C_2: $p=0.988$). Therefore, the side of paresis/hand dominance did not seem to affect the C_1 and C_2 values and their reliability. For subsequent analysis, only the values obtained when the sensor was placed on the paretic side (stroke group) or the non-dominant side (control group) were used.

It is known that certain cardiovascular measures may have a circadian rhythm (Fagard et al., 2008; Rosa et al., 2008). In light of this, reliability tests were performed to determine whether the time of testing (i.e., morning vs. afternoon) has any effect on the C_1 and C_2 values. Ten non-disabled subjects (five men and five women) were enrolled in these reliability tests. No significant difference was found in mean values of C_1 and C_2 when the testing was conducted in the morning or afternoon (C_1: p=0.634; C_2: p=0.713).

The reliability measures obtained in our study seem to be similar with those previously reported in the literature. In Zimlichman et al (2005), intra-visit measurements of arterial compliance indices differed by less than 3% (range, 0.36% to 2.97%), and all inter-visit measures differed by less than 4% (range, 0.24% to 3.67%). In our study, the intra-visit measurements, differed by less than 3.5% (range, 0.17% to 3.44%). On the other hand, the inter-visit measures between am & pm differed by less than 5.5% (range, 1.34% to 5.33%). Difference in subject characteristics and experimental procedures may partially account for the difference in findings. For example, a sample of healthy subjects was used in their study whereas people with stroke are used in ours. A 5-min rest period was provided between the two assessments on the same day (intra-visit measurements), which is longer than what is used in our study (1 min). For inter-visit measurements, the time lapse between assessments was 4 weeks in their study. In contrast, we assessed the difference of the measurements taken in morning session and those taken in the afternoon session.

2.4.4.2 Impedance Cardiograph

Stroke volume and cardiac output profile at rest were measured by impedance cardiograph (ICG) (HIC-3000/T model, BIT Inc., Chapel Hill, NC, USA). ICG is a non-invasive

method to evaluate cardiac output and has been validated in various populations (Charloux et al., 2000; Pianosi, 2004; Richard et al., 2001). The ICG technology involves emission of a low-grade electrical current of 2-4 mA through skin electrodes. Based on information of returned current received by the sensing electrodes, the impedance to current flow is calculated via an algorithm. ICG basically measures the changes of electrical resistance in the thorax. As blood offers the least resistance to electrical current in the thorax, and with each heartbeat, conductivity changes as the blood distends the heart and the aorta.

Subjects were instructed to rest in a supine position. After proper skin preparation, four dual electrodes were placed on specific areas on the neck and thorax (Figure 6). Two electrodes were positioned directly inferior to the ear lobe and the other two were positioned on each side of the rib cage on the mid-axillary line at the level of the xiphoid process. Data were recorded for a period of 10 minutes, and the data for the last 3 minutes of the recording period were used to calculate the mean value of each key parameter. The major parameters generated by this system are listed in Table 5.

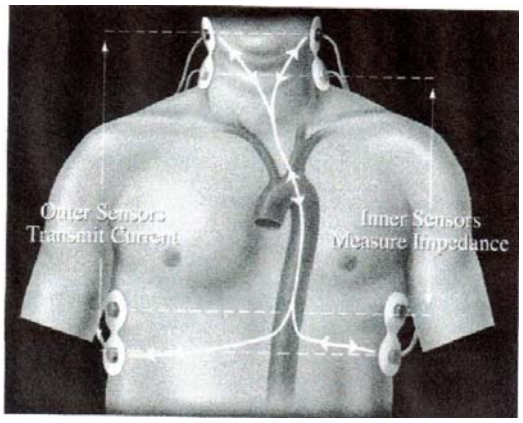

Figure 6. Impedance cardiograph (ICG)

Eight-spot surface electrode array for thoracic electrical bioimpedance application. The outer electrodes introduce the current, and the inner electrodes sense the corresponding voltage changes through the cardiac cycle and the electrocardiogram (Reprinted from CardioDynamics, The ICG Company Webs).

Table 5. Major parameters generated by impedance cardiograph (ICG)

Hemodynamic	Explanation
Cardiac Index (CI)	Cardiac output normalized for the body surface area of subject ($L/min/m^2$)
Stroke Index (SI)	Stroke volume normalized for the body surface area of subject ($mL/beat/m^2$)

The major parameters generated by the impedance cardiograph system (ICG) are cardiac index (CI) and stroke index (SI).

Test-retest reliability of the ICG system to measure cardiac index (CI) and stroke index (SI) was established by repeating the recording procedures in all subjects after a brief rest. The reliability was found to be high in both the stroke group (CI: $ICC_{3,1} = 0.832$, SI: $ICC_{3,1} = 0.904$) and the control group (CI: $ICC_{3,1} = 0.948$, SI: $ICC_{3,1} = 0.955$).

Reliability tests were performed to determine whether the time of testing (i.e., morning vs. afternoon) had any significant influence on the CI and SI values. Ten non-disabled subjects (five men and five women) participated in these experiments. No significant difference was found in CI ($p=0.311$) and SI values ($p=0.743$) obtained in the morning and afternoon sessions.

2.4.4.3 Walking Endurance and Oxygen Consumption (VO_{2max}): FitMate™

The FitMate™ metabolic system (Cosmed, Rome, Italy) was used to measure the oxygen consumption rate (VO_2) during the 6 minute walking test (6MWT). FitMate™ is a reliable and

valid metabolic device designed for measuring oxygen consumption and energy expenditure during rest and exercise (Nieman et al., 2006, 2007). A turbine flowmeter was used to measure the ventilation, and a galvanic fuel cell oxygen sensor was used to analyze the fraction of expired oxygen. VO_2 was calculated intrinsically using the equation below:

$$\mathbf{VO_2 = (FiO_2 * IV - FeO_2 * EV) * RF * STPD}$$

where FiO_2 is the fraction of inspired O_2, IV is the inspired volume, FeO_2 is the fraction of expired O_2, EV is the expired volume, RF is the respiratory frequency, and STPD refers to standard temperature and pressure, dry.

Subjects were instructed to walk along a 15 m corridor and cover as much distance as they could in 6 minutes. The total distance walked (m) was recorded. The 6MWT is a common submaximal exercise test used for various populations, and the VO_2 obtained during the 6MWT is significantly associated with maximal VO_2 (the gold standard of cardiorespiratory fitness) in stroke patients (Eng et al., 2004; Pang & Eng, 2005b). Data on VO_2 during the last 30 seconds of the 6MWT were averaged for each subject (Pang & Eng, 2005b). This method of data analysis was previously used by Eng et al. (2004) and Pang et al. (2005). It is well known that the VO_2 rate would plateau after approximately 3 minutes at a given submaximal exercise workload, as in the 6MWT (Pang et al. 2005; Tomczak et al. 2008). Before data averaging, the profile of the VO_2 data obtained was checked visually to make sure that there was no unusual fluctuation of the values during the last 30s of the 6MWT. This parameter has been shown to have a good correlation with maximal VO_2 in chronic stroke patients ($r = 0.66$) (Eng et al., 2004).

2.5 Statistical Analysis

All statistical analyses were performed using SPSS 17.0 software (SPSS Inc., Chicago, IL, USA) and a significance level of 0.05 (two-tailed). In the primary analysis, analysis of variance (ANOVA) with mixed design [within-subject factor: side (paretic or non-dominant Vs non-paretic or dominant), between-subject factor: group (stroke Vs control)] were performed for each outcome (i.e. aBMD) (hypothesis 1 and 2). Post-hoc paired t-tests were then used to compare the bone parameters between the paretic and non-paretic sides among the stroke subjects, and between the dominant and non-dominant sides among the controls. The level of significance was adjusted according to the number of comparisons made (i.e., 0.025). Next, for each bone parameter, a percent side-to-side difference score was computed by first subtracting the value of the non-paretic side from that of the paretic side and then dividing it by the value of the non-paretic side and multiplying with a factor of 100. Thus, a negative percent side-to-side difference score indicated that the value on the paretic side was lower than that on the non-paretic side. In the secondary analysis, the data collected from the male and female subjects were analyzed separately using the above statistical methods.

Pearson product-moment correlation coefficient (r) or Spearman's rho was used to determine the degree of association of the bone parameters of interest with other demographic (e.g., age, post-stroke duration) and clinical factors (e.g., muscle strength, spasticity, elasticity indices) within the stroke group), depending on whether the assumptions of parametric statistics were fulfilled. Hierarchical multiple regression analyses were then performed to identify the significant determinants of the aBMD of the 1/3, mid-, ultradistal, and total forearm regions, as well as the bone strength indices measured at the radius epiphysis (i.e., CBSI) and diaphysis (i.e., p-SSI) among stroke patients (i.e., dependent variables in regression) (hypothesis 3). The selection of the predictor variables was based on biological relevance and the results from the

bivariate correlations. To check for multicollinearity, bivariate correlation analyses were performed to assess the degree of association among the stroke impairment variables. Any predictor variables that had a correlation of > 0.5 should not be included in the same regression model (Darren & Mallery, 2010). First, age, sex, body mass index (BMI), and post-stroke duration were forced into the regression model (Enter method). Next, those stroke impairments that were found to have significant association with the dependent variable in the bivariate correlation analysis were then entered into the regression model (Enter method). For spasticity, the stroke subjects were categorized into either of two groups for the regression analysis: no or mild spasticity (MAS score = 0-1.5) and moderate to severe spasticity (MAS score = 2-4).

The above analysis would yield a large number of regression models, but it was necessary for several reasons. The bone structure is different between the ultradistal region (mostly trabecular bone) vs the diaphyseal sites (mostly cortical bone). The former is primarily subjected to compressive forces whereas the latter is more exposed to torsional and bending forces (Hayes & Myers, 1997; Frank et al., 2010). It is thus likely that different regions of the bone may respond to the same pathology differently. Indeed, there is evidence from previous stroke literature that different stroke impairments may have differential effects on various parts of the long bone. For example, spasticity and mobility are related to the bone strength index of the tibial distal epiphysis, but not the tibial diaphysis (Pang et al. 2008; Pang et al. 2010). In aerospace research, it has also been shown that the tibial distal epiphysis sustains more rapid bone loss than tibial diaphysis in response to microgravity (Vico et al. 2000). We feel that the association of different stroke impairments with bone density/strength of various parts of the forearm is worth exploring.

CHAPTER 3

RESULTS

3.1 Demographics

Sixty-five patients after stroke and 34 control subjects participated in this study. The demographic data of the subjects are presented in Table 6. The majority of the stroke subjects had an ischemic stroke, and the mean post-stroke duration was 47.8 months. The control group had significantly higher physical activity (i.e., PASE) score than the stroke group ($p<0.001$). The stroke group also had significantly higher proportion of people with hypertension (Chi square test; $p<0.001$), diabetes (Chi square test; $p<0.001$), and high cholesterol (Chi square test; $p<0.001$). Not surprisingly, significantly higher proportion of people in the stroke group were taking antihypertensive agents ($p<0.001$), hypoglycemic agents ($p<0.05$), and hypolipidemic agents ($p<0.001$). There was no significant difference between the two groups in other variables listed in the Table 6, such as age, male-to-female ratio, height, weight, and BMI. There was also no significant correlation between the side of paresis and side of hand dominance in the stroke group ($p=0.727$).

Table 6. Subject characteristics

	Stroke group (n=65)			Control group (n=34)			p
	male	**female**	**all**	**male**	**female**	**all**	
Basic demographics							
Age (year)	60.9±10.3	59.4±11.3	60.1±10.7	60.7±7.3	59.6±9.8	60.2±8.5	0.997
Sex (men/women), n			33:32			17:17	0.920
Height (cm)	165.8±6.8	154.7±6.2	160.3±8.5	164.3±5.5	153.3±5.8	159.8±7.2	0.743
Weight (kg)	67.8±12.4	56.1±9.9	62.1±12.6	64.4±6.9	59.1±10.2	61.7±9.0	0.888
Body Mass Index (BMI), kg/m^2	24.5±3.5	23.3±3.4	23.9±3.5	23.8±2.4	24.7±3.7	24.2±3.1	0.439
Number of postmenopausal women		23			13		0.862
Postmenopausal years (female only)		10.3±11.5			10.1±11.3		0.951
Physical activity score (PASE)	103.1±63.4	85.0±53.9	94.0±59.15	160.0±81.6	138.3±38.9	152.3±61.7	**<0.001***
Receiving physiotherapy or occupational therapy during the study period, n	2	6	8	NA	NA	NA	---
Side of hand dominance (left/right), n	2/31	1/31	3/62	0/17	0/17	0/34	---
Medical history							
Hypertension, n	27	21	48	8	6	14	**0.001***
Diabetes, n	7	11	18	1	0	1	**0.003***
High cholesterol, n	12	11	23	0	2	2	**0.001***
Total number of co-morbid conditions, n	1.6±1.0	1.9±1.3	1.7±1.2	0.7±0.8	0.8±0.9	0.7±0.8	**0.026***
Medications							
Antihypertensive agents, n	20	20	40	3	4	7	**<0.001***
Anticoagulants, n	22	19	41	0	0	0	**<0.001***
Anticonvulsive agents, n	2	2	4	0	0	0	0.140
Hypolipidemic agents, n	12	14	26	1	0	1	**<0.001***

Hypoglycemic agents, n	2	5	7	0	0	0	**0.047***
Analgesics, n	0	2	2	0	0	0	0.301
Antidepressants, n	1	5	6	0	1	1	0.246
Antipsychotic agents, n	1	2	3	0	0	0	0.203
Anti-inflammatory agents, n	1	3	4	0	0	0	0.140
Immunosuppressive, n	1	1	2	0	0	0	0.301
Vitamin D supplementation, n	1	1	2	2	2	4	0.089
Calcium supplementation, n	3	3	6	1	3	4	0.710
Stroke characteristics							
Type of stroke (ischemic/hemorrhagic/unknown), n	22/9/2	14/15/3	36/24/5	NA	NA	NA	---
Side of paresis (left/right), n	21/12	7/25	28/37	NA	NA	NA	---
Duration after stroke (month)	45.3±43.7	48.3±41.8	47.8±46.0	NA	NA	NA	---

Data are mean ± SD unless indicated otherwise; NA, not applicable; *statistically significant difference between the stroke and control groups ($p<0.05$)

3.2 Comparison of Bone Parameters: DXA

3.2.1 Areal Bone Mineral Density

When the aBMD data of all stroke and control subjects were analyzed (i.e., primary analysis), a significant main effect of side for all four regions of the forearm was found ($p<0.005$) (Table 7). There was also a significant side × group interaction effect for the ultradistal ($p=0.006$), and mid regions of the forearm ($p=0.007$), as well as the total forearm ($p=0.001$). There was a trend for a side × group interaction effect for the 1/3 region of the forearm, but it did not quite reach statistical significance ($p = 0.083$). Post-hoc analysis demonstrated that in the

stroke group, the aBMD values on the paretic side were significantly lower than those in the non-paretic side ($p<0.001$) in all measured forearm regions, with a side-to-side difference varying from 5.4% to 9.6%. In contrast, while the control group also demonstrated a significant side-to-side difference of aBMD in the mid ($p=0.016$), 1/3 ($p=0.012$), and total forearm regions ($p=0.009$), the magnitude of the difference was much smaller, ranging from 1.3-1.9%.

The data of male and female subjects were also analyzed separately (i.e., secondary analysis). Regardless of gender, the aBMD values on the paretic side were significantly lower than those on the non-paretic side in the stroke group ($p<0.025$), except the aBMD of the 1/3 region among the male subjects ($p=0.197$) (Table 7). In contrast with the female stroke survivors, the side × group interaction effect was not significant for the mid ($p=0.306$), 1/3 ($p=0.955$) and total forearm ($p=0.084$) regions in the male group, indicating that the side-to-side difference of aBMD in these skeletal sites between the male stroke and control groups was less pronounced than their female counterparts.

The overall significant main effect of side was attributable to the remarkable difference between the paretic and non-paretic sides in the stroke group, despite that the side-to-side difference in the same parameters demonstrated by the control group is much smaller. The lack of significant main effect of group, on the other hand, is mainly due to the fact that the bone parameters measured on the dominant side of control subjects are not that different from those on the non-paretic side of stroke subjects. The results may indicate that the non-paretic side has similar bone health status as in non-disabled controls. It is likely that the overall bone health condition was not particularly compromised prior to stroke, compared with age-matched controls. After the stroke, however, the paretic side may have sustained substantial changes in bone density/geometry, which accounted for the side-to-side difference in bone parameters in the

stroke group, and also the significant side × group interaction effect (i.e. the magnitude of side-to-side difference in bone parameters is group-dependent).

Table 7. Comparison of DXA parameters

Parameter	Stroke group (n=65)			Control group (n=34)			Main effect: Side		Main effect: Group		Side × group interaction effect	
aBMD (g/cm^2)	**Paretic side**	**Non-paretic side**	**Percent difference**	**Non-dominant side**	**Dominant side**	**Percent difference**	**F**	**p**	**F**	**p**	**F**	**p**
Ultradistal												
Male	0.396±0.074	0.436±0.088	**-8.6±11.8†**	0.409±0.061	0.411±0.055	-0.6±5.1	6.260	**0.016***	0.104	0.748	5.106	**0.028***
Female	0.306±0.100	0.343±0.714	**-10.7±23.7†**	0.345±0.046	0.348±0.052	-0.8±5.4	4.238	**0.045***	1.116	0.296	2.989	0.090
All	0.352±0.098	0.390±0.093	**-9.6±18.5†**	0.377±0.062	0.379±0.062	-0.7±5.1	10.476	**0.002***	0.166	0.684	7.978	**0.006***
Mid												
Male	0.597±0.099	0.627±0.076	**-5.1±8.4†**	0.588±0.066	0.604±0.069	-2.6±4.8	12.585	**0.001***	0.463	0.500	1.070	0.306
Female	0.453±0.111	0.501±0.091	**-9.9±12.7†**	0.516±0.067	0.522±0.070	-1.2±4.2	12.972	**0.001***	2.530	0.118	7.785	**0.008***
All	0.556±0.127	0.565±0.105	**-7.4±10.9†**	0.552±0.075	0.563±0.080	**-1.9±4.5†**	25.198	**<0.001***	0.311	0.578	7.607	**0.007***
1/3												
Male	0.727±0.093	0.745±0.082	**-1.9±14.4**	0.727±0.059	0.746±0.069	**-2.6±3.0†**	3.614	**0.063**	<0.001	0.985	0.003	0.955
Female	0.550±0.121	0.603±0.091	**-9.0±13.3†**	0.620±0.068	0.621±0.070	-0.1±3.1	7.475	**0.009***	2.624	0.112	6.995	**0.011***
All	0.640±0.139	0.675±0.112	**-5.4±14.2†**	0.673±0.083	0.684±0.094	**-1.4±3.3†**	10.2424	**0.002***	0.807	0.371	3.078	0.083
Total forearm												
Male	0.574±0.089	0.604±0.068	**-5.1±8.1†**	0.575±0.063	0.584±0.065	**-1.6±2.7†**	11.331	**0.002***	0.2314	0.646	3.117	0.084
Female	0.435±0.103	0.483±0.084	**-10.3±12.5†**	0.492±0.059	0.497±0.060	-1.1±3.5	14.786	**<0.001***	2.185	0.146	9.564	**0.003***
All	0.506±0.111	0.544±0.097	**-7.7±10.7†**	0.533±0.073	0.541±0.076	**-1.3±3.1†**	25.739	**<0.001***	0.345	0.558	12.072	**0.001***

Significant side × group interaction effect was found in ultradistal, mid, and total forearm regions (aBMD = areal bone mineral density).
Data are mean ± SD indicated.
*Statistically significant results (two-way ANOVA with repeated measures, $p<0.05$).
† Statistically significant difference between the two sides (post-hoc paired t-test, $p<0.025$)

3.3 Comparison of Bone Parameters: pQCT

3.3.1 Radius Distal Epiphysis (4% site)

The pQCT images of the radius distal epiphysis of a representative stroke subject are shown in Figure 7A & 7B. When compared with the non-paretic side (Fig. 7B), it was clear that the paretic side (Fig. 7A) had less trabecular bone surrounded by a thinner cortical shell. Such a pronounced side-to-side difference in bone macrostructure was not observed in the control subject (Fig. 7C & 7D).

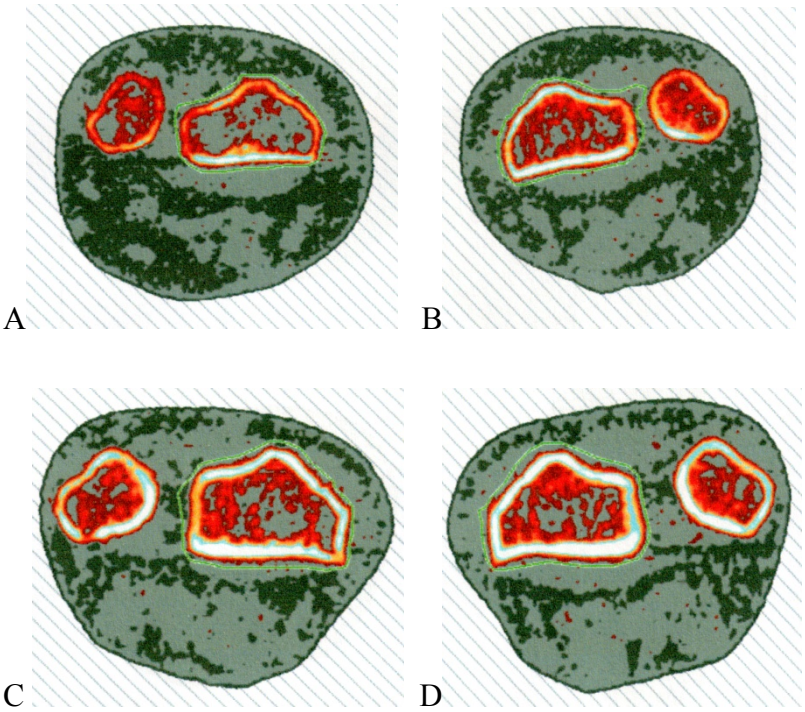

Figure 7. Difference in macrostructure of the distal radius epiphysis between a stroke subject and a control subject.
The figure shows the pQCT images of the paretic side (A) and non-paretic side (B) of a 61-year-old female stroke patient, and those of the non-dominant side (C) and dominant side (D) of a 54-year-old control female subject.

In the primary analysis of group data, there was a significant main effect of side and side × group interaction effect for total BMC, total vBMD, trabecular vBMD, and CBSI ($p<0.05$), but not total area ($p>0.4$) (Table 8). Post-hoc analysis revealed that in the stroke group, the paretic side had significantly lower total BMC ($p<0.001$), total vBMD ($p<0.001$) and trabecular vBMD ($p<0.001$) than the non-paretic side by 9.8%, 10.5% and 11.4%, respectively. In contrast, the side-to-side difference in these parameters was not statistically significant in the control group (0.5-3.0%; $p>0.2$). Moreover, no significant difference in total area was identified between the two sides in both groups of subjects. Nevertheless, the compromised total vBMD led to a significantly lower CBSI on the paretic side than the non-paretic side by 17.8% in the stroke group ($p<0.001$). The corresponding side-to-side difference in p-SSI was much less remarkable in the control group, at only 4.1% ($p=0.126$) (Table 8).

For the most part, the male and female stroke subjects yielded similar results. Regardless of gender, the mean total BMC, total vBMD, trabecular vBMD and CBSI values on the paretic side were all significantly lower than their corresponding values on the non-paretic side among stroke subjects ($p<0.025$) while none of the pQCT variables demonstrated any significant difference between the two sides in the control group ($p>0.1$)(Table 8).

Table 8. Comparison of pQCT parameters: 4% site

	Stroke group			Control group			Main effect: Side		Main effect: Group		Side × group interaction effect	
	Paretic	**Non-paretic**	**%Δ**	**Non-dominant**	**Dominant**	**%Δ**	**F**	**p**	**F**	**p**	**F**	**p**
Total BMC (mg/mm)												
Male	116.2±25.4	125.6±19.8	**-7.8±12.8†**	116.1±24.3	118.3±21.6	-2.1±7.5	8.203	**0.006***	0.321	0.573	3.139	0.083
Female	71.0±20.2	81.5±18.2	**-11.9±19.0†**	81.3±11.4	84.9±12.7	-3.9±7.2	11.543	**0.001***	2.123	0.152	2.689	0.108
All	94.0±32.2	103.9±29.1	**-9.8±16.2†**	98.7±25.7	101.6±24.3	-3.0±7.3	20.001	**<0.001***	0.043	0.837	5.910	**0.017***
Total area (mm^2)												
Male	313.9±43.3	317.6±41.1	-3.1±18.1	320.5±54.3	322.8±65.1	-1.7±9.5	0.462	0.500	0.183	0.671	0.023	0.879
Female	269.8±42.5	264.5±47.8	3.1±15.3	248.4±37.4	253.9±40.9	-1.7±10.6	<0.001	0.990	1.766	0.190	1.192	0.280
All	292.2±48.0	291.4±51.6	-0.1±17.0	284.5±58.7	288.4±64.0	-1.7±9.9	0.228	0.634	0.242	0.624	0.489	0.486
Total vBMD (mg/cm^3)												
Male	372.9±79.2	398.1±59.4	**-6.7±13.5†**	365.4±65.9	372.0±58.9	-1.7±9.5	5.143	**0.028***	0.788	0.379	1.743	0.193
Female	268.8±85.8	313.3±71.4	**-14.4±20.2†**	332.3±58.0	340.5±60.9	-1.7±10.6	10.492	**0.002***	4.985	**0.030***	4.977	**0.030***
All	321.7±97.2	356.3±77.9	**-10.5±17.4†**	348.9±63.4	356.3±61.1	-1.7±9.9	15.344	**<0.001***	0.708	0.402	6.431	**0.013***
Trabecular vBMD (mg/cm^3)												
Male	199.8±42.0	215.7±31.0	**-7.9±11.0†**	208.8±27.7	211.6±27.9	-1.2±5.8	10.321	**0.002***	0.063	0.802	4.993	**0.030***
Female	149.1±38.9	173.7±28.9	**-15.1±16.1†**	183.4±28.0	180.9±33.9	2.2±7.7	10.860	**0.002***	4.876	**0.032***	16.237	**<0.001***
All	174.9±47.6	195.0±36.5	**-11.4±14.1†**	196.1±30.3	196.3±34.3	0.5±6.9	20.725	**<0.001***	1.966	0.164	19.953	**<0.001***
CBSI (g^2/cm^4)												
Male	0.45±0.17	0.50±0.13	**-12.4±22.5†**	0.43±0.14	0.44±0.12	-3.2±15.2	5.870	**0.019***	2.771	0.103	2.771	0.103
Female	0.21±0.12	0.26±0.10	**-23.4±35.2†**	0.27±0.07	0.29±0.08	-5.1±16.0	12.928	**0.001***	3.072	0.086	3.170	0.081
All	0.33±0.19	0.39±0.17	**-17.8±29.7†**	0.35±0.14	0.37±0.13	-4.1±15.4	17.142	**<0.001***	0.007	0.935	5.868	**0.017***

Significant side × group interaction was found in total BMC, total vBMD, trabecular vBMD and CBSI (total BMC = total bone mineral content; total vBMD = total volumetric bone mineral density; trabecular vBMD = trabecular volumetric bone mineral density; CBSI = compute a compressive bone strength index).
Data are mean ± SD indicated
%Δ percent side-to-side difference
*Statistically significant results (two-way ANOVA with repeated measures, $p<0.05$)
† Statistically significant difference between the two sides (post-hoc paired t-test, $p<0.025$);

3.3.2 Radius Diaphysis (33% site)

Figure 8A & 8B show the pQCT images of the radius diaphysis of a representative stroke subject. In this predominantly cortical bone site, the paretic side (Fig. 8A) demonstrated a substantially thinner cortical bone shell and a larger marrow cavity area when compared with the non-paretic side (Fig. 8B). The control subject (Fig. 8C & 8D), on the other hand, showed no such difference between the two sides.

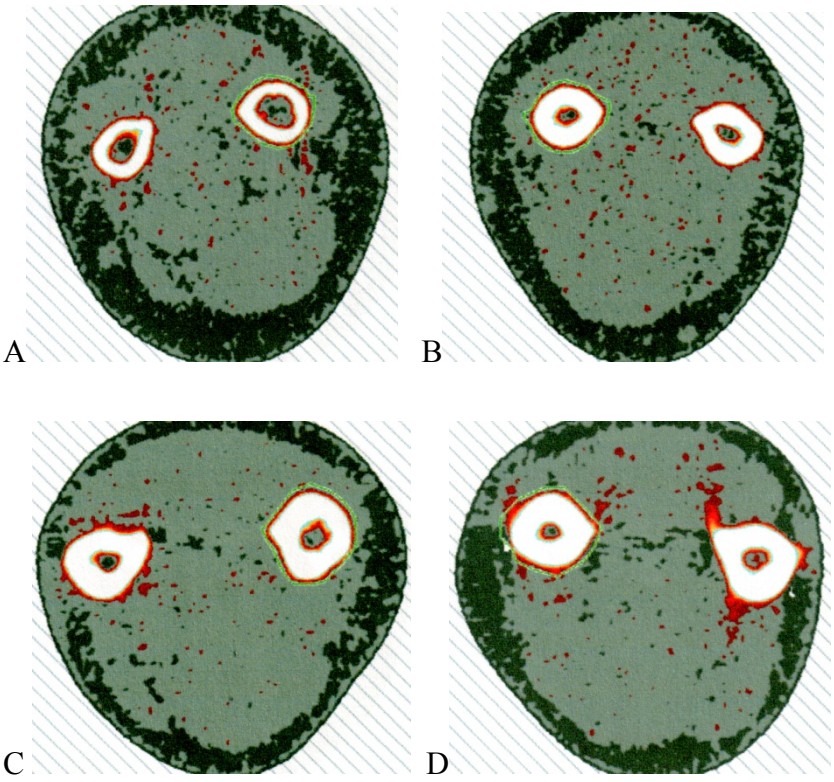

Figure 8. Difference in macrostructure of the radius diaphysis between a stroke subject and a control subject.
The figure shows the pQCT images of the paretic side (A) and non-paretic side (B) of a 61-year-old female stroke patient, and those of the non-dominant side (C) and dominant side (D) of a 54-year-old control female subject.

When the data of all subjects were analyzed in the primary analysis (Table 9), ANOVA revealed a significant main effect of side for cortical BMC ($p<0.001$), cortical vBMD ($p=0.002$), cortical area ($p<0.001$), cortical thickness ($p<0.001$), marrow cavity area ($p=0.025$), and p-SSI ($p=0.032$), but not total area ($p= 0.746$). There was also a significant side × group interaction effect on cortical BMC ($p=0.026$), cortical vBMD ($p=0.001$), and p-SSI ($p=0.021$). The significance level of the side × group interaction effect for cortical area ($p=0.058$) and cortical thickness ($p=0.060$) was marginal.

Post-hoc paired t-tests showed that in the stroke group, the cortical BMC, cortical vBMD, cortical area, and cortical thickness in the paretic radius diaphysis were significantly lower than the corresponding values on the non-paretic side. The marrow cavity on the paretic side, in contrast, was significantly greater than the non-paretic side (<0.001), while the total area did not demonstrate any significant side-to-side difference ($p=0.866$). As a result of the differences in densitometric and geometric parameters, the p-SSI on the paretic side was also significantly lower than that on the non-paretic side by 5.9% ($p<0.001$) (Table 9). In the control group, the difference in pQCT parameters between the two sides was largely unremarkable. There was no significant difference in any of the parameters except the cortical area on the dominant side (0.3%, $p=0.028$).

In the secondary analysis, the female stroke group demonstrated significantly lower values in cortical BMC ($p<0.001$), cortical vBMD ($p<0.001$), cortical area ($p<0.001$), cortical thickness ($p<0.001$) and p-SSI ($p<0.001$), but greater marrow cavity area ($p=0.001$) (Table 9). On the other hand, although the male stroke subjects demonstrated a similar trend, the side-to-side difference in the same bone parameters was less pronounced than their female counterparts, with significant findings only in cortical BMC ($p=0.019$), cortical vBMD ($p=0.013$), and p-SSI

(p=0.017). Regardless of gender, the total area demonstrated no significant side-to-side difference in the stroke group (p>0.80). While many pQCT parameters at the radius diaphysis showed significant side-to-side differences in the stroke group, the only significant finding in the control group was a slightly greater cortical area on the dominant side among men (0.9%, p=0.019).

Table 9. Comparison of pQCT parameters: 33% site

	Stroke group			Control group			Main effect: Side		Main effect: Group		Side × group interaction effect	
	Paretic	Non-paretic	%Δ	Non-dominant	Dominant	%Δ	F	p	F	p	F	p
Cortical BMC (mg/mm)												
Male	106.0±18.7	111.2±14.1	**-4.8±11.2†**	103.0±16.7	106.7±15.2	**-3.6±4.7†**	8.266	**0.006***	0.656	0.422	0.245	0.623
Female	65.8±20.5	73.7±16.1	**-11.7±15.9†**	74.9±9.3	75.1±9.9	-0.0±5.3	9.376	**0.004***	1.299	0.260	8.534	**0.005***
All	86.2±28.1	92.7±24.1	**-8.2±14.1†**	88.9±19.5	90.9±20.4	-1.8±5.3	17.233	**<0.001***	0.008	0.929	5.093	**0.026***
Cortical vBMD (mg/cm^2)												
Male	1195.4±36.0	1208.6±28.2	**-1.1±2.4†**	1196.6±30.8	1197.4±24.8	-0.1±1.5	3.362	0.073	0.355	0.554	2.612	0.113
Female	1147.9±82.0	1180.6±67.2	**-2.9±4.4†**	1189.4±43.2	1185.9±45.3	0.3±1.2	6.831	**0.012***	1.509	0.225	10.522	**0.002***
All	1172.0±66.9	1194.8±52.8	**-2.0±3.4†**	1193.0±37.1	1162.2±188.2	0.1±1.4	9.681	**0.002***	0.683	0.411	12.263	**0.001***
Total area (mm^2)												
Male	133.1±13.3	133.4±12.4	-0.2±6.6	159.8±148.1	163.8±140.0	-4.8±14.0	1.520	0.224	1.304	0.259	1.118	0.296
Female	99.8±12.0	99.8±13.6	0.2±3.9	123.5±111.2	117.2±83.8	1.3±8.4	1.591	0.213	1.403	0.242	1.577	0.215
All	116.7±21.0	116.9±21.3	0.0±5.4	141.6±130.3	140.5±116.0	-1.8±11.8	0.106	0.746	2.428	0.122	0.178	0.674
Cortical area (mm^2)												
Male	88.3±14.1	91.9±11.0	-4.0±10.1	86.3±13.6	89.0±11.9	**-0.9±5.7†**	6.993	**0.011***	0.474	0.495	0.114	0.738
Female	56.5±14.9	62.0±11.5	**-11.2±15.5†**	62.9±6.9	63.3±7.2	0.4±5.0	8.862	**0.005***	1.301	0.260	6.834	**0.012***
All	72.7±21.5	77.2±18.8	**-7.6±13.5†**	74.6±15.9	76.1±16.3	-0.3±5.3	15.214	**<0.001***	0.012	0.913	3.680	0.058
Marrow cavity area (mm^2)												
Male	44.8±11.2	41.5±9.2	8.1±22.3	73.49±140.7	74.8±133.3	1.0±14.3	0.391	0.534	1.710	0.197	2.024	0.161
Female	43.3±15.6	37.8±13.4	**17.6±22.6†**	60.6±111.6	54.0±84.0	3.0±15.8	5.188	**0.027***	0.044	0.835	0.044	0.835
All	44.0±13.4	39.7±11.5	**12.6±22.8†**	67.1±125.2	64.4±110.2	2.0±14.3	5.158	**0.025***	2.654	0.107	0.291	0.591
Cortical thickness (mm)												
Male	2.77±0.45	2.88±0.36	-3.9±11.4	2.67±0.29	2.79±0.25	-3.8±7.5	6.487	**0.014***	<0.001	0.990	<0.001	0.990
Female	1.97±0.59	2.21±0.45	**-14.2±18.9†**	2.28±0.29	2.29±0.32	0.0±5.1	8.874	**0.005***	2.129	0.151	8.025	**0.007***
All	2.38±0.65	2.55±0.53	**-9.0±16.3†**	2.48±0.35	2.54±0.38	-1.9±6.6	14.673	**<0.001***	0.171	0.680	3.635	0.060
p-SSI (mm^3)												
Male	285.3±54.1	298.6±46.7	**-4.7±10.1†**	288.0±78.1	291.0±70.7	-1.3±7.7	3.709	0.060	0.020	0.888	1.478	0.230
Female	166.4±43.1	177.4±36.6	**-7.2±13.1†**	179.8±22.7	175.9±27.9	3.2±11.6	1.108	0.298	0.334	0.566	4.899	**0.032***
All	226.8±77.1	239.0±73.9	**-5.9±11.6†**	233.9±78.9	233.5±78.9	1.0±10.0	4.734	**0.032***	0.002	0.961	5.494	**0.021***

Significant side × group interaction effect was found in cortical BMC, cortical vBMD, and p-SSI (Cortical BMC: cortical bone mineral content; Cortical vBMD: cortical volumetric bone mineral density; p-SSI: polar stress-strain index

Data are mean ± SD indicated

%Δ percent side-to-side difference

*Statistically significant results (two-way ANOVA with repeated measures, $p<0.05$)

† Statistically significant difference between the two sides (post-hoc paired t-test, $p<0.025$)

3.5 Comparison of Neuromuscular Parameters

The hand grip strength on the paretic side was significantly lower by 44.5% when compared with the non-paretic side, indicating substantial paresis on the affected side (Table 10). In contrast, although the control group also demonstrated a significant difference in hand-grip strength between the dominant and non-dominant sides, the magnitude of the difference was significantly smaller (6.9%). The mean FMA score was 46 out of 66, indicating moderate impairment (Table 11). The majority of the subjects did not have severe spasticity in the affected upper extremity. Twelve stroke subjects (18.5%) had a MAS score of ≥2, with only 2 of these subjects (3.1%) obtaining a MAS score of ≥3. Given the prominent sensorimotor impairment, it was not surprising that the mean MAL score was only at 3.2, indicating moderate disuse of the affected upper extremity. The secondary analysis revealed that the female stroke subjects had a significantly lower FMA score (p=0.002) but higher MAS score (p=0.020) than the male stroke subjects.

Table 10. Comparison of grip strength data

Parameter	Stroke group (n=65)			Control group (n=34)		
	Paretic	Non-paretic	p	Non-dominant	Dominant	p
Grip strength (kg)						
Male	20.3±11.1	30.8±6.7	**<0.001***	29.3±6.2	32.2±7.0	**<0.001***
Female	6.4±7.3	17.5±5.1	**<0.001***	21.6±5.1	20.8±4.0	0.138
All	13.45±11.65	24.25±8.97	**<0.001***	25.1±6.7	26.9±8.1	**<0.001***

The grip strength on the paretic side was significantly lower than the non-paretic side in the stroke group. The side-to-side difference in the control group was much smaller.
Data are mean ± SD.
*Statistically significant (paired t-test, p<0.025).

Table 11. Other neuromuscular impairments in the stroke group

Parameter	All	Male	Female	p
MAS				
(0/1/1+/2/3/4), n	25/16/12/10/2/0	17/8/4/3/1/0	8/8/8/7/1/0	---
Median±IQR	1.0±1.5	0.0±1.25	1.25±1.63	**0.020***
FMA (0-66)	46.00±20.31	53.7±15.9	38.1±21.5	**0.002†**
MAL (0-5)	3.17±1.75	3.57±1.52	2.75±1.89	0.059

Overall, the subjects in the stroke group had moderate motor impairment and disuse in the paretic upper extremity. Only a small proportion of subjects had severe spasticity (MAS = Modified Ashworth Scale; FMA = Fugl-Meyer motor assessment; MAL = Motor Activity Log).
Data are mean ± SD unless otherwise indicated.
*Statistically significant (Mann Whitney U test, $p<0.05$
† Statistically significant (independent t- test, $p<0.05$);

3.6 Comparison of Cardiovascular Parameters

Regarding the various cardiovascular-related measures, the 6MWT distance recorded in the stroke group was significantly shorter than that in controls, indicating impaired walking endurance ($p<0.001$). The mean VO_2 during the 6MWT was also significantly lower in the stroke group than controls ($p<0.001$). There was no significant between-group difference in other cardiovascular measures, except a significantly lower C_2 value in the female stroke group ($p=0.020$) (Table 12).

Separate analyses were performed for those with ischemic stroke and those with hemorrhagic stroke. No significant difference was found in the cardiovascular measures between these two sub-groups of patients after stroke ($p=0.125$-0.937). In addition, their cardiovascular measures were not significantly different from the control group, regardless of whether the stroke was ischemic or hemorrhagic in nature ($p>0.05$).

Table 12. Comparison of cardiovascular parameters

Parameters	Stroke group (n=65)			Control group (n=34)			p
	male	female	all	male	female	all	
C_1 (mL/mmHg × 10)	14.72±4.94	12.58±5.52	13.63±5.30	15.9±45.77	13.52±4.46	14.74±5.23	0.339
C_1(Abnormal/borderline/normal),n	0/6/27	1/10/21	1/16/48	0/1/16	0/3/14	0/4/30	0.164
C_2 (mL/mmHg × 100)	5.95±4.07	3.29±2.06	4.64±3.48	4.98±1.90	4.86±2.40	4.92±2.13	0.676
C_2 (Abnormal/borderline/normal),n	15/5/13	21/6/5	36/11/18	9/5/3	3/5/9	12/10/12	0.918
CI (L/min/m^2)	2.41±0.70	2.61±0.68	2.51±0.69	2.70±1.94	2.42±0.66	2.56±1.44	0.801
SI (mL/beat/m^2)	38.43±11.32	40.20±9.68	39.30±10.50	39.87±25.70	39.55±8.14	39.71±18.77	0.890
6MWT distance (m)	318.39±108.11	214.05±116.76	267.02±123.33	445.06±50.29	446.56±53.09	445.81±50.93	**<0.001***
VO_2 during 6MWT (mL/kg/min)	12.82±3.13	10.67±3.12	11.76±3.28	14.76±4.54	15.22±3.36	14.99±3.94	**<0.001***

Significant difference in 6MWT distance and VO_2 rate during 6MWT was found between the stroke and control groups (C_1 = large artery elasticity index; C_2 = small artery elasticity index; CI = cardiac index; SI = stroke index; 6MWT = 6 minute walking test; VO_2 = oxygen consumption).
Data are mean ± SD
*Statistically significant (independent t-tests, $p<0.025$)

3.7 First line of Correlation Analysis: Association of Absolute Bone Parameters with Neuromuscular and Cardiovascular Function in Stroke Group

Bivariate correlation analyses were first performed to assess the degree of association of DXA-derived aBMD values and pQCT-derived bone strength indices with indicators of muscle and cardiovascular function (Table 13). It was clear that sensorimotor impairments were highly associated with the various key bone parameters. FMA, grip strength, and MAL had significant, positive correlations with aBMD in all the measured forearm regions (i.e., ultradistal, mid-, 1/3, and total forearm), and with the bone strength indices measured at the 4% (i.e., CBSI) and 33% radius sites (i.e., p-SSI), indicating that those individuals with better motor recovery and muscle strength and more frequent use of the affected upper limb tended to have higher aBMD and bone strength index values. On the other hand, negative associations of the bone parameters with MAS score were identified, demonstrating that more severe spasticity was related to lower aBMD and bone strength indices.

Among the various cardiovascular parameters, C_1 and C_2 showed a significant, positive correlation with all the key bone parameters, whereas CI, SI, and VO_2 rate during the 6MWT had no significant relationship with any of the bone parameters.

Table 13. Association of absolute values of bone parameters with neuromuscular and cardiovascular function

	Areal bone mineral density (DXA)				Bone strength index (pQCT)	
Parameters	Ultradistal forearm	Mid-forearm	1/3-forearm	Total forearm	CBSI	p-SSI
MAS	**-0.361***	**-0.346***	**-0.344***	**-0.362***	**-0.412***	**-0.331***
MAL	**0.340***	**0.331***	**0.291***	**0.333***	**0.342***	**0.258***
FMA	**0.339***	**0.364***	**0.378***	**0.375***	**0.462***	**0.377***
Grip strength	**0.531***	**0.592***	**0.613***	**0.607***	**0.689***	**0.712***
CI	0.098	0.075	-0.023	0.052	0.064	-0.032
SI	0.092	0.184	0.171	0.163	0.059	0.119
VO_2 during 6MWT	0.081	0.102	0.140	0.109	0.112	0.214
C_1	**0.289***	**0.365***	**0.401***	**0.371***	**0.332***	**0.417***
C_2	**0.393***	**0.505***	**0.476***	**0.496***	**0.517***	**0.497***

Areal bone mineral density of different forearm subregions and bone strength indices of the radius were significantly correlated to all neuromuscular variables measured, C_1 and C_2 (MAS = Modified Ashworth Scale; MAL = Motor Activity Log; FMA = Fugl-Meyer motor assessment; CI = cardiac index; SI = stroke index; VO_2 = oxygen consumption; 6MWT = 6 minute walking test; C_1 = large artery elasticity index; C_2 = small artery elasticity index). *Statistically significant (Pearson's r or Spearman's rho, $p<0.05$)

3.8 First Set of Regression Analyses: Predicting Absolute Forearm aBMD and Radius Bone Strength Index Values on the Paretic Side

Multiple regression analyses were performed to identify the determinants of aBMD of the four forearm regions (i.e., ultradistal, mid, 1/3, and total), and bone strength indices in the radius epiphysis (i.e., CBSI), and diaphysis (i.e., p-SSI) on the paretic side. To check for multicollinearity, bivariate correlation analyses were first performed to determine the degree of association between the stroke impairment variables. It was found that MAL, MAS, FMA, grip strength were highly correlated with each other ($r > 0.5$) (Appendix 9). Therefore, separate

regression analyses were used to identify the relative contributions of individual neuromuscular impairments to the dependent variable. In all regression analyses, relevant variables such as age, sex, BMI, and post-stroke duration were forced into the regression model first, followed by the stroke impairments that showed significant bivariate correlations with the dependent variable

3.8.1 Determinants of Ultradistal Forearm aBMD

Regression analyses were performed to identify the significant determinants of ultradistal aBMD of the forearm on the paretic side (Appendix 10). After accounting for age, sex, BMI, and post-stroke duration, MAS (model 1), MAL (model 3), and grip strength (model 4) remained independently associated with ultradistal aBMD. These models explained a total of 51.6-54.8% of the variance in ultradistal aBMD. There was a tendency for the FMA score (model 2) to be associated with ultradistal forearm aBMD ($p=0.099$). Although C_1 and C_2 had significant association with ultradistal forearm aBMD in bivariate correlation analysis, their effects were diminished in multivariate analysis ($p>0.50$).

3.8.2 Determinants of Mid-forearm aBMD

Regressions analyses were performed to identify the determinants of aBMD of the mid-forearm region on the paretic side (Appendix 11). After accounting for age, sex, BMI, and years after stroke onset, MAS (model 1) and grip strength (model 4) remained independently associated with mid-region aBMD ($p<0.05$) (Appendix 11). The total variance accounted for in these models varied from 64.3% to 66.5%. On the other hand, there was a trend for the FMA score (model 2) ($p=0.059$) and MAL score (model 3) ($p=0.073$) to be associated with mid-

forearm aBMD, after adjusting for other relevant factors. C_1 and C_2 were not independently associated with the mid-forearm aBMD ($p>0.20$).

3.8.3 Determinants of 1/3 Forearm aBMD

Appendix 12 shows the results of the regression analyses for predicting aBMD of the 1/3 forearm region on the paretic side. After accounting for age, sex, BMI, and years after stroke onset, MAS (model 1) and grip strength (model 4) remained independently associated with aBMD of the 1/3 forearm region, accounting for a total of 63.1-64.8% of the variance ($p<0.05$). There was also a trend for the FMA score to be associated with aBMD of the 1/3 forearm region (model 2) ($p=0.082$). MAL score, on the other hand, was not significantly correlated with aBMD of the 1/3 forearm region, after adjusting for other relevant factors (model 3) ($p=0.318$). Similar to the other two sub-regions of the forearm, C_1 and C_2 were not independently associated with aBMD of the 1/3 region of the paretic forearm.

3.8.4 Determinants of Total Forearm aBMD

Multiple regression analyses showed that MAS (model 1) and grip strength (model 4) remained independently associated with total BMD of paretic forearm after accounting for the effects of age, sex, BMI and post-stroke duration, accounting for a total of 65.2-67.5% of the variance ($p<0.05$) (Appendix 13). There was also a tendency for a positive association of the total forearm aBMD with FMA (model 2) ($p=0.054$) and MAL scores (model 3) ($p=0.077$). No significant association was identified between total forearm aBMD and C_1 and C_2 in multivariate analysis.

3.8.5 Determinants of CBSI at the Distal Radius Epiphysis

After accounting for age, sex, BMI, and years after stroke onset, MAS (Appendix 14, model 1), FMS (model 2), and grip strength (model 4) remained independently associated with CBSI at 4% site of radius. These models explained 64.3-67.3% of the variance in CBSI ($p<0.05$) (Appendix 14). The associations of CBSI with MAL score (model 3), C_1, and C_2 were no longer significant after adjusting for the effects of other relevant factors ($p>0.05$).

3.8.6 Determinants of p-SSI of the Radius Diaphysis

After accounting for age, sex, BMI, and years after stroke onset, grip strength was independently associated with p-SSI at the 33% site of radius (model 4). In addition, C_1 was independently associated with p-SSI in regression model 2 ($p<0.05$). There was also a tendency for the C_1 to be associated with p-SSI in the other three regression models with marginal significance level ($p=0.055-0.058$). MAS, FMS, and MAL scores, on the other hand, were not significantly associated with p-SSI after accounting for the relevant factors ($p>0.05$) (Appendix 15). These models accounted for 73.5-77.8% of the variance in p-SSI.

3.9 Second Line of Correlation Analysis: Association of Percent Side-to-side Difference of Bone Parameters with Neuromuscular and Cardiovascular Function in Stroke Group

To further explore the association of stroke impairments with upper extremity bone status, a second line of correlation analyses was conducted. The dependent variable was now the percent side-to-side difference values in bone parameters, rather than the absolute values

obtained on the paretic side (Table 14). Most of the measured sensorimotor impairments were highly associated with the various key bone parameters. Specifically, FMA, percent side-to-side difference in grip strength, and MAL had significant, positive correlations with percent side-to-side difference in DXA-derived aBMD values, and pQCT-derived bone strength indices. In addition, negative associations between percent side-to-side difference in the various bone parameters and MAS were found, reflecting that more severe spasticity was related to greater side-to-side difference of aBMD and bone strength indices.

Table 14. Correlation between percent side-to-side difference in bone parameters and neuromuscular and cardiovascular function

	Percent side-to-side difference (%Δ)					
	Areal bone mineral density (DXA)				**Bone strength index (pQCT)**	
Parameter	**Ultradistal forearm**	**Mid-forearm**	**1/3-forearm**	**Total forearm**	**CBSI**	**p-SSI**
MAS	**-0.411***	**-0.583***	**-0.341***	**-0.584***	**-0.576***	**-0.180**
MAL	**0.410***	**0.497***	**0.273***	**0.512***	**0.523***	**0.353***
FMA	**0.353***	**0.553***	**0.392***	**0.566***	**0.532***	**0.286***
%Δ grip strength	**0.393***	**0.639***	**0.461***	**0.663***	**0.634***	**0.382***
CI	0.152	0.056	0.009	0.056	-0.005	0.048
SI	-0.044	0.086	0.205	0.080	-0.011	0.112
VO_2	0.060	-0.089	0.147	-0.057	-0.127	0.061
C_1	0.033	0.237	**0.297***	**0.246***	0.188	**0.343***
C_2	0.134	**0.271***	**0.261***	**0.288***	**0.275***	0.228

The percent side-to-side difference in areal bone mineral density of different forearm subregions and bone strength indices of the radius were significantly related to all neuromuscular variables measured (%Δ = Percent side-to-side difference, DXA = dual-energy X-ray absorptiometry ; pQCT = peripheral quantitative computed tomography; CBSI = compressive bone strength index ; p-SSI = polar stress-strain index ; MAS = Modified Ashworth Scale; MAL = Motor Activity Log**;** FMA: Fugl-Meyer motor assessment; CI = cardiac index; SI = stroke index; VO_2 = oxygen consumption; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant (Pearson's r or Spearman's rho, $p<0.05$)

Similar to the results yielded in the first line of correlation analysis, CI, SI, and VO_2 were not significantly related to the percent side-to-side difference in any of the bone parameters of

interest. C_1 showed a significant, positive correlation with percent side-to-side difference of 1/3-forearm aBMD, total forearm aBMD and p-SSI. C_2 showed a significant, positive correlation with percent side-to-side difference of mid-forearm aBMD, 1/3-forearm aBMD, total forearm aBMD, and CBSI.

3.10 Second Set of Regression Analyses: Predicting Percent Side-to-side Difference in Forearm aBMD and Radius Bone Strength Indices on the Paretic Side

Multiple regression analyses were performed to identify the determinants of percent side-to-side difference of aBMD of the four forearm regions (i.e., ultradistal, mid, 1/3, and total), and bone strength indices in the radius epiphysis (i.e., CBSI), and diaphysis (i.e., p-SSI) on the paretic side. To check for multicollinearity, bivariate correlation analyses were performed to determine the degree of association of the percent side-to-side difference of the grip strength with stroke impairment variables. It was found that the percent side-to-side difference of grip strength was highly correlated with other motor impairments (i.e., MAL, MAS, FMA) ($r > 0.5$), but not the cardiovascular indexes ($r < 0.5$) (Appendix 16).

3.10.1 Determinants of Percent Side-to-side Difference of Ultradistal Forearm aBMD

Regression analyses were performed to identify the significant determinants of percent side-to-side difference of ultradistal forearm aBMD (Appendix 17). After accounting for age, sex, BMI, and post-stroke duration, MAL (model 2), FMA (model 3), and percent side-to-side difference in grip strength (model 4) remained independently associated with percent side-to-side

difference in ultradistal forearm aBMD. These models explained a total of 19.0-23.5% of the variance in percent side-to-side difference of ultradistal forearm aBMD. There was a tendency for the MAS group (model 1) to be associated with percent side-to-side difference of ultradistal forearm aBMD ($p=0.067$).

3.10.2 Determinants of Percent Side-to-side Difference in Mid-forearm aBMD

Regressions analyses were performed to identify the determinants of percent side-to-side difference in aBMD of the mid-forearm region (Appendix 18). After accounting for age, sex, BMI, and years after stroke onset, MAS (model 1), FMS score (model 2), MAL score (model 3) and grip strength (model 4) remained independently associated with percent side-to-side difference of mid-region aBMD ($p<0.01$). The total variance accounted for in these models ranged from 36.6% to 51.8%. Among the various neuromuscular impairments, grip strength was the most powerful determinant (model 4), as reflected by the large R^2 change (0.289) and beta values (0.663), compared with the other three regression models. In none of these regression models did C_2 demonstrate significant associations with the percent side-to-side difference of mid-forearm aBMD.

3.10.3 Determinants of Percent Side-to-side Difference in 1/3 Forearm aBMD

Appendix 19 shows the results of the regression analyses for predicting percent side-to-side difference of BMD of the 1/3 forearm region on the paretic side. After accounting for age, sex, BMI, and years after stroke onset, MAS (model 1), FMS (model 2), and percent side-to-side difference in grip strength (model 4) remained independently associated with percent side-to-side difference of aBMD of the 1/3 forearm region, accounting for a total of 30.7 and 37.7% of the

variance ($p<0.05$), respectively. MAL score, on the other hand, was not significantly correlated with percent side-to-side difference of BMD of the 1/3 forearm region, after adjusting for other relevant factors (model 3) ($p=0.510$). Similar to the other two subregions of the forearm, C_1 and C_2 were not independently associated with percent side-to-side difference in aBMD of the 1/3 forearm region.

3.10.4 Determinants of Percent Side-to-side Difference in Total Forearm aBMD

Multiple regression analyses showed that MAS (model 1), FMA score (model 2), MAL score (model 3) and grip strength (model 4) remained independently associated with percent side-to-side difference of total aBMD after accounting for the relevant factors, accounting for a total of 38.6-58.2% of the variance ($p<0.05$) (Appendix 20). Among the four neuromuscular impairments measured, grip strength was the most important determinant of total forearm aBMD, as reflected by the magnitude of its beta value of 0.687), which was greater than that of the MAS, FMA and MAL (0.385-0.492). No significant association was identified between the percent side-to-side difference of total forearm aBMD and C_1 and C_2.

3.10.5 Determinants of Percent Side-to-side Difference in CBSI at the Radius Epiphysis

After accounting for age, sex, BMI, and years after stroke onset, FMA (model 2), MAL score (model 3) and percent side-to-side difference in grip strength (model 4) remained independently associated with the percent side-to-side difference of CBSI at 4% site of radius. Model 4 using grip strength as the predictor variable explained the most variance in percent side-to-side difference of CBSI ($R^2=0.483$, $p<0.05$) (Appendix 21). There was also a tendency for the MAS (model 1) to be associated with percent side-to-side difference of CBSI ($p=0.066$). The

cardiovascular parameter—namely, C_2—was not significantly associated with the percent side-to-side difference of CBSI in multivariate analysis.

3.10.6 Determinants of Percent Side-to-side Difference in p-SSI at the Radius Diaphysis

After accounting for age, sex, BMI, and years after stroke onset, only grip strength was independently associated with percent side-to-side difference in p-SSI at the 33% site of radius (model 3), explaining 32.7% of the variance. FMA, and MAL scores, on the other hand, were not significantly associated with percent side-to-side difference in p-SSI after accounting for the relevant factors ($p>0.05$) (Appendix 22). C_1 was not a significant determinant of the percent side-to-side difference in p-SSI ($p>0.05$).

3.11 Summary of Results on Regression Analyses

Overall, multiple regression analyses showed that aside from basic demographic factors such as age and sex, the neuromuscular impairments played a more predominant role in determining the paretic forearm aBMD and bone strength indices than the cardiovascular measures. In particular, grip strength stood out as the most powerful determinant of these bone parameters. The results of the regression analyses for DXA-derived aBMD variables are summarized in Table 15 and Table 16, respectively.

Table 15. The significant determinants identified in regression analyses: DXA

	Dependent variable							
	Paretic forearm aBMD (g/cm^2)				%Δ forearm aBMD			
	Ultradistal	**mid**	**1/3**	**Total**	**Ultradistal**	**mid**	**1/3**	**Total**
Predictor variables								
Demographics								
Age	√	√	√	√		√	√	√
Sex	√	√	√	√				
BMI								
Post-stroke duration								
Neuromuscular								
MAS	√	√	√	√		√	√	√
FMA					√	√	√	√
MAL	√				√	√		√
Grip strength	√	√	√	√	NA	NA	NA	NA
%ΔGrip strength	NA	NA	NA	NA	√	√	√	√
Cardiovascular								
CI								
SI								
C_1								
C_2								
VO_2								

Grip strength was identified as a significant determinant of the dependent variables in all regression models (%Δ Percent side-to-side difference = aBMD: areal bone mineral density; MAS = Modified Ashworth Scale; MAL = motor activity log**;** FMA = Fugl-Meyer motor assessment; CI = cardiac index; SI = stroke index; C_1 = large artery elasticity index; C_2 = small artery elasticity index; VO_2 = oxygen consumption; NA = Not applicable; √ = Significant determinant of the dependent variable).

Table 16. The significant determinants identified in regression analyses: pQCT

	Dependent variable			
	Radius epiphysis (4% site)		Radius diaphysis (33% site)	
	Paretic side CBSI (in g^2/cm^4)	%Δ Paretic side CBSI	Paretic side p-SSI (in mm^3)	%Δ Paretic side p-SSI
Predictor variables				
Demographics				
Age	√		√	
Sex	√		√	
BMI				
Post-stroke duration		√		√
Neuromuscular				
MAS	√			
FMA	√		√	
MAL			√	
Grip strength	√	√	NA	NA
%ΔGrip strength	NA	NA	√	√
Cardiovascular				
CI				
SI				
C_1			√	
C_2				
VO_2				

Grip strength was identified as a significant determinant of the dependent variables in all regression models (%Δ Percent side-to-side difference = aBMD: areal bone mineral density; MAS = Modified Ashworth Scale; MAL = motor activity log**;** FMA = Fugl-Meyer motor assessment; CI = cardiac index; SI = stroke index; C_1 = large artery elasticity index; C_2 = small artery elasticity index; VO_2 = oxygen consumption; NA = Not applicable; √ = Significant determinant of the dependent variable).

CHAPTER 4

DISCUSSION

4.1 Pronounced Side-to-side Difference in Areal Bone Mineral Density Measured by DXA

The degree of bone loss in the paretic upper extremity can be estimated by comparing the bone mineral levels between the paretic and non-paretic sides. In this study, we found a significant side-to-side difference in aBMD (5%-10%) in all measured regions of the forearm in the stroke group (Table 7). Our results thus extend the findings from previous cross-sectional studies, which showed that individuals with chronic stroke had compromised aBMD in other skeletal sites of the hemiparetic upper extremity, including the total arm (Hamdy et al., 1993; Iversen et al., 1989; Pang et al., 2005a), and proximal humerus (Jørgensen et al., 2000). The magnitude of the side-to-side differences in aBMD values reported in this study is also comparable with what was previously reported in the chronic stroke population. For example, Pang et al. (2005a) reported a 4.5% side-to-side difference in total arm aBMD in a sample of 56 older individuals with chronic stroke (mean post-stroke duration = 197 weeks). Using dual photon absorptiometry, Hamdy et al. (1993) showed that an 8% side-to-side difference existed in the total arm aBMD in a sample of 30 subjects with chronic stroke (mean post-stroke duration = 484 weeks).

Few studies have employed a prospective design to examine the aBMD changes in the paretic upper extremity among patients in the chronic stage of stroke recovery (Jørgensen& Jacobsen, 2001; Ramnemark et al., 1999a, 1999b). The extent of changes varied greatly, depending on the skeletal sites measured and subject characteristics. For example, it has been demonstrated that the one-year reduction of aBMD in the humerus (17.4%) is much higher than that in the total arm (7.6%) in the same group of stroke subjects (Ramnemark et al., 1999b). Only one study has examined the aBMD of the ultradistal radius in the chronic stage of stroke recovery. Ramnemark et al. (1999b) showed that the ultradistal radius region sustained a

significant 8.6% decline in aBMD whereas the non-paretic side had a significant 5.5% increase in the same variable within the first year following the onset of stroke. Such differential changes in the paretic and non-paretic sides resulted in a 15.7% side-to-side difference in aBMD when measured 12 months post-stroke. In comparison, the side-to-side difference in the ultradistal region of the forearm in our stroke subjects was less (9.6%), despite the fact that our subjects were more chronic (mean post-stroke duration = 47.8 months). The difference in results was probably due to the difference in subject characteristics. First, their subjects were relatively older (mean age = 77) than ours (mean age = 60). The difference in severity of hemiparesis may also explain the different results. The subjects in Ramnemark et al. (1999b) were more severely impaired (Morticity Index score = 45/100) than ours (mean FMA score = 46/66).

The side-to-side difference in bone mineral levels observed in our stroke group could not be explained by hand dominance. First, our analysis showed that there was no significant relationship between hand dominance and the side of paresis. Second, the side-to-side difference in aBMD in the stroke group (5-10%) was much greater than that in the control group (0-2%). The hand dominance-related difference in aBMD observed in our control group was consistent with the results found by Taaffe et al. (1994), who reported a significant but small difference in total arm aBMD (1.0%) between the dominant and non-dominant sides in elderly women. Overall, the results of this study have clearly demonstrated the deleterious effect of stroke on aBMD in the hemiparetic forearm.

4.2 Pronounced Side-to-side Difference in Volumetric Bone Mineral Density, Bone Geometry, and Bone Strength Indices of the Radius Measured by pQCT

Along with bone density, bone geometry is an important determinant of bone strength (Burr & Turner 2003; Frost, 2003). Using pQCT, this study provides insight into the influence of stroke on both bone density and geometry at the radius epiphysis and diaphysis.

4.2.1 Radius Distal Epiphysis

At the 4% site of the radius (distal epiphysis), the paretic side had significantly lower total vBMD (10.5%) and trabecular vBMD (11.4%) than the non-paretic side. Only two studies have used pQCT to examine vBMD at the radius distal epiphysis (Ashe et al., 2006; Lazoura et al., 2008) and in general, their results are consistent with what is reported in this study. In a small sample of 15 chronic stroke patients, Ashe et al. (2006) reported a significantly lower total vBMD (15%) at the 4% site of the radius on the paretic side. Lazoura et al. (2008) also showed that at one-year post-stroke, the trabecular vBMD at the 4% site of the radius on the paretic side was 14% lower than that on the non-paretic side. The most interesting finding of the study, however, is that no significant side-to-side difference in total area was found in the stroke group, just as in the control group. A similar finding was also reported by Ashe et al. (2006), based on data from 13 patients after chronic stroke. Taken together, the results may indicate that the overall bone size is relatively preserved on the paretic side, despite the decline in vBMD. This observation is distinctly different from the bone changes associated with aging, which are characterized by the reduction of vBMD with a concomitant increase in total area (Ahlborg et al., 2003; Riggs et al., 2004). For example, in a population-based study of age differences in bone properties using pQCT, Riggs et al. (2004) found that although the total vBMD at the distal radius was reduced by 28-30% between 20 and 90 years of age, the total area increased by

approximately 15%. Since the CBSI is proportional to the load-bearing area of the bone, the increase in total area would increase bone strength against compressive forces. It is thought that the increase in total area is a compensatory mechanism to offset the detrimental effect of vBMD decline in bone strength during the aging process (Riggs et al., 2004). This results obtained in the distal radius epiphysis in our stroke patients seemed to be distinct from that observed in aging.

Despite the preservation of the total area on the paretic side, the CBSI remained significantly lower than on the non-paretic side by 17.8%, owing to the effects of compromised total vBMD. The lower CBSI denotes reduced ability of the bone segment to resist compressive loads, which may, in turn, increase the risk of fracture. Only one study has examined the bone strength index at the radius distal epiphysis in stroke patients, although geometric properties were not specifically reported. Based on a sample of 67 patients after stroke, Lazoura et al. (2008) showed that p-SSI was reduced by 28.6% and 19.2% within the first year post-stroke in male and female subjects, respectively. However, the non-paretic side also sustained a corresponding reduction in p-SSI of 11.5% and 6.1% in male and female subjects, respectively. Such changes resulted in a side-to-side difference in the p-SSI of 26.5% and 28.2% in male and female stroke patients, respectively, at 12 months post-stroke. The extent of side-to-side difference in the bone strength index at the radius epiphysis reported in their study seemed to be greater than that observed in this study. However, it is difficult to compare our findings with theirs. Firstly, details on upper extremity function in their subjects were not documented, whereas a thorough examination of neuromuscular and cardiovascular function was included in our study protocol. Secondly, a different bone strength index was used. The CBSI was used in this study, whereas the p-SSI (a torsional bone strength index) was used in Lazoura et al. (2008). It is more appropriate to use the CBSI rather than the p-SSI for the epiphyseal region, as

compressive loads are more predominant at this site than torsional loads are (Hayes, 1997). It has been demonstrated in non-disabled older adults that the maximal voluntary isometric, concentric or eccentric hand-grip torques can predict only 38-42% of the variance in CBSI at the distal radius epiphysis, but these same muscle torques could account for as much as 78-90% of the variance in the p-SSI at the radius diaphysis (Frank et al., 2010). Therefore, torsional forces from muscle contractions play a less important role in determining bone strength at the radius epiphyseal site.

4.2.2 Radius Diaphysis

The results showed that at the 33% site of the radius, the paretic side showed significantly lower values in cortical BMC (8%), cortical vBMD (2%), cortical area (8%), and cortical thickness (9%) than the non-paretic side. Such differences were not observed in the control subjects. The results thus indicate a possible decline in bone mass in the hemiparetic radius diaphysis after stroke, with increased cortical bone porosity and thinning of the cortical shell. The findings are in agreement with those of Ashe et al. (2006) and Pang et al. (2007), who also examined the radius diaphysis (30% site) in patients after chronic stroke using pQCT and found a significant side-to-side difference in cortical BMC, cortical vBMD, cortical area, and cortical thickness.

The total area demonstrated no significant side-to-side difference in patients after chronic stroke, indicating relative preservation of overall bone size, a finding similar to what was reported by Ashe et al. (2006) and Pang et al. (2007). Moreover, we found that the marrow cavity area on the paretic side was significantly greater than that on the non-paretic side (Table

9). The combination of preservation of total area with enlarged marrow cavity area indicates possible endosteal resorption after stroke (i.e., bone loss on the endosteal surface rather than on the periosteal surface). These results are in stark contrast with the age-related alterations in macrostructure of the long bone mid-shaft. It has been shown that in the diaphyseal site of the radius, endosteal resorption (i.e., expansion of the marrow cavity area) is accompanied by periosteal apposition (i.e., bone gain on the periosteal surface), leading to outward displacement of the cortex and an increase in total area (Ahlborg et al., 2003; Riggs et al., 2004; Szulc et al., 2006). As adding bone to the periosteal surface would increase the cross-sectional moment of inertia, it could partially offset the detrimental effect of endosteal resorption on bone strength (Burr & Turner, 2003). There was no evidence of periosteal apposition in the radius diaphyseal site among our patients after stroke. Compared with age-related bone changes, endosteal resorption without compensatory periosteal apposition after stroke would lead to a more compromised p-SSI.

Due to the cross-sectional nature of the study, we cannot prove that endosteal resorption indeed was taking place in our stroke subjects. The side-to-side difference in bone parameters detected may be due to bone changes on the paretic side or/and the non-paretic side. Moreover, we also cannot rule out the possibility of existence of bone structural adaptation in response to stroke-induced bone loss on the paretic side. For example, it is possible that stroke may induce bone resorption on both the endosteal and periosteal surfaces. Any compensatory periosteal apposition may then be masked by the concomitant stroke-induced periosteal resorption. It would require a longitudinal study to confirm the occurrence of endosteal resorption in patients after stroke. Although Lazoura et al. (2008) used a longitudinal study design to examine the change in p-SSI at the 20% radius site, and found a significant 7.3% decline on the paretic side,

changes in geometric properties (i.e., total area, marrow cavity area, cortical thickness) were not documented. Therefore, it remains unknown whether bone resorption takes place at the endosteal or/and periosteal surface after stroke.

4.3 Comparison between Trabecular and Cortical Bone Sites

This is the first study to investigate bone density and geometry in different sub-regions of the forearm in the same group of patients after chronic stroke. Overall, the greatest side-to-side difference in aBMD was found in the ultradistal site (9.6%), followed by the mid forearm (7.4%) and 1/3 forearm regions (5.4%). Similarly, the side-to-side difference in the bone strength index at the distal epiphysis (17.8%) was also more pronounced than that in the diaphyseal site (5.9%). These observations indicate that the trabecular bone site is more sensitive to stroke-related impairments than the cortical bone site is, and may partially explain why the radius distal epiphysis is a more common site of fracture in individuals with stroke compared with other upper extremity skeletal sites (Dennis et al., 2002). A previous study by Lazoura et al. (2008) also reported that bone loss is more severe in the trabecular bone site after stroke. Their data showed that the 1-year decline in trabecular vBMD at the 4% radius site was 12.3%, whereas that in cortical vBMD at the 20% radius site was only 3.5%. Research in other populations has also shown more severe bone loss in distal sites of long bones in response to lack of mechanical loading. In a study of cosmonauts after spaceflight, it was found that the onset of bone loss at the distal tibial epiphysis was earlier than that at the tibial diaphysis. Additionally, the extent of microgravity-induced bone loss was greater in the tibial epiphyseal site than in the tibial diaphyseal site at the end of a 6-month spaceflight (Vico et al., 2000). It has also been shown in

patients with spinal cord injury that the loss in bone mass was also faster and more extensive in the tibial epiphysis than in the tibial diaphysis (Eser et al., 2004).

4.4 Sex-related Differences

Some interesting sex-related differences were found between male and female stroke subjects. In general, female stroke subjects tended to demonstrate more compromised bone density and bone strength than their male counterparts did (Table 7-9). For example, a significant side × group interaction effect for the total forearm aBMD, total vBMD of the 4% radius site, and cortical BMC, cortical vBMD and p-SSI at the 33% radius site was detected only in the female group. This phenomenon may be explained by several factors. Firstly, most female stroke patients in this study were post-menopausal. It is known that at menopause, the level of estradiol (the main form of circulating estrogen) is drastically reduced by 90% (Eastell et al., 2003). In contrast, the level of androgens in older men is reduced at a much lower rate, at approximately 1.2% per year (Rosen et al., 2003). The level of testosterone and estrogens (through peripheral conversion of testosterone) in older men is thus much higher than that in older women (Falahati-Nini et al., 2000; Pfeilschifter et al., 1996; Slemenda et al., 1997). Androgens are capable of stimulating periosteal bone formation in the rat model (Turner et al., 1990). Although it remains uncertain whether androgens can exert a similar effect on bone tissue in humans, it cannot be ruled out those higher levels of androgens in our male stroke patients may have resulted in less compromised densitometric and geometric bone properties (Pang et al., 2008). Secondly, the female stroke patients were more severely impaired than their male counterparts, as reflected by a significantly lower mean FMA score and higher MAS score, indicating poorer motor control and more severe spasticity (Table 11). These factors may lead to more compromised bone health

in the paretic upper extremity in female stroke patients. Our results are in contrast with those of Lazoura et al. (2008), who found that male stroke patients sustained greater bone loss than females did. For example, their male subjects suffered a significant decline in p-SSI of 28.6% and 7.4% at the 4% and 20% site of the paretic radius, respectively, within the first year post-stroke. The corresponding decline in female patients after stroke was less, at 19.2 and 7.0%, respectively. Perhaps their male patients after stroke were more severely impaired than the female group, but unfortunately, information on motor impairment and disuse of the paretic upper limb was not documented, thus making interpretation and comparison of the results difficult.

There are also sex-related differences in bone geometry in the tibial diaphysis of patients after chronic stroke (Pang et al., 2008). In their male stroke group, the total area displayed no side-to-side difference, whereas the marrow cavity area on the paretic side was significantly greater than that on the non-paretic side. In the female group, however, the total area on the paretic side was significantly smaller than that on the non-paretic side, whereas the marrow cavity area demonstrated no significant side-to-side difference (Pang et al., 2008). These findings indicate that endosteal resorption in the tibia may be more predominant among male patients after stroke, whereas periosteal resorption may be more apparent in female patients after stroke. The present study, however, provides no evidence of such phenomenon in the radius diaphysis. Although the female subjects tended to have more compromised bone health indicators in the paretic radius diaphysis compared with the male subjects, as reflected by the greater side-to-side difference in bone parameters, both male and female stroke groups showed a greater marrow cavity area on the paretic side, with no significant side-to-side difference in total area. Apparently, the gender-related differences in bone structural adaptation post-stroke for a non-

weight-bearing bone, such as the radius, are distinct from that for a weight-bearing bone, such as the tibia.

4.5 Lack of Significant Main Effects of Group

The overall significant main effect of side was attributable to the remarkable difference between the paretic and non-paretic sides in the stroke group, despite that the side-to-side difference in the same parameters demonstrated by the control group is much smaller. On the other hand, the main effects of Group for all DXA and pQCT parameters were not significant, except for total vBMD for women. The lack of significant main effect of group is mainly due to the fact that the bone parameters measured on the dominant side of control subjects are not that different from those on the non-paretic side of stroke subjects. The results may indicate that the non-paretic side has similar bone health status as in non-disabled controls, despite that the physical activity level is significantly lower in the stroke group. Other factors such as genetic make-up , dietary habits, may contribute to the lack of between-group difference. The comparison of bone parameters between the two sides within the same subjects would provide some control of these co-factors that influence bone health.

The results likely indicate that that the overall bone health condition was not particularly compromised prior to stroke, compared with age-matched controls. After the stroke, however, the paretic side may have sustained substantial changes in bone density/geometry, which accounted for the side-to-side difference in bone parameters in the stroke group, and also the significant side × group interaction effect (i.e. the magnitude of side-to-side difference in bone parameters is group-dependent).

4.6 Determinants of DXA-derived aBMD and pQCT-derived Bone Strength Indices

One of the objectives of this study was to identify significant determinants of important bone parameters in the paretic forearm. Overall, the results showed that, aside from advancing age and female gender, neuromuscular factors, including muscle weakness, spasticity and chronic disuse, are more important determinants of DXA-derived aBMD and pQCT-derived bone strength indices than are cardiovascular factors.

4.6.1 Demographic factors

BMI and post-stroke duration were entered in the regression model to account for the effect of body size and chronicity of stroke. It was found that BMI and post-stroke duration were not significant determinants of aBMD, CBSI and p-SSI. The results are consistent with previous studies in stroke. For example, BMI and post-stroke duration were not significantly related to percent side-to-side difference in cortical thickness and cortical BMC measured at the radius diaphysis in individuals with chronic stroke (Pang et al. 2007). One potential explanation is that other factors, such as muscle strength, may have precedence over BMI and post-stroke duration in determining the bone density/strength indices. For example, the integrity of bone tissue may be better for someone who have had a mild stroke (mild muscle weakness) for 5 years, compared with someone who have had a severe stroke (complete paralysis) for 1 year.

4.6.2 Neuromuscular Function

4.6.2.1 Muscle Weakness

Among the various neuromuscular factors, reduced grip strength turned out to be the most predominant determinant of the paretic forearm bone health. Firstly, grip strength or percent side-to-side difference in grip strength was identified as a significant determinant in all the regression models constructed. Secondly, when compared with other neuromuscular impairments measured, grip strength contributed the most variance to the dependent variable.

Muscle weakness is a common symptom after stroke (Nakayama et al., 1994; Parker et al., 1986), According to our findings, the hand-grip strength on the paretic side was significantly lower (44.5%) than on the non-paretic side, indicating substantial paresis on the affected side. In addition to the decrease in central drive due to the disruption of the motor pathways (Mima et al., 2001), other factors may contribute to the reduced ability to voluntarily generate muscle force, including a reduction in the number of functioning motor units (Hara et al., 2000; McComas et al., 1973), changes in motor unit recruitment and discharge rate (Frontera et al., 1997; Gemperline et al., 1995; Rosenfalck et al., 1980), and muscle atrophy (Iversen et al., 1989; Pang et al., 2005a; Ryan et al., 2002). It is well known that muscle contractions provide a good source of mechanical strain, which plays an important role in osteogenesis (Robling et al., 2002). Reduced muscle loading after a stroke event may thus have a negative impact on bone health.

Our results thus extend the findings of previous studies in highlighting the intimate relationship between bone health and muscle strength in other populations (i.e., the muscle-bone unit) (Di Monaco et al., 2000; Frank et al., 2010; Hughes et al., 1995; Osei-Hyiaman et al., 1999; Ozdurak et al., 2003). For example, among non-disabled older adults, hand-grip strength was significantly related to the bone strength indices at the radius epiphysis and diaphysis (Frank et al., 2010). Hand-grip strength was also independently associated with the distal radius BMC (Di

Monaco et al., 2000) and metacarpal aBMD (Osei-Hyiaman et al., 1999) in postmenopausal women.

The muscle-bone relationship has also been reported in previous stroke studies. Pang et al. (2005a) reported that the composite muscle-strength score of the paretic upper extremity was a significant predictor of the paretic total arm BMC in individuals with chronic stroke. Using pQCT, Ashe et al. (2006) demonstrated a positive relationship between the composite paretic arm muscle score and p-SSI measured at the 30% site of the radius in a small sample of 15 patients with chronic stroke (R^2=0.72). However, Pang et al. (2007) found that the percent side-to-side difference in paretic arm muscle strength was not correlated with percent side-to-side difference in p-SSI in a group of 47 patients after chronic stroke (r=0.224, p>0.05), which is in contrast with the results of the present study. Several factors may account for the difference in results. Firstly, the paretic arm composite muscle strength score was used in their study, whereas the grip-strength score was used here. It is known that muscle weakness is typically more severe in the distal part of the extremity than in the proximal part (Adams et al., 1990; Colebatch & Gandevia, 1989). It is thus likely that the grip-strength score is a stronger predictor of bone parameters of the distal skeletal sites of the upper limb, such as the radius. Secondly, this study had a larger sample size. The associated increase in statistical power would increase the ability to detect a significant correlation.

4.6.2.2 Motor Function

Recovery of motor function, as measured by the FMA score, was significantly associated with the paretic radius CBSI and p-SSI and percent side-to-side difference in aBMD of all forearm regions. Previous stroke studies have also indicated a positive relationship between

BMD and degree of motor recovery (Ashe et al., 2006; Jørgensen & Jacobsen, 2001; Pang et al., 2007; Prince et al., 1988; Sato et al., 1998). For example, a significant relationship between aBMD of the second metacarpal and Brunstrom's staging was identified by Sato et al. (1998). Jørgensen & Jacobsen (2001) also showed that motor function, as measured by the Scandinavian Stroke Scale, at baseline was a powerful predictor of 1-year aBMD decline in the proximal humerus after stroke. Specifically, those with severe upper-limb motor impairment throughout the first year post-stroke suffered a much greater degree of bone loss (25%) than those who had recovered (8%). A significant relationship between FMA score and p-SSI (R^2=0.38) at the 30% site of the radius in patients after chronic stroke was also demonstrated by Ashe et al. (2006). Compared with grip strength, the FMA was not as strong a determinant of forearm aBMD and radius-bone strength indices. The FMA provides an overall assessment of selective joint movements and reflex activity of both proximal and distal parts of the upper limbs, rather than the distal part only. In addition, the tasks involved in the FMA do not require a high level of muscle force for successful execution. It is also a laboratory-based performance test, which does not quantify the actual amount of use of the paretic upper extremity in daily activities.

4.6.2.3 Chronic Disuse

In general, our stroke patients did not use their affected upper limb in functional activities as frequently as they did prior to the stroke event, as indicated by the compromised MAL score. Many stroke patients tend to adopt a compensatory strategy, using only the non-paretic upper limb to perform daily functional activities (Taub et al., 1993). In the present study, the MAL score was identified as one of the significant predictors in some of our regression models. Our results are thus in line with a previous study in showing that the MAL score was

significantly related to the side-to-side difference in cortical BMC and cortical thickness of the 30% site of the radius in patients after chronic stroke (Pang et al, 2007). Again, compared with grip strength, the MAL was not as strong a determinant of the bone parameters of interest. It may be related to the fact that the 30 functional tasks included in the MAL were predominantly light functional activities (e.g., brushing teeth, opening doors), whereas testing of grip strength requires maximal effort from the patient.

4.6.2.4 Spasticity

The MAS score remained independently associated with most of the forearm aBMD parameters and CBSI of the paretic radius, after accounting for the effects of other relevant factors. The relationship is negative, indicating those with more severe spasticity tend to have more compromised aBMD and CBSI. However, the MAS was not significantly associated with p-SSI in multivariate analysis ($p = 0.496$). Previous studies on spasticity and bone health also reported conflicting results. It has been shown that in patients with spinal cord injury, patients with flaccid paralysis had more bone loss than those with spastic paralysis did (Wilmet et al., 1995). Pang et al. (2005a) identified no significant relationship between total arm BMC and the MAS score in patients after chronic stroke. On the other hand, more severe spasticity was related to a greater side-to-side difference in cortical BMC and cortical thickness in the midshaft radius (Pang et al, 2007) and trabecular bone loss at the ultradistal radius and ulna (Prince et al., 1988). The different results may be partially attributable to the different skeletal sites measured, as well as the method of bone imaging. The conflicting results may also be related to the complex effect of spasticity on bone tissue. On one hand, increased muscle tone associated with spasticity may provide a source of mechanical loading to the bone and may thus have a protective effect on

bone. On the other hand, severe spasticity may impair upper extremity function, which leads to chronic disuse and subsequent bone loss. Whether a spasticity threshold exists above or below which bone health would not be significantly influenced is currently unknown (Pang et al., 2008; Pang et al., 2010). It is also noteworthy that only 18.5% of the sample were categorized as having moderate/severe spasticity. Thus, the interpretation of results warrants caution.

4.6.3 Cardiovascular Function

Another important purpose of this study was to investigate the association of cardiovascular function with bone-health indicators of the paretic upper extremity in patients after chronic stroke. In general, the study showed that among the various cardiovascular variables measured, only C_1 was associated with the paretic radius p-SSI.

4.6.3.1Vascular Elasticity

Although C_1 and C_2 showed a significant, positive association with paretic forearm aBMD and radius bone strength indices (Table 13) in bivariate correlation analysis, their effects were diminished in multivariate analysis. Only C_1 remained independently associated with the p-SSI of the paretic radius epiphysis. Why is C_1 more powerful than C_2 in determining the p-SSI of the radius diaphysis? Vascular disease involves the dysfunction of the endothelial lining of small arteries and arterioles, and is initiated by one or more factors, such as aging, inactivity, diabetes, high cholesterol and pressure, all of which are prevalent in patients post-stroke (Kopunek et al., 2007; Michael et al., 2005). This early change in endothelial function may be detected as a premature loss of arterial elasticity (Hypertension Diagnostics Inc., 2005).. Such dysfunction of

the endothelium can occur in the entire arterial system, but is easier to detect in the very small arteries and arterioles, and results in a reduction of C_2. With the progression of vascular disease, large arteries become affected and C_1 begins to decline (Hypertension Diagnostics Inc., 2005).. Thus, a reduction in C_1 is indicative of more advanced cardiovascular dysfunction, and may have a more adverse influence on the integrity of bone tissue. This may partially explain why C_1, rather than C_2, is a significant determinant of the p-SSI of the hemiparetic radius.

It is also interesting that C_1 was significantly related to bone strength index at the radius diaphysis (i.e., p-SSI), but not at the radius epiphysis (i.e., CBSI). This is in contrast with the results reported in the tibia (Pang et al., 2008; Pang et al., 2010). Peak VO_2 has been found to be more strongly associated with the CBSI at the tibial epiphysis (Pang et al., 2010) than the p-SSI at the tibial diaphysis (Pang et al., 2008) among chronic stroke patients. It is thought that the epiphyseal site may be more sensitive to changes in cardiovascular function due to the fact that trabecular bone is more vascularized and metabolically active than cortical bone (Whitney et al., 2004). The difference may also be related to the difference in cardiovascular parameters used and skeletal sites measured. Taken together, the results suggest site-specific differences in the relationship between bone alterations and stroke impairment. The implication is that enhancing bone strength in the paretic upper extremity may require different rehabilitative strategies from those used for the paretic lower extremity.

4.6.3.2 Oxygen Consumption Rate, Cardiac Index and Stroke-volume Index

The results showed that VO_2 rate during the 6MWT in the stroke group was significantly lower than that in the control group, but was not significantly related to the forearm aBMD or

bone strength indices. Previous published studies of chronic stroke patients, however, have shown that cardiovascular fitness, as measured by the peak VO_2, is strongly associated with hip aBMD and tibial bone strength index (Pang & Eng, 2005b; Pang et al., 2008; Pang et al., 2010). The difference in results may be explained by several factors. Firstly, the method of VO_2 measurement differed. We measured the VO_2 during the 6MWT (a submaximal exercise test), whereas peak VO_2 was evaluated during a maximal exercise test using cycle ergometry in previous studies. Although the VO_2 rate during the 6MWT had significant association with peak VO_2 in patients after chronic stroke, the correlation was only moderate (Eng et al., 2004; Pang & Eng, 2005a). Secondly, the site of bone measurement was different. The association between VO_2 and bone health may be stronger in the lower extremity sites than in the upper extremity sites. Poor cardiovascular fitness is common among patients after chronic stroke (Mackay-Lyons & Makrides, 2002; Pang & Eng, 2005a) and lack of ambulatory activity may be a major contributing factor. It is known that the ambulatory activity of patients after chronic stroke is extremely low (mean = 2,837 steps per day) compared with that of sedentary older adults (5,000-6,000 steps per day) (Michael et al., 2005). This lack of ambulatory activity, which is a loading activity in itself, may adversely influence both bone health and cardiovascular health. This may explain the stronger association of cardiovascular health indicators with lower limb-bone parameters.

It is intriguing that CI and SI values showed no significant between-group difference. The stroke subjects in this study were all community-dwelling and had relatively good mobility function, which may thus explain the lack of significant findings. Moreover, the measurement of CI and SI was taken during the resting condition. It is possible that the between-group difference in these parameters may be apparent only when the subjects are exposed to more demanding

conditions. Physical exercise can often elicit cardiovascular abnormalities that are not present at rest (Fletcher et al., 2001). For example, systolic cardiac dysfunction and abnormal blood pressure can often be detected during the early stage of exercise, but not at rest (Fletcher et al., 2001). We acknowledge that ICG has been used to measure cardiac output during exercise by others (Scherhag et al., 2005). We originally attempted to use the ambulatory ICG unit to measure cardiac output during the 6MWT. However, the signals obtained were really unstable with substantial movement artifacts and could not be used for analysis.

4.7 Clinical Implications

This study has several important clinical implications. Among the various stroke impairments studied, muscle weakness is the strongest determinant of forearm aBMD and bone strength indices of the radius in the paretic forearm. Grip-strength assessment may be an easy-to-administer, inexpensive and reliable clinical tool to screen those post-stroke patients who have compromised upper extremity bone health and requires further investigation.

Our results also point to the potential importance of muscle strengthening as a way to enhance bone health in the paretic forearm. Previous studies have shown that muscle-strengthening work can induce corticalization of trabecular bone at the endosteal surface and periosteal apposition in the ultradistal radius region among post-menopausal women (Adami et al., 1999). A randomized controlled study has also shown that the combination of impact aerobic exercise and functional muscle strengthening is effective in enhancing trabecular bone mineral content in the tibial distal epiphysis and cortical thickness in the tibial diaphysis (Pang et al. 2006). Whether muscle strengthening (i.e., resistance training, electrical muscle stimulation, etc.)

can enhance bone density and the geometry of the radius in the stroke population remains to be investigated.

A higher level of spasticity was also independently associated with aBMD of the forearm and CBSI of the radius epiphysis of the radius on the paretic side. Intervention to decrease spasticity is indicated when spasticity is so severe that it affects daily functioning (Yelnik et al., 2010). Common clinical management of spasticity includes pharmacological treatment, such as baclofen and tizanidine, and non-pharmacological treatment, such as orthoses, injection of botulinum neurotoxins, electrical stimulation, passive stretching and active exercise (Bhakta 2000; Elia et al., 2009; Rekand, 2010; Yelnik et al., 2010). It would be interesting to determine the influence of these different interventions on the integrity of bone tissue in this patient group.

Chronic disuse of the paretic upper extremity was also identified as a significant determinant of many bone parameters measured. Many patients after stroke have the tendency to compensate by performing all daily activities using the non-paretic upper limb, especially when the affected upper limb happens to be on the non-dominant side (Harris et al., 2007; Taub et al., 1993), a phenomenon termed "learned non-use". One clinical implication of our results is that bone health in the paretic upper limb could be potentially improved by increasing the functional use of the paretic upper limb. Certain therapeutic approaches, such as the constraint-induced movement therapy, are specifically designed to counteract the habitual non-use of the paretic upper limb (Kunkel et al., 1999; Page et al., 2004; Sterr et al., 2002). It would be clinically relevant to examine whether intensive movement therapy of the paretic limb would have any beneficial effects on bone-health indicators in the stroke population.

C_1 is also independently associated with the bone strength index in the radius diaphysis, indicating that cardiovascular health may affect the integrity of bone tissue at this site, although

its influence is less predominant than grip strength. Modifications of vascular health may thus play a potentially important role in enhancing bone health of the paretic upper limb. There is evidence that moderate aerobic training can reduce large artery stiffness in young individuals (Kingwell, 2002). Impact aerobic exercises have also been shown to improve aBMD of the hip in postmenopausal women and patients with chronic stroke (Chien et al., 2000; Pang & Eng, 2005b; Pang et al., 2006). It would be worth investigating the effects of aerobic exercise training on upper extremity bone health in patients after stroke.

4.8 Limitations and Future Research Directions

This study has several limitations. Firstly, a cross-sectional design was employed, and the influence of stroke on bone was assessed mainly by examining the side-to-side difference in bone parameters. The advantage of this approach is that it allowed us to evaluate the effects of stroke on bone health, while providing some control for the various genetic and environmental factors (e.g., nutrition) that may influence bone parameters across different subjects. However, the cross-sectional design rendered us unable to evaluate longitudinal bone changes over time. Although a control group was incorporated in the study, it could not be ruled out that changes in both the paretic and non-paretic sides may contribute to the observed side-to-side difference in bone parameters. For example, in a prospective study, Ramnemark et al. (1999a) showed that reduction in aBMD of the ultradistal radius, proximal humerus and total arm on the paretic side was accompanied by an increase in aBMD in the corresponding skeletal sites on the non-paretic side within the first year post-stroke. On the other hand, Lazoura et al. (2008) demonstrated that both the paretic and non-paretic sides sustained significant decline in vBMD at the radius distal epiphysis, though the degree of bone loss was less extensive on the non-paretic side (7%) than on

the paretic side (12%). To date, no prospective studies have examined the changes in both aBMD and geometry in the forearm region in the same patients after stroke. This important area of research awaits further investigation.

While significant correlations between bone health indicators and certain stroke impairments were identified, causality could not be established. Randomized controlled studies are required to determine whether modification of neuromuscular factors through muscle strengthening, constraint-induced therapy or other rehabilitative approaches could successfully enhance or maintain bone density and geometry in the hemiparetic upper extremity among patients after chronic stroke.

The resolution of the pQCT images (0.4mm voxel size) was not high enough to accurately measure cortical thickness in the distal radius epiphysis. Partial volume effect occurs when there is heterogeneous material within a single voxel. In skeletal sites where cortical bone shell is thin (<2mm), such as the distal radius epiphysis on the paretic side, the voxels may be only partly filled by bone material and soft tissue (Hangartner et al., 1996). These voxels will inaccurately yield lower BMD because a voxel's BMD value is the mean density of all the tissues within it. Therefore, cortical thickness in the distal radius epiphysis was not used as an outcome measure in this study, as in previous pQCT studies in patients after stroke (Ashe et al., 2006; Pang et al., 2007). While using a higher resolution may improve the accuracy of measurement, it necessitates a longer scanning time, which may increase the probability of movement artifacts, especially considering that individuals with stroke have impaired motor control, with tremor and spasticity not being uncommon (Pang et al., 2007). In the study by Pang et al. (2007) using a voxel size of 0.3mm, out of 63 patients who underwent pQCT scanning of the radius, 15 scans (24%) were eliminated from analysis because of movement artifacts related

to movement, tremor or spasticity. We believe that the current pQCT protocol is appropriate, considering the need to achieve a balance between maintaining precision of measurement and reducing movement artifacts.

The MAS has a number of inadequacies as a measure of spasticity. A recent study (Fleurens et al., 2010) has shown that the interrater reliability is modest (0.58 for elbow flexors and 0.63 for knee extensors). The rating on the Ashworth Scale is only moderately associated with electromyographic parameters (Fleurens et al., 2010).

In this study, we only measured the spasticity of elbow flexors, but the assessment of "spasticity" could have been extended to other regions of the paretic upper extremity, thereby obtaining a more comprehensive picture of the severity of spasticity. However, the disadvantage of this would be the increase in duration of the assessment session. The elbow region was chosen as a representative measure because it is commonly used to evaluate spasticity in the stroke population (O'Dwyer et al. 1996; Watkins et al. 2002). Spasticity of the elbow flexors as measured by the MAS has also been identified as a significant determinant of activity participation (Harris & Eng, 2007).

Another limitation of the study is related to potential sampling bias. The post-stroke patients in this study were all community-dwelling and the majority was recruited from stroke self-help groups, which hold regular monthly activities for their members. Therefore, the subjects in this study tended to be more physically able and socially active than their counterparts who did not participate in these self-help groups. Our sample thus may not be representative of the greater chronic stroke population.

The vascular elasticity indices (C_1, C_2) are indicators of systemic vascular health only. Perhaps measures of localized blood flow in the paretic wrist region may be more strongly

associated with the bone parameters on the same side. One interesting research area worth exploring is to use other technologies, such as dynamic contrast-enhanced magnetic resonance (MR) imaging, to measure local perfusion in the radius and its association with BMD and bone strength (Griffith et al., 2008).

Finally, our various regression models explained only 19-78% of the variance in different bone parameters, indicating that some potentially important factors underlying bone health post-stroke were not captured in this study. For example, dietary factors and sunshine exposure may have an important influence on bone health, but were not systematically evaluated in this study. Increasing the number of predictors in the regression models, however, necessitates a greater sample size. Further studies should employ a larger sample size and address the relationship between these factors and bone health in patients after chronic stroke.

4.9 Conclusion

Using both DXA and pQCT techniques, this novel study aimed to examine the influence of stroke on bone density, geometry and bone strength indices of the forearm region among patients after chronic stroke, and to identify their clinical correlates. The results showed that in the stroke group, the DXA-derived aBMD values in different regions of the forearm on the paretic side were significantly lower than those on the non-paretic side, and that the side-to-side differences in aBMD values in these patients were greater than those of their non-disabled counterparts. The pQCT results also showed more compromised vBMD values in the paretic radius epiphysis and diaphysis when compared with controls, leading to lower bone strength

index values, despite no significant side-to-side difference in total area. These findings suggest a lack of compensatory structural adaptations in response to stroke-related bone loss.

Overall, among the various stroke-related neuromuscular and cardiovascular impairments studied, muscle weakness was the most important determinant of the DXA-derived aBMD values and pQCT-derived bone strength indices. Promoting muscle strength of the paretic upper extremity may be an important treatment strategy to enhance or maintain bone mass in the paretic upper extremity, and warrants further investigation.

References

1. Adami S, Gatti D, Braga V, Bianchini D, Rossini M. Site-specific effects of strength training on bone structure and geometry of ultradistal radius in postmenopausal women. J Bone Miner Res 1999; 14: 120-4.
2. Adams RW, Gandevia SC, Skuse NF. The distribution of muscle weakness in upper motoneuron lesions affecting the lower limb. Brain 1990; 113: 1459-76.
3. Agabiti-Rosei E, Muiesan ML. Carotid atherosclerosis, arterial stiffness and stroke events. Adv Cardiol 2007; 44: 173-86.
4. Ahlborg HG, Johnell O, Turner CH, Rannevik G, Karlsson MK. Bone loss and bone size after menopause. N Engl J Med 2003; 349: 327-34.
5. Alberts MJ, Atkinson R. Risk reduction strategies in ischaemic stroke, the role of antiplatelet therapy. Clin Drug Invest 2004; 24: 245-54.
6. American Heart Association. Heart Disease and Stroke Statistics—2008 Update. Dallas, Texas: American Heart Association; 2008.
7. American Heart Association: Heart Disease and Stroke Statistics - 2005 Update. Dallas, Texas: American Heart Association; 2005.
8. Andersson AG, Kamwendo K, Seiger A, Appelros P. How to identify potential fallers in a stroke unit: validity indexes of 4 test methods. J Rehabil Med 2006; 38: 186-91.
9. Ashe MC, Fehling P, Eng JJ, Khan KM, McKay HA. Bone geometric response to chronic disuse following stroke: a pQCT study. J Musculoskelet Neuronal Interact 2006; 6: 226-33.

10. Baim S, Wilson CR, Lewiecki EM, Luckey MM, Downs RW Jr, Lentle BC. Precision assessment and radiation safety for dual-energy X-ray absorptiometry: position paper of the International Society for Clinical Densitometry. J Clin Densitom 2005; 8: 371-8.
11. Barengolts EI, Berman M, Kukreja SC, Kouznetsova T, Lin C, Chomka EV. Osteoporosis and coronary atherosclerosis in asymptomatic postmenopausal women. Calcif Tissue Int 1998; 62: 209 –213.
12. Beaupre GS, Lew HL. Bone-density changes after stroke. Am J Phys Med Rehabil 2006; 85: 464-72.
13. Bhakta BB, Cozens JA, Chamberlain MA, Bamford JM. Impact of botulinum toxin type A on disability and carer burden due to arm spasticity after stroke: a randomised double blind placebo controlled trial. J Neurol Neurosurg Psychiatry 2000; 69: 217-21.
14. Blake GM, Fogelman I. Role of dual-energy X-ray absorptiometry in the diagnosis and treatment of osteoporosis. J Clin Densitom 2007; 10: 102-10.
15. Bohannon RW, Smith MB. Interrater reliability of a modified Ashworth scale of muscle spasticity. Phys Ther 1987; 67: 206-7.
16. Brass LM. Strategies for primary and secondary stroke prevention. Clin Cardiol 2006; 29: II21-7.
17. Browner WS, Seeley DG, Vogt TM, Cummings SR. Non-trauma mortality in elderly women with low bone mineral density. Lancet 1991; 338: 355-8.
18. Browner WS, Pressman AR, Nevitt MC, Cauley JA, Cummings SR. Association between low bone density and stroke in elderly women. The study of osteoporotic fractures. Stroke 1993; 24: 940-46.

19. Burr DB, Turner Ch. Biomechanics of bone. In: Favus MJ (ed) Primer on the metabolic bone diseases and disorders of mineral metabolism, 5th edn. American Society for Bone and Mineral Research, Washington DC, 2003; 58–64
20. Cameron JD, Rajkumar C, Kingwell BA, Jennings GL, Dart AM. Higher systemic arterial compliance is associated with greater exercise time and lower blood pressure in a young older population. J Am Geriatr Soc 1999; 47: 653–6.
21. Carter KN, Anderson CS, Hackett ML, Barber PA, Bonita R; Auckland Regional Community Stroke Study Group. Improved survival after stroke: Is admission to hospital the major explanation? Trend analyses of the Auckland Regional Community Stroke Studies. Cerebrovasc Dis 2007; 23: 162-8.
22. Celermajer DS, Sorensen KE, Gooch VM, Spiegelhalter DJ, Miller OI, Sullivan ID, Lloyd JK, Deanfield JE. Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. Lancet 1992; 340: 1111-5.
23. Centre for Health Protection, Department of Health, Hong Kong, 2010, website: http://www.chp.gov.hk/en/data/4/10/27/340.html.
24. Chaiwanichsiri D, Jiamworakul A, Kitisomprayoonkul W. Falls among stroke patients in Thai Red Cross rehabilitation center. J Med Assoc Thai 2006; 89: S47-52.
25. Charloux A, Lonsdorfer-Wolf E, Richard R, Lampert E, Oswald-Mammosser M, Mettauer B, Geny B, Lonsdorfer J.A new impedance cardiograph device for the non-invasive evaluation of cardiac output at rest and during exercise: comparison with the "direct" Fick method. Eur J Appl Physiol 2000; 82: 313-20.

26. Chau JP, Thompson DR, Chang AM, Woo J, Twinn S, Cheung SK, Kwok T. Depression among Chinese stroke survivors six months after discharge from a rehabilitation hospital. J Clin Nurs. 2010; 19: 3042-50.
27. Cheng PT, Liaw MY, Wong MK, Tang FT, Lee MY, Lin PS. The sit-to-stand movement in stroke patients and its correlation with falling. Arch Phys Med Rehabil 1998; 79: 1043-6.
28. Chien MY, Wu YT, Hsu AT, Yang RS, Lai JS. Efficacy of a 24-week aerobic exercise program for osteopenic postmenopausal women. Calcif Tissue Int 2000; 67: 443-8.
29. Chiu KY, Pun WK, Luk KD, Chow SP. A prospective study on hip fractures in patients with previous cerebrovascular accidents. Injury 1992; 23: 297-9.
30. Cohn JN, Finkelstein SM, McVeigh G, Morgan D, LeMay L, Robinson J, Mock J. Non-invasive pulse wave analysis for early detection of vascular disease. H ypertension 1995; 26: 503-8.
31. Colebatch JG, Gandevia SC. The distribution of muscular weakness in upper motor neuron lesions affecting the arm. Brain 1989; 112: 749-63.
32. Collins TC, Ewing SK, Diem SJ, Taylor BC, Orwoll ES, Cummings SR, Strotmeyer ES, Ensrud KE. Peripheral arterial disease is associated with higher rates of hip bone loss and increased fracture risk in older men. Circulation 2009; 119: 2305-12.
33. Cooper C. The crippling consequences of fractures and their impact on quality of life. Am J Med 1997; 103: 12S-19S.
34. Davenport RJ, Dennis MS, Wellwood I, Warlow CP. Complications after acute stroke. Stroke 1996; 27: 415-20.

35. Demaerschalk BM, Hwang HM, Leung G. US cost burden of ischemic stroke: a systematic literature review. Am J Manag Care 2010; 16: 525-33.
36. Dennis MS, Lo KM, McDowall M, West T. Fractures after stroke. Frequency, types and associations. Stroke 2002; 33: 728-34.
37. Di Monaco M, Di Monaco R, Manca M, Cavanna A. handgrip strength is an independent predictor of distal radius bone mineral density in postmenopausal women. Clin Rheumatol 2000; 19: 473– 6.
38. Di Monaco M, Vallero F, Di Monaco R, Mautino F, Cavanna A. Functional recovery and length of stay after hip fracture in patients with neurologic impairment. Am J Phys Med Rehabil 2003; 82: 143-8.
39. Duprez DA, De Buyzere MM, De Bruyne L, Clement DL, Cohn JN. Small and large artery elasticity indices in peripheral arterial occlusive disease (PAOD). Vasc Med 2001; 6: 211-4.
40. Duprez DA, De Buyzere ML, De Backer TL, Van DeVeire N, Clement DL, Cohn JN. Relationship between arterial elasticity indices and carotid artery intima-media thickness. Am J Hypertens 2000; 13: 1226-32.
41. Eastell R. Pathogenesis of postmenopausal osteoporosis. In: Favus MJ (ed.) Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism, 5th ed. American Society for Bone and Mineral Research, Washington, DC, USA, 200 ; 314–316.
42. Elia AE, Filippini G, Calandrella D, Albanese A. Botulinum neurotoxins for post-stroke spasticity in adults: a systematic review. Mov Disord 2009; 24: 801-12.

43. Eng JJ, Dawson AS, Chu KS. Submaximal exercise in persons with stroke: test-retest reliability and concurrent validity with maximal oxygen consumption. Arch Phys Med Rehabil 2004; 85: 113–8.

44. Ensrud KE, Blackwell T, Mangione CM, Bowman PJ, Bauer DC, Schwartz A, Hanlon JT, Nevitt MC, Whooley MA. Central nervous system active medications and risk of fractures in older women. Arch Intern Med 2003; 163: 949-57.

45. Eser P, Schiessl H, Willnecker J. Bone loss and steady state after spinal cord injury: a cross-sectional study using pQCT. J Musculoskelet Neuronal Interact 2004; 4: 197-8.

46. Eugene M, Vandewalle H, Bertholon JF, Teillac A. Arterial elasticity and physical working capacity in young men. J Appl Physiol 1986; 61: 1720-3.

47. Evenson KR, Rosamond WD, Cai J, Toole JF, Hutchinson RG, Shahar E, Folsom AR. Physical activity and ischemic stroke risk. The atherosclerosis risk in communities study. Stroke 1999; 30: 1333-9.

48. Everson SA, Lynch JW, Kaplan GA, Lakka TA, Sivenius J, Salonen JT. Stress-induced blood pressure reactivity and incident stroke in middle-aged men. Stroke 2001; 32: 1263-70.

49. Fagard RH, Celis H, Thijs L, Staessen JA, Clement DL, De Buyzere ML, De Bacquer DA. Daytime and nighttime blood pressure as predictors of death and cause-specific cardiovascular events in hypertension. Hypertension 2008; 51: 55-61.

50. Falahati-Nini A, Riggs BL, Atkinson EJ, O'Fallon WM, Eastell R, Khosla S. Relative contributions of testosterone and estrogen in regulating bone resorption and formation in normal elderly men. J Clin Invest 2000; 106: 1553–60.

51. Farhat GN, Cauley JA, Matthews KA Newman AB, Johnston J, Mackey R, Edmundowicz D, Sutton-Tyrrell K. Volumetric BMD and vascular calcification in middle-aged women: the Study of Women's Health Across the Nation. J Bone Miner Res 2006; 21: 1839-46.

52. Farhat GN, Newman AB, Sutton-Tyrrell K Matthews KA, Boudreau R, Schwartz AV, Harris T, Tylavsky F, Visser M, Cauley JA. The association of bone mineral density measures with incident cardiovascular disease in older adults. Osteoporos Int 2007; 18: 999-1008.

53. Feigin VL, Lawes CM, Bennett DA, Barker-Collo SL, Parag V. Worldwide stroke incidence and early case fatality reported in 56 population-based studies: a systematic review. Lancet Neurol 2009; 8: 355-69.

54. Ferretti JL, Cointry GR, Capozza RF. Noninvasive analysis of bone mass, structure, and strength. In: An YH, editor. Orthopaedic Issues in Osteoporosis. Boca Raton, FL: CRC Press; 2002; 145-67.

55. Feske W, Finkelstein SM, Francis G, Cohn JN. Arterial vascular compliance response to exercise in hypertension. Biomed Sci Instrum 1988; 24: 161–5.

56. Fletcher GF, Balady GJ, Amsterdam EA, Chaitman B, Eckel R, Fleg J, Froelicher VF, Leon AS, Piña IL, Rodney R, Simons-Morton DA, Williams MA, Bazzarre T. Exercise standards for testing and training: a statement for healthcare professionals from the American Heart Association. Circulation 2001; 104: 1694-740.

57. Fleuren JF, Voerman GE, Erren-Wolters CV, Snoek GJ, Rietman JS, Hermens HJ, Nene AV. Stop using the Ashworth Scale for the assessment of spasticity. J Neurol Neurosurg Psychiatry. 2010; 81: 46-52.

58. Frost HM. Bone's mechanostat: a 2003 update. Anat Rec Discov Moll Cell Evol Biol 2003; 275A: 1081-101.

59. Forster A, Young J. Incidence and consequences of falls due to stroke: a systematic inquiry. BMJ 1995; 311: 83-6.

60. Frank AW, Lorbergs AL, Chilibeck PD, Farthing JP, Kontulainen SA. Muscle cross sectional area and grip torque contraction types are similarly related to pQCT derived bone strength indices in the radii of older healthy adults. J Musculoskelet Neuronal Interact 2010; 10: 136-41.

61. Frontera WR, Grimby L, Larsson L. Firing rate of the lower motorneuron and contractile properties of its muscle fibres after upper motorneuron lesion in man. Muscle Nerve 1997; 20: 938– 47.

62. Fugl-Meyer AR, Jaasko L, Leyman I, Olsson S, Steglind S. The post-stroke hemiplegic patient. 1. a method for evaluation of physical performance. Scand J Rehabil Med 1975; 7: 13-31.

63. Garrett NA, Brasure M, Schmitz KH, Schultz MM, Huber MR. Physical inactivity: direct cost to a health plan. Am J Prev Med 2004; 27: 304-9.

64. Genant HK, Lang TF, Engelke K Fuerst T, Glüer C, Majumdar S, Jergas M. Advances in the noninvasive assessment of bone density, quality, and structure. Calcif Tissue Int 1996; 59: S10-5.

65. Gemperline JJ, Allen S, Walk D, Rymer WZ. Characteristics of motor unit discharge in subjects with hemiparesis. Muscle Nerve 1995; 18: 1101– 14.

66. Darren G, Mallery P. SPSS for Windows Step by Step A Simple Guide and Reference 17.0 Update, 10th Edition. Pearson Education. 2010.

67. Gladstone DJ, Danells CJ, Black SE. The fugl-meyer assessment of motor recovery after stroke: a critical review of its measurement properties. Neurorehabil Neural Repair. 2002; 16: 232-40.

68. Gresham GE, Fitzpatrick TE, Wolf PA, McNamara PM, Kannel WB, Dawber TR. Residual disability in survivors of stroke-The Framingham study. N Engl J Med 1975; 293: 954-6.

69. Grey E, Bratteli C, Glasser SP Alinder C, Finkelstein SM, Lindgren BR, Cohn JN. Reduced small artery but not large artery elasticity is an independent risk marker for cardiovascular events. Am J Hypertens 2003; 16: 265-9.

70. Griffith JF, Yeung DK, Tsang PH Choi KC, Kwok TC, Ahuja AT, Leung KS, Leung PC. Compromised bone marrow perfusion in osteoporosis. J Bone Miner Res 2008; 23: 1068-75.

71. Grisso JA, Chiu GY, Maislin G, Steinmann WC, Portale J. Risk factors for hip fractures in men: a preliminary study. J Bone Miner Res 1991; 6: 865-8.

72. Grisso JA, Kelsey JL, Strom BL Chiu GY, Maislin G, O'Brien LA, Hoffman S, Kaplan F. Risk factors for falls as a cause of hip fracture in women. The Northeast Hip Fracture Study Group. N Engl J Med 1991; 324: 1326-31.

73. Haapasalo H, Kontulainen S, Sievanen H, Kannus P, Jarvinen M, Vuori I. Exercise-induced bone gain is due to enlargement in bone size without a change in volumetric bone density: a peripheral quantitative computed tomography study of the upper arms of male tennis players. Bone 2000; 27: 351-7.

74. Håheim LL, Holme I, Hjermann I, Leren P. Risk factors of stroke incidence and mortality. A 12-year follow-up of the Oslo Study. Stroke 1993; 24: 1484-9.

75. Hak AE, Pols HA, van Hemert AM, Hofman A, Witteman JC. Progression of aortic calcification is associated with metacarpal bone loss during menopause: a population-based longitudinal study. Aterioscler Thromb Vasc Biol 2000; 20: 1926-31.

76. Hamdy RC, Krishnaswamy G, Cancellaro V, Whalen K, Harvill L. Changes in bone mineral content and density after stroke. Am J Phys Med Rehabil 1993; 72: 188-91.

77. Hamdy RC, Moore SW, Cancellaro VA, Harvill LM. Long-term effects of strokes on bone mass. Am J Phys Med Rehabil 1995; 74: 351-6.

78. Hangartner TN, Gilsanz V. Evaluation of cortical bone by computed tomography. J Bone Miner Res 1996; 11; 1518-25.

79. Hara Y, Akaboshi K, Masakado Y, Chino N. Physiologic decrease of single thenar motor units in the F-response in stroke patients. Arch Phys Med Rehabil 2000; 81: 418–23.

80. Harris JE, Eng JJ. Individuals with the dominant hand affected following stroke demonstrate less impairment than those with the nondominant hand affected. Neurorehabil Neural Repair 2006; 20: 380-9.

81. Harris JE, Eng JJ. Paretic upper-limb strength best explains arm activity in people with stroke. Phys Ther 2007; 87: 88-97.

82. Hayes WC, Myers ER. Biomechanical considerations of hip and spine fractures in osteoporotic bone. Instr Course Lect 1997; 46: 431-8.

83. HDI/PulseWaveTM CR-2000 Research CardioVascular Profiling System Manual. HDI Hypertension Diagnostics Inc. 2005.

84. Holloway RG, Tuttle D, Baird T, Skelton WK. The safety of hospital stroke care. Neurology 2007; 68: 550-5.

85. Hughes VA, Frontera WR, Dallal GE, Lutz KJ, Fisher EC, Evans WJ. Muscle strength and body composition: associations with bone density in older subjects. Med Sci Sports Exerc 1995; 27: 967– 74.

86. Hyndman D, Ashburn A. Stops walking when talking as a predictor of falls in people with stroke living in the community. J Neurol Neurosurg Psychiatry 2004; 75: 994-7.

87. Hyndman D, Ashburn A, Stack E. Fall events among people with stroke living in the community: circumstances of falls and characteristics of fallers. Arch Phys Med Rehabil 2002; 83: 165-70.

88. Iversen E, Hassager C, Christiansen C. The effect of hemiplegia on bone mass and soft tissue body composition. Acta Neurol Scand 1989; 79: 155-9.

89. Iwamoto J, Takeda T, Ichimura S. Relationships between physical activity and metacarpal cortical bone mass and bone resorption in hemiplegic patients. J Orthop Sci 2001; 6: 227-33.

90. Järvinen TL, Kannus P, Sievänen H. Have the DXA-based exercise studies seriously underestimated the effects of mechanical loading on bone? J Bone Miner Res 1999; 14: 1634-5.

91. Jitapunkul S, Pillay I, Ebrahim S. The abbreviated mental test: its use and validity. Age Ageing. 1991; 20: 332-6.

92. Jørgensen L, Engstad T, Jacobsen BK. Higher incidence of falls in long-term stroke survivors than in population controls: depressive symptoms predict falls after stroke. Stroke 2002; 33: 542-7.

93. Jørgensen L, Jacobsen BK, Wilsgaard T, Magnus JH. Walking after stroke: does it matter? Changes in bone mineral density within the first 12 months after stroke. A longitudinal study. Osteoporos Int 2000; 11: 381-7.

94. Jørgensen L, Jacobsen BK. Functional status of the paretic arm affects the loss of bone mineral in the proximal humerus after stroke: a 1-year prospective study. Calcif Tissue Int 2001; 68: 11-5.

95. Kado DM, Browner WS, Blackwell T, Gore R, Cummings SR. Rate of bone loss is associated with mortality in older women: a prospective study. J Bon Miner Res 2000; 15: 1974-80.

96. Kannel WB, Wolf PA, McGee DL, Dawber TR, McNamara P, Castelli WP. Systolic blood pressure, arterial rigidity, and risk of stroke. The Framingham study. JAMA 1981; 245: 1225-9.

97. Kanis JA, Gluer CC. An update on the diagnosis and assessment of osteoporosis with densitometry. Committee of Scientific Advisors, International Osteoporosis Foundation. Osteoporos Int 2000; 11: 192-202.

98. Kanis J, Oden A, Johnell O. Acute and long-term increase in fracture risk after hospitalization for stroke. Stroke 2001; 32: 702-6.

99. Kelly BM, Pangilinan PH Jr, Rodriguez GM. The stroke rehabilitation paradigm. Phys Med Rehabil Clin N Am 2007; 18: 631-50.

100. Kontulainen SA, Johnston JD, Liu D, Leung C, Oxland TR, McKay HA. Strength indices from pQCT imaging predict up to 85% of variance in bone failure properties at tibial epiphysis and diaphysis. J Musculoskelet Neuronal Interact 2008; 8 :401-9.

101. Kontulainen S, Sievanen H, Kannus P, Pasanen M, Vuori L. Effect of long-term impact-loading on mass, size, and estimated strength of humerus and radius of female racquet-sports players: a peripheral quantitative computed tomography study between young and old starters and controls. J Bone Miner Res 2002; 17: 2281-9.

102. Kopunek SP, Michael KM, Shaughnessy M, Resnick B, Nahm ES, Whitall J, Goldberg A, Macko RF. Cardiovascular risk in survivors of stroke. Am J Prev Med 2007; 32: 408-12.

103. Kotila M, Numminen H, Waltimo O, Kaste M. Depression after stroke: results of the FINNSTROKE Study. Stroke 1998; 29: 368-72.

104. Kouwenhoven SE, Kirkevold M, Engedal K, Kim HS. Depression in acute stroke: prevalence, dominant symptoms and associated factors. A systematic literature review. Disabil Rehabil 2011; 33: 539-56.

105. Kiel DP, Kauppila LI, Cupples LA, Hannan MT, O'Donnell CJ, Wilson PW. Bone loss and the progression of abdominal aortic calcification over a 25 year period: the Framingham Heart Study. Calcif Tissue Int 2001; 68: 271-6.

106. Kingwell BA. Large artery stiffness: implications for exercise capacity and cardiovascular risk. Clin Exp Pharmacol Physiol. 2002; 29: 214-7.

107. Kiratli BJ, Smith AE, Nauenberg T, Kallfelz CF, Perkash I. Bone mineral and geometric changes through the femur with immobilization due to spinal cord injury. J Rehabil Res Dev 2000; 37: 225-33.

108. Kunkel A, Kopp B, Müller G, Villringer K, Villringer A, Taub E, Flor H. Constraint-induced movement therapy for motor recovery in chronic stroke patients. Arch Phys Med Rehabil 1999; 80: 624-8.

109. Lamb SE, Ferrucci L, Volapto S, Fried LP, Guralnik JM; Women's Health and Aging Study. Risk factors for falling in home-dwelling older women with stroke: the women's Health and Aging study. Stroke 2003; 34: 494-501.

110. Lau EM, Suriwongpaisal P, Lee JK, Das De S, Festin MR, Saw SM, Khir A, Torralba T, Sham A, Sambrook P. Risk factors for hip fracture in Asian men and women: the Asian osteoporosis study. J Bone Miner Res 2001; 16: 572-80.

111. Laurent S, Kingwell B, Bank A, Weber M, Struijker-Boudier H. Clinical applications of arterial stiffness: therapeutics and pharmacology. Am J Hypertens 2002; 15: 453–8.

112. Lazoura O, Groumas N, Antoniadou E, Papadaki PJ, Papadimitriou A, Thriskos P, Fezoulidis I, Vlychou M. Bone mineral density alterations in upper and lower extremities 12 months after stroke measured by peripheral quantitative computed tomography and DXA. J Clin Densitom 2008; 11: 511-7.

113. Lenzi GL, Altieri M, Maestrini I. Post-stroke depression. Rev Neurol (Paris). 2008; 164: 837-40.

114. Leonard MB, Shore RM. Radiologic evaluation of bone mineral in children. In: Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism, 5th ed. Favus MJ, editor. Washington, DC: American Society for Bone and Mineral Research; 2003.; 173- 89.

115. Liu-Ambrose TY, Khan KM, Eng JJ, Heinonen A, McKay HA. Both resistance and agility training increase cortical bone density in 75- to 85-year-old women with low bone mass: a 6-month randomized controlled trial. J Clin Densitom 2004; 7: 390-8.

116. Lochmüller EM, Lill CA, Kuhn V, Schneider E, Eckstein F. Radius bone strength in bending, compression, and falling and its correlation with clinical densitometry at multiple sites. J Bone Miner Res 2002; 17: 1629-38.

117. Macdonald H, Kontulainen S, Petit M, Janssen P, McKay H. Bone strength and its determinants in pre- and early pubertal boys and girls. Bone 2006; 39: 598-608.

118. MacKay-Lyons MJ, Makrides L. Exercise capacity early after stroke. Arch Phys Med Rehabil 2002; 83: 1697-702.

119. MacKay-Lyons MJ, Makrides L. Longitudinal changes in exercise capacity after stroke. Arch Phys Med Rehabil 2004; 85: 1608-12.

120. Mackintosh SF, Hill KD, Dodd KJ, Goldie PA, Culham EG. Balance score and a history of falls in hospital predict recurrent falls in the 6 months following stroke rehabilitation. Arch Phys Med Rehabil 2006; 87: 1583-9.

121. Marigold DS, Eng JJ. Altered timing of postural reflexes contributes to falling in persons with chronic stroke. Exp Brain Res 2006; 171: 459-68.

122. McComas AJ, Sica REP, Upton ARM, Aguilera N. Functional changes in motoneurons of hemiparetic patients. J Neurol, Neurosurg Psychiatry 1973; 36: 183– 93.

123. McVeigth G, Brennan G, Hayes R, Cohn J, Finelstein S, Johnston D. Vascular abnormalities in non-insulin-dependent diabetes mellitus identified by arterial waveform analysis. Am J Med 1993; 95: 424-30.

124. Melton LJ 3rd, Brown RD Jr, Achenbach SJ, O'Fallon WM, Whisnant JP. Long-Term fracture risk following ischemic stroke: a population-based study. Osteoporos Int 2001; 12: 980-6.

125. Michael KM, Allen JK, Macko RF. Reduced ambulatory activity after stroke: the role of balance, gait, and cardiovascular fitness. Arch Phys Med Rehabil 2005; 86: 1552-6.

126. Mima T, Toma K, Koshy B, Hallet M. Coherence between cortical and muscular activities after subcortical stroke Stroke 2001; 32: 2597– 601.

127. Murray CJL, Lopez AD. The global burden of disease—volume 1: a comprehensive assessment of mortality and disability from disease, injuries, and risk factors in 1990 and projected to 2020. Boston: Harvard University Press; 1996.

128. Naftchi NE, Viau AT, Marshall CH, Davis WS, Lowman EW. Bone mineralization in the distal forearm of hemiplegic patients. Arch Phys Med Rehabil 1975; 56: 487-92.

129. Nakayama H, Jorgensen HS, Raaschou HO, Olsen TS. Recovery of upper extremity function in stroke patients: the Copenhagen Stroke Study. Arch Phys Med Rehabil 1994; 75: 394–8.

130. Naves M, Rodríguez-García M, Díaz-López JB, Gómez-Alonso C, Cannata-Andía JB. Progression of vascular calcifications is associated with greater bone loss and increased bone fractures. Osteoporos Int 2008; 19: 1161-6.

131. Nguyen TV, Center JR, Eisman JA. Osteoporosis in elderly men and women: effects of dietary calcium, physical activity, and body mass index. J Bone Miner Res 2000; 15: 322-31.

132. Nichols WW, O'Rourke MF. McDonald's Blood Flow in Arteries: Theoretical, Experimental and Clinical Principles. 3rd ed. London, UK: Edward Arnold; 1990: 77–142,216–269,283–359,398–437.

133. Nidhinandana S, Sithinamsuwan P, Chinvarun Y, Wongmek W, Supakasem S, Suwantamee J. Prevalence of poststroke depression in Thai stroke survivors studied in Phramongkutklao Hospital. J Med Assoc Thai. 2010; 93 Suppl 6: S60-4.

134. Nieman DC, Lasasso H, Austin MD, Pearce S, Mclnnis T, Unick J. Validation of Cosmed's FitMate in measuring exercise metabolism. Res Sports Med 2007; 15: 67-75.

135. Nieman DC, Austin MD, Benezra L, Pearce S, McInnis T, Unick J, Gross SJ. Validation of Cosmed's FitMate in measuring oxygen consumption and estimating resting metabolic rate. Res Sports Med 2006; 14: 89-96.

136. Nyberg L, Gustafson Y. Patient falls in stroke rehabilitation. A challenge to rehabilitation strategies. Stroke1995; 26: 838-42.

137. O'Dwyer NJ, Ada L, Neilson PD. Spasticity and muscle contracture following stroke. Brain 1996; 119: 1737-49.

138. Osei-Hyiaman D, Ueji M, Toyakawa S, Takahashi H, Kano K. Influence of hand grip strength on metacarpal bone mineral density in postmenopausal Japanese women: a cross-sectional study. Calcif Tissue Int 1999; 64: 263– 6.

139. Ozdurak RH, Duz S, Arsal G, Akinci Y, Kablan N, Isikli S, et al. Quantitative forearm muscle strength influences radial bone mineral density in osteoporotic and healthy males. Tech Health Care 2003; 11: 253– 61.

140. Page SJ, Sisto S, Levine P, McGrath RE. Efficacy of modified constraint-induced movement therapy in chronic stroke: a singleblinded randomized controlled trial. Arch Phys Med Rehabil 2004; 85:14– 8.

141. Pandyan AD, Johnson GR, Price CI, Curless RH, Barnes MP, Rodgers H. A review of the properties and limitations of the Ashworth and modified Ashworth Scales as measures of spasticity. Clin Rehabil 1999; 13: 373-83.

142. Pang MY, Ashe MC, Eng JJ. Compromised bone strength index in the hemiparetic distal tibia epiphysis among chronic stroke patients: the association with cardiovascular function, muscle atrophy, mobility, and spasticity. Osteoporos Int 2010; 21: 997-1007.

143. Pang MYC, Ashe MA, Eng JJ, McKay HA, Dawson AS. A 19-week exercise program for people with chronic stroke enhances bone geometry at the tibia: a pQCT study. Osteoporos Int 2006; 17: 1615-25.

144. Pang MYC, Ashe MA, Eng JJ. Muscle weakness, spasticity and disuse contribute to demineralization and geometric changes in the radius following chronic stroke. Osteoporos Int 2007; 18: 1243-52.

145. Pang MYC, Ashe MC, Eng JJ. Tibial bone geometry in chronic stroke patients: influence of sex, cardiovascular health, and muscle mass. J Bone Miner Res 2008; 23: 1023-30.

146. Pang MYC, Eng JJ, Dawson AS. Relationship between ambulatory capacity and cardiorespiratory fitness in chronic stroke: influence of stroke-specific impairments. Chest 2005a; 127: 495-501.

147. Pang MYC, Eng JJ, McKay HA, Dawson AS. Reduced hip bone mineral density is related to physical fitness and leg lean mass in ambulatory individuals with chronic stroke. Osteoporos Int 2005b; 16: 1769-79.

148. Pang MYC, Eng JJ. Muscle strength is a determinant of bone mineral content in the hemiparetic upper extremity: implications for stroke rehabilitation. Bone 2005a; 37: 103-11.

149. Pang MYC, Eng JJ, Dawson AS, McKay HA, Harris JE. A community-based Fitness and Mobility Exercise program for older adults with chronic stroke: a randomized controlled trial. Journal of the American Geriatrics Society 2005b; 53: 1667-74.

150. Parker VM, Wade DT, Hewer RL. Lossof arm function after stroke: measurement, frequency, and recovery. Int Rehabil Med 1986; 8: 69-73.

151. Peacock M, Turner CH, Liu G, Manatunga AK, Timmerman L, Johnston CC Jr. Better discrimination of hip fracture using bone density, geometry and architecture. Osteoporos Int 1995; 5: 167-73.

152. Pfeilschifter J, Scheidt-Nave C, Leidig-Bruckner G, Woitge HW, Blum WF, Wüster C, Haack D, Ziegler R. Relationship between circulating insulin-like growth factor components and sex hormones in a population-based study of 50- to 80- year-old men and women. J Clin Endocrinol Metab 1996; 81: 2534–40.

153. Pianosi PT. Measurement of exercise cardiac output by thoracic impedance in healthy children. Eur J Appl Physiol 2004; 92: 425-30.

154. Poole KE, Reeve J, Warburton EA. Falls, fractures, and osteoporosis after stroke: time to think about protection? Stroke 2002; 33: 1432-6.

155. Prince RL, Price RI, Ho S. Forearm bone loss in hemiplegia: a model for the study of immobilization osteoporosis. J Bone Miner Res 1988; 3: 305-10.

156. Qin L, Au SK, Leung PC, Lau MC, Woo J, Choy WY, Hung WY, Dambacher MA, Leung KS. Baseline BMD and bone loss at distal radius measured by peripheral

quantitative computed tomography in peri- and postmenopausal Hong Kong Chinese women. Osteoporos Int 2002; 13: 962-70.

157. Ramnemark A, Nilsson M, Borssén B, Gustafson Y. Stroke, a major and increasing risk factor for femoral neck fracture. Stroke 2000; 31:1572-7.

158. Ramnemark A, Nyberg L, Borssen B, Olsson T, Gustafson Y. Fractures after stroke. Osteoporos Int 1998; 8: 92-5.

159. Ramnemark A, Nyberg L, Lorentzon R, Englund U, Gustafson Y. Progressive hemiosteoporosis on the paretic side and increased bone mineral density in the nonparetic arm the first year after severe stroke. Osteoporos Int 1999a; 9: 269-75.

160. Ramnemark A, Nyberg L, Lorentzon R, Olsson T, Gustafson Y. Hemiosteoporosis after severe stroke, independent of change in body composition and weight. Stroke 1999b; 30: 755-60.

161. Rekand T. Clinical assessment and management of spasticity: a review. Acta Neurol Scand Suppl 2010; 190: 62-6.

162. Richard R, Lonsdorfer-Wolf E, Charloux A, Doutreleau S, Buchheit M, Oswald-Mammosser M, Lampert E, Mettauer B, Geny B, Lonsdorfer J. Non-invasive cardiac output evaluation during a maximal progressive exercise test, using a new impedance cardiograph device. Eur J Appl Physiol 2001; 85: 202-7.

163. Riggs BL, Melton Iii LJ 3rd, Robb RA, Camp JJ, Atkinson EJ, Peterson JM, Rouleau PA, McCollough CH, Bouxsein ML, Khosla S. Population-based study of age and sex differences in bone volumetric density, size, geometry, and structure at different skeletal sites. J Bone Miner Res 2004; 19: 1945-54.

164. Robling AG, Hinant FM, Burr DB, Turner Ch. Improved bone structure and strength after long-term mechanical loading is greatest if loading is separated into short bouts. J Bone Miner Res 2002; 17: 1545–54.

165. Rodriguez-Garcia M, Naves-Diaz M, Andia JBC. Bone metabolism, vascular calcifications and mortality: Associations beyond mere coincidence. J Nephrol 2005; 18: 458-63.

166. Roth EJ. Heart disease in patients with stroke: incidence, impact, and implications for rehabilitation. Part I: classification and prevalence. Arch Phys Med Rehabil 1993; 74: 752-60.

167. Rosa J, Strauch B, Petrák O, Pikus T, Holaj R, Zelinka T, Wichterle D, Widimský J Jr. Relationship between clinical, 24-hour, average day-time and night-time blood pressure and measures of arterial stiffness in essential hypertension. Physiol Res 2008; 57: 303-6.

168. Rosen CJ, Kiel DP. Age-related osteoporosis. In: Favus MJ (ed.) Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism, 5th ed. American Society for Bone and Mineral Research, Washington, DC, USA, 2003; 89–92.

169. Rosenfalck A, Andreassen S. Impaired regulation of force and firing pattern of single motor units in patients with spasticity. J Neurol, Neurosurg Psychiatry 1980; 43: 907–16.

170. Ryan AS, Dobrovolny CL, Smith GV, Silver KH, Macko RF. Hemiparetic muscle atrophy and increased intramuscular fat in stroke patients. Arch Phys Med Rehabil 2002; 83: 1703-7.

171. Sacco RL, Adams R, Albers G, Alberts MJ, Benavente O, Furie K, Goldstein LB, Gorelick P, Halperin J, Harbaugh R, Johnston SC, Katzan I, Kelly-Hayes M, Kenton EJ, Marks M, Schwamm LH, Tomsick T. Guidelines for prevention of stroke in patients with ischemic stroke or transient ischemic attack: a statement for healthcare professionals from the American Heart Association/ American Stroke Association Council on Stroke: cosponsored by the Council on Cardiovascular Radiology and Intervention: the American Academy of Neurology affirms the value of this guideline. Stroke 2006; 37: 577-617.

172. Sackley CM. Falls, sway, and symmetry of weight-bearing after stroke. Int Disabil Stud 1991; 13: 1-4.

173. Safar ME. Pulse pressure in essential hypertension: clinical and therapeutical implications. J Hypertens 1989; 7: 769–776.

174. Sahadevan S, Lim PP, Tan NJ, Chan SP. Diagnostic performance of two mental status tests in the older chinese: influence of education and age on cut-off values. Int J Geriatr Psychiatry. 2000; 15: 234-41.

175. Sanford J, Moreland J, Swanson LR, Stratford PW, Gowland C. Reliability of the Fugl-Meyer assessment for testing motor performance in patients following stroke. Phys Ther 1993; 73: 447-454.

176. Sato Y, Kuno H, Kaji M, Ohshima Y, Asoh T, Oizumi K. Increased bone resorption during the first year after stroke. Stroke 1998; 29: 1373-7.

177. Scherhag A, Pfleger S, Garbsch E, Buss J, Sueselbeck T, Borggrefe M. Automated impedance cardiography for detecting ischemic left ventricular dysfunction during exercise testing. Kidney Blood Press Res 2005; 28: 77-84.

178. Schiessl H, Ferretti JL, Tysarczyk-Niemeyer G, Willnecker J. Nonivasive bone strength index as analyzed by peripheral quantitative computed tomography (pQCT). In: Schoenau E, editor. Paediatric osteology: new developments in diagnostics and therapy. Amsterdam: Elsevier; 1996; 141- 6.

179. Schulz E, Arfai K, Liu X, Sayre J, Gilsanz V. Aortic calcification and the risk of osteoporosis and fractures. J Clin Endocrinol Metab. 2004;89: 4246–4253.

180. Slemenda CW, Longcope C, Zhou L, Hui S, Peacock M, Johnston CC. Sex steroids and bone mass in older men. J Clin Invest 1997; 100: 1755–59.

181. Sterr A, Freivogel S, Voss A. Exploring a repetitive training regime for upper limb hemiparesis in an in-patient setting: a report on three case studies. Brain Inj 2002; 16: 1093-107.

182. Suzuki T, Sonoda S, Misawa K, Saitoh E, Shimizu Y, Kotake T. Incidence and consequence of falls in inpatient rehabilitation of stroke patients. Exp Aging Res 2005; 31: 457-69.

183. Syeda B, Gottsauner-Wolf M, Denk S, Pichler P, Khorsand A, Glogar D. Arterial compliance: a diagnostic marker for atherosclerotic plaque burden? Am J Hypertension 2003; 16: 356-62.

184. Sze KH, Wong E, Leung HY, Woo J. Falls among Chinese stroke patients during rehabilitation. Arch Phys Med Rehabil 2001; 82: 1219-25.

185. Szulc P, Seeman E, Duboeuf F, Sornay-Rendu E, Delmas PD. Bone fragility: failure of periosteal apposition to compensate for increased endocortical resorption in postmenopausal women. J Bone Miner Res 2006; 21: 1856-63.

186. Taaffe DR, Lewis B, Marcus R. Quantifying the effect of hand preference on upper limb bone mineral and soft tissue composition in young and elderly women by dual-energy X-ray absorptiometry. Clin Physiol 1994; 14: 393-404.

187. Talbot LA, Metter EJ, Fleg JL. Leisure-time physical activities and their relationship to cardiopulmonary fitness in healthy men and women 18-95 years old. Med Sci Sports Exerc 2000; 32: 417-25.

188. Taub E, Miller NE, Novack TA Cook EW 3rd, Fleming WC, Nepomuceno CS, Connell JS, Crago JE. Technique to improve chronic motor deficit after stroke. Arch Phys Med Rehabil 1993; 74: 347-54.

189. Teasell R, McRae M, Foley N, Bhardwaj A. The incidence and consequences of falls in stroke patients during inpatient rehabilitation: factors associated with high risk. Arch Phys Med Rehabil 2002; 83: 329-33.

190. Tomczak CR, Jelani A, Haennel RG, Haykowsky MJ, Welsh R, Manns PJ. Cardiac reserve and pulmonary gas exchange kinetics in patients with stroke. Stroke 2008; 39: 3102-6.

191. Towfighi A, Saver JL, Engelhardt R, Ovbiagele B. Factors associated with the steep increase in late-midlife stroke occurrence among US men. J Stroke Cerebrovasc Dis 2008; 17: 165-8.

192. Truelsen T, Piechowski-Józwiak B, Bonita R, Mathers C, Bogousslavsky J, Boysen G. Stroke incidence and prevalence in Europe: a review of available data. Eur J Neurol 2006; 13: 581-98.

193. Tutuarima JA, Van der Meulen JH, De Haan RJ, Van Straten A, Limburg M. Risk factors for falls of hospitalized stroke patients. Stroke1997; 28: 297-301.

194. Turner RT, Wakley GK, Hannon KS. Differential effects of androgens on cortical bone histomorphology in gonadectomized male and female rats. J Orthop Res 1990; 8: 612–7.

195. Ugur C, Gücüyener D, Uzuner N, Ozkan S, Ozdemir G . Characteristics of falling in patients with stroke. J Neurol Neurosurg Psychiatry 2000; 69: 649-51.

196. Uusi-Rasi K, Sievänen H, Pasanen M, Oja P, Vuori I. Maintenance of body weight, physical activity and calcium intake helps preserve bone mass in elderly women. Osteoporos Int 2001; 12: 373-9.

197. van der Lee JH, Beckerman H, Knol DL, de Vet HCW, Bouter LM. Clinimetric properties of the motor activity log for the assessment of arm use in hemiparetic patients. Stroke 2004; 35:1410–4.

198. Vico L, Collet P, Guignandon A, Lafage-Proust MH, Thomas T, Rehaillia M, Alexandre C. Effects of long-term microgravity exposure on cancellous and cortical weight-bearing bones of cosmonauts. Lancet 2000; 355: 1607-11.

199. Vogt MT, Cauley JA, Kuller LH, Nevitt MC. Bone mineral density and blood flow to the lower extremities: the study of osteoporotic fractures. J Bone Miner Res 1997; 12: 283–9.

200. Wang JS, Yang CF, Liaw MY, Wong MK. Suppressed cutaneous endothelial vascular control and hemodynamic changes in paretic extremities with edema in the extremities of patients with hemiplegia. Arch Phys Med Rehabil 2002; 83: 1017-23.

201. Washburn RA, Smith KW, Jette AM, Janney CA. The Physical Activity Scale for the Elderly (PASE): Development and Evaluation. J Clin Epidemiol 1993; 46: 153–162.

202. Watkins CL, Leathley MJ, Gregson JM, Moore AP, Smith TL, Sharma AK. Prevalence of spasticity post stroke. Clin Rehabil 2002; 16: 515-22.

203. Weerdesteyn V, de Niet M, van Duijnhoven HJ, Geurts AC. Falls in individuals with stroke. J Rehabil Res Dev 2008; 45: 1195-213.

204. White HC. Post-stroke hip fractures. Arch Orthop Trauma Surg 1988; 107: 345-7.

205. Whitney C, Warburton ER, Frohlich J, Chan SY, McKay H, Khan K. Are cardiovascular disease and osteoporosis directly linked? Sports Med 2004; 34: 779-807.

206. Whitson HE, Pieper CF, Sanders L, Horner RD, Duncan PW, Lyles KW. Adding injury to insult: Fracture risk after stroke in veterans. J Am Geriatr Soc 2006; 54: 1082-88.

207. Wilhelm G, Felsenberg D, Bogusch G. Biomechanical examinations for validation of the Bone Strength-Strain Index SSI, calculated by peripheral quantitative computed tomography. In: Lyrithis GP, editor. Musculoskeletal Interactions. Vol II. Athens: Hylonome 1999: 105-11.

208. Wilmet E, Ismail AA, Heilporn A, Welraeds D, Bergmann P. Longitudinal study of the bone mineral content and of soft tissue composition after spinal cord section. Paraplegia 1995; 33: 674-7.

209. Wilson AM, O'Neal D, Nelson CL, Prior DL, Best JD, Jenkins AJ. Comparison of arterial assessments in low and high vascular disease risk groups. Am J Hypertension 2004; 17: 285-91.

210. Winer N, Sowers JR, Weber MA. Gender differences in vascular compliance in young, healthy subjects assessed by pulse contour analysis. J Clin Hypertens (Greenwich) 2001; 3: 145-52.

211. Wong KS, Huang YN, Gao S, Lam WW, Chan YL. Cerebrovascular disease among Chinese populations--recent epidemiological and neuroimaging studies. Hong Kong Med J 2001; 7: 50-7.

212. World Health Organization (WHO): Assessment of Fracture Risk and Its Application to Screening for Postmenopausal Osteoporosis: Report of a WHO Stud y Group. WHO Technical Report Series, Report No. 843. Geneva, WHO, 1994

213. Yavuzer G, Ataman S, Sulder N, Mesut A. Bone mineral density in patients with stroke. Int J Rehabil Res 2002; 25: 235-9.

214. Yelnik AP, Simon O, Parratte B, Gracies JM. How to clinically assess and treat muscle overactivity in spastic paresis. J Rehabil Med 2010; 42: 801-7.

215. Zimlichman R, Shargorodsky M, Boaz M, Duprez D, Rahn KH, Rizzoni D, Payeras AC, Hamm C, McVeigh G. Determination of arterial compliance using blood pressure waveform analysis with the CR-2000: reliability, repeatability, and establishment of normal values for healthy European population – the Seven European Sites Study (SESS). Am J Hypertension 2005; 18: 65-71.

Appendix 1

Abbreviated Mental Test (Hong Kong version)

	Scores
1. Age (+/- 5 years)	0 / 1
2. Time (nearest hour, or a, p, n)	0 / 1
3. Address for recall at the end of the test: 42 Shanghai Street	0 / 1
4. Year (+/- 1 year)	0 / 1
5. Place name	0 / 1
6. Recognition of two persons (doctor, nurse)	0 / 1
7. Date of birth (day and month)	0 / 1
8. Date of mid-Autumn festival	0 / 1
9. Name of present Governor or Chinese leader	0 / 1
10. Count 20 - 1 backwards	0 / 1
Total Scores	____

Communication barriers present at the time of the test:—Y / N

Deafness _____ Depression ____ Dysphasia _____
Language barriers _____

(Others: _____)

Appendix 2

THE HONG KONG
POLYTECHNIC UNIVERSITY
香港理工大學

MEMO

To : PANG Marco Yiu Chung, Department of Rehabilitation Sciences
From : NG Yin Fat, Chairman, Departmental Research Committee, Department of Rehabilitation Sciences

Ethical Review of Research Project Involving Human Subjects

I write to inform you that approval has been given to your application for human subjects ethics review of the following research project for a period from 17/05/2010 to 01/05/2012:

Project Title : Bone density and macrostructure of the radius in chronic stroke patients: relationship to muscle function and cardiovascular health.

Department : Department of Rehabilitation Sciences

Principal Investigator : PANG Marco Yiu Chung

Please note that you will be held responsible for the ethical approval granted for the project and the ethical conduct of the research personnel involved in the project. In the case the Co-PI has also obtained ethical approval for the project, the Co-PI will also assume the responsibility in respect of the ethical approval (in relation to the areas of expertise of respective Co-PI in accordance with the stipulations given by the approving authority).

You are responsible for informing the Departmental Research Committee Department of Rehabilitation Sciences in advance of any changes in the research proposal or procedures which may affect the validity of this ethical approval.

You will receive separate notification should you be required to obtain fresh approval.

NG Yin Fat
Chairman
Departmental Research Committee
Department of Rehabilitation Sciences

Appendix 3A

The Hong Kong Polytechnic University

Department of Rehabilitation Sciences

Research of Stroke Subjects Informed Consent Form

Project title: Bone density and macrostructure of the radius in chronic stroke patients: relationship to muscle function and cardiovascular health.

Investigator(s):

- Ms. Qun Cheng
- Mphil student, Department of Rehabilitation Science, The Hong Kong Polytechnic University
- Dr. Marco Yiu Chung Pang (PhD)
  Associate Professor, Department of Rehabilitation Science, The Hong Kong Polytechnic University
- Prof. Alice Jones (PhD)
  Professor, Department of Rehabilitation Science, The Hong Kong Polytechnic University
- Dr. Darren Warburton (PhD)
  Associate Professor, University of British Columbia, Vancouver, Canada

Project information: Previous studies have suggested a link between cardiovascular disease and bone loss. We would like to determine the relationship between bone health and cardiovascular function following stroke.

You will be assessed once. Physical measurements will take place in the Hong Kong Polytechnic University while bone scanning will take place in the Jockey Club Centre for Osteoporosis Care and Control. You will undergo the following outcome measurement:

Bone density

pQCT: You will undergo bone scans called Peripheral Quantitative Computed Tomography (pQCT) to measure your bone structure in your radius bone on both side. This technique will give us additional information on your bone structure and bone strength. This procedure will take about 25 minutes.

DXA: You will also undergo bone scans by Dual-energy X-ray absorptiometry (DXA) to measure the bone mineral density of your forearm on each side. This scan will take about 20 minutes.

Vascular Elasticity

You will be lying on your back. We will place a blood pressure cuff on the upper-arm and a sensor placed over the opposite wrist for measuring the integrity of the vascular system.

Cardiac Function

You will be required to walk along a corridor and cover as much distance as you can in 6 minutes. We will use an electronic device to measure your heart rate, stroke volume and cardiac output. This will involve placing a few electrodes on your chest. You should not feel any discomfort or pain.

Secondary Outcome measurements

Upper Extremity Muscle Strength: We will assess the strength of your upper extremity muscles by using a hand-held dynamometer. We will ask you to contract your arm or hand muscles as hard as you can and maintain it for 5 seconds.

Recovery of Motor Skills: We will ask you to perform some specific movement patterns using your affected arm. This is to assess the recovery of your arm function. This part takes about 20 minutes.

Spasticity: We will test the level of spasticity of your muscles. You will relax and we will perform some passive movements in the wrist on the paretic side.

Sensory impairment: We will carry out a few simple tests to assess your sensation to light touch. With your eyes closed, the monofilament will be applied to your hand and fingers in random order.

Benefits and risks of undertaking this study:

There are no known risks associated with these measurements. The major benefit from participating in this study is that you will have the opportunity to know your bone health status and recovery of your arm function. The transportation cost involved for the assessment sessions will be reimbursed (a maximum of $100.00 per assessment).

Confidentiality:

All information and data collected from this study will be treated in strict confidence. Your name and personal data will not be disclosed to anyone except the project investigators.

Consent:

I, _____, have been explained the details of this study. I voluntarily consent to participate in this study. I understand that I can withdraw from this study at any time without giving reasons, and my withdrawal will not lead to any punishment or prejudice against me. I am aware of any potential risk in joining this study. I also understand that my personal information will not be disclosed to people who are not related to this study and my name will not appear on any publications resulted from this study. I also understand that the video taken on me will be edited and used for educational purpose and for conference presentation.

I can contact the chief investigator, Dr Marco Pang at telephone 2766 7156 for any questions about this study. If I have complaints related to the investigator(s), I can contact Mrs Michelle Leung, secretary of Departmental Research Committee, at 2766 5397. I know I will be given a signed copy of this consent form.

Signature (subject): _____ **Date:** _____

Signature (witness): _____ **Date:** _____

Appendix 3B

香港理工大學康復治療科學系

中風康復治療科研項目參加者同意書

科研題目：中風後橈骨的機械性能與血流動力狀態及血管彈性的關聯。

科研人員：

- 程群
- 香港理工大學，康復治療科學系碩士研究生
- 彭耀宗博士
  香港理工大學，康復治療科學系副教授
- 鍾斯何綺文教授
  香港理工大學，康復治療科學系教授
- Dr. Darren Warburton
  加拿大溫哥華英屬哥倫比亞大學副教授

科研內容：以往研究顯示心血管疾病與骨質流失息息相關。是項研究目的在探討中風後骨骼健康跟心血管功能的關聯。

研究人員將會收集中風病人在復康過程中不同時期的數據作分析。體格檢查將會在香港理工大學進行， 而骨質密度檢查將會在賽馬會骨質疏鬆預防及治療中心進行。

參與研究人仕將接受下列檢查：

骨質密度

pQCT ：我們會以肢體定量計算機斷層掃描骨質密度儀（pQCT）量度雙手手腕橈骨的骨質密度。整個素描的過程需時大約 25 分鐘。

DXA ：我們會以雙能量 X 光骨質密度儀（DXA）量度雙手前臂骨質密度。素描過程需時大約 20 分鐘。

血管彈性

首先，我們會要求你放鬆仰臥，為你的上臂量度血壓，另一組感應器同時測量另一手腕處的血流。

心臟功能

測試之前我們會先在你上身胸前貼上感應器，並為你佩上一個小型電子儀器，用作心臟功能分析；然後請你在走廊裏來回步行六分鐘，記錄你的最多步行距離。以上程序應不會令你感到不適。

其他肢體能力測量

上肢肌肉強度：我們會使用肌力計，測試你的肩肘及手部的肌肉力量。我們會要求你以最大的力量持續 5 秒緊握肌力計，每組的肌肉力量測試需要重複量度 3 次，每次量度之間會稍作休息。

肌動技能：你需要按指示以弱側上肢做出一套指定動作，目的是要測試你的肌動技能受損程度。過程需時約 20 分鐘。

痙攣：我們會在你完全放鬆的情況下，進行數項肢體動作來測試你弱側手腕肌肉的痙攣程度。

感官：我們會要求你在閉眼的情況下，進行數項簡單測試來了解你手部的輕觸覺。

對參與人士的益處或潛在危險性：
本研究並無任何其他已知的潛在損害或危險性。透過參與本研究，參加者能知道自己的骨質密度及肢體的康復情況。參與是項研究之相關交通費用將可得津貼（最高款項為港幣一百元）。

保密性：

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參加者同意書

本人_____已瞭解此次研究的具體情況。本人願意參加是項計劃，並有權在任何時候、不論任何原因放棄參與此項計劃，而此舉不會導致我受到任何懲罰或不公平對待。本人明白參與此項計劃的潛在危險性以及本人的資料將不會洩露給與此計劃無關的人員，我的名字不會出現在任何影帶或出版物上。本人亦明白製作人員可剪輯本人之訪問或錄音或錄影片段，而片段將製作成教學用具，作為教學用途或於學術會議中播放。

本人可以用電話 2766 7156 來聯絡此計劃負責人彭耀宗博士。若本人對此計劃之研究人員有任何投訴，可以聯絡部門科研委員會秘書梁女士(電話: 2766 5397)。本人亦明白，參與此計劃需要本人簽署一份同意書。

簽名（參與者） ：_____ 日期 ：_____

簽名（證人） ：_____ 日期 ：_____

Appendix 4

Physical Activity Scale for the Elderly (PASE)

以下幾條問題喺問你喺過去７日內嘅體能活動。如果喺過去７日因為唔舒服或者天氣唔好而影響你往常嘅活動，請你根據兩三個星期前嘅活動嚟做估計。

1. 喺過去７日，你用咗幾多時間嚟做一啲坐喺度嘅活動，例如閱讀、睇電視、做手工藝、打麻雀、捉棋、玩啤牌、玩電腦等？

○冇　○好少(1 至 2 日)　○有時(3 至 4 日)　○經常(5 至 7 日)

直去第 2 題　係啲乜嘢活動呢？_____

你每日平均會用幾多個鐘頭嚟做呢啲坐喺度嘅活動？

○少過 1 個鐘　○1 至 2 個鐘　○2 至 4 個鐘　○多過 4 個鐘

2. 喺過去７日，你通常用幾多時間喺屋外行路(唔理為咗乜嘢原因)？例如：去玩或者做運動、行路返工、帶狗散步、去買餸、掉垃圾、去飲茶、去行街等？

○冇　○好少(1 至 2 日)　○有時(3 至 4 日)　○經常(5 至 7 日)

直去第 3 題　係啲乜嘢活動呢？_____

你每日平均會用幾多個鐘頭嚟行路？

○少過 1 個鐘　○1 至 2 個鐘　○2 至 4 個鐘　○多過 4 個鐘

3. 喺過去７日，你用咗幾多時間嚟做一啲輕量嘅運動或者消遣嘅活動？例如：打保齡球、打高爾夫球(乘車)、在碼頭或坐船釣魚、耍太極、氣功、打乒乓球或者其他類似嘅活動。

○冇　○好少(1 至 2 日)　○有時(3 至 4 日)　○經常(5 至 7 日)

直去第 4 題　係啲乜嘢活動呢？_____

你每日平均會用幾多個鐘頭嚟做一啲輕量嘅運動或者消遣嘅活動？

○少過 1 個鐘　○1 至 2 個鐘　○2 至 4 個鐘　○多過 4 個鐘

4. 喺過去７日，你用咗幾多時間嚟做一啲溫和嘅運動同消閒活動，例如：網球雙打、羽毛球、社交舞、高爾夫球(冇乘車)、拎重嘢行平路(少過 5 公斤)或者做其他類似嘅活動？

○冇　○好少(1 至 2 日)　○有時(3 至 4 日)　○經常(5 至 7 日)

直去第 5 題　係啲乜嘢活動呢？_____

你每日平均會用幾多個鐘頭嚟做呢啲溫和嘅運動同消閒活動？

○少過 1 個鐘　○1 至 2 個鐘　○2 至 4 個鐘　○多過 4 個鐘

5. 喺過去7日，你用咗幾多時間嚟做一啲劇烈嘅運動同消閒活動，例如：跑步、游泳、踩單車、網球單打、跳健康舞、咩背囊行山、壁球、籃球、健身單車、划船、拎重嘢上樓梯(例如一包5公斤嘅米)或者做其他類似嘅活動？

○冇　○好少(1 至2日)　○有時(3 至4日)　○經常(5至7日)

直去第6題　係啲乜嘢活動呢？_____

你每日平均會用幾多個鐘頭嚟做呢啲劇烈嘅運動同消閒活動？

○少過 1個鐘　○1 至2個鐘　○2 至4個鐘　○多過 4個鐘

6. 喺過去7日，你用咗幾多時間嚟特登做一啲增強肌肉力量同持久力嘅運動？例如：舉重、掌上壓或者其他類似嘅活動。

○冇　○好少(1 至2日)　○有時(3 至4日)　○經常(5至7日)

直去第7題　係啲乜嘢活動呢？_____

你每日平均會用幾多個鐘頭嚟做一啲增強肌肉力量同持久力嘅運動？

○少過 1個鐘　○1 至2個鐘　○2 至4個鐘　○多過 4個鐘

7. 喺過去7日，你有冇做過一啲輕巧嘅家務，例如：打掃或者洗碗、手洗、熨、晾衫、煮飯、買餸？　○有　○冇

8. 喺過去7日，你有冇做過一啲粗重嘅家務或者雜務，例如：吸塵、擦地板、拖地、洗窗、洗車、搬傢俬或者石油氣？　○有　○冇

9. 喺過去7日，你有冇做以下任何嘅活動？(請答有或者冇)

A. 家居維修，例如：油漆油、貼牆紙、整電器等等　○有　○冇

B. 草地或者庭院工作，例如：剪草、掃樹葉、斬木等　○有　○冇

C. 戶外園藝　○有　○冇

D. 照顧其他人，例如：小孩、配偶、或者其他成人　○有　○冇

10. 喺過去7日，你有冇做工(包括有支薪水或係義工)？

○有　○冇

過去一個星期，你做咗幾多個鐘頭有支薪水嘅工作或者係義工 [ ][ ] 小時

以下邊一個類別最好用嚟形容你做工時(受薪或係義工)，所需要嘅體能活動？

○ 坐喺度為主，只有少量嘅手部活動
(例如：文員、手錶匠、生產線工人、巴士司機等。)

○ 坐或者企，有時需要行
(例如：收銀員、文員、技工、信差)

○ 需要行，有時要搬唔超過50磅重嘅嘢
(例如：郵差、侍應、建築工人、重機械工人)

○ 需要行同經常要搬超過50磅重嘅嘢
(例如：建築工人、搬運工人、農夫、漁夫、礦工、修路工人)

Appendix 5

Fugl Meyer Motor Assessment (Upper Extremity)

TEST	SCORING CRITERIA	SCORE
I. Reflexes	0 - no reflex 2 - reflex elicited	
Biceps		
Triceps		
IIa. Flexor Synergy	Contralateral knee to ear 0 - cannot be performed 1 – performed partly 2 – performed faultlessly	
Elevation		
Retraction		
Abduction (at least 90°)		
External Rotation		
Elbow Flexion		
Forearm Supination		
IIb. Extensor Synergy	Ear to contralateral knee.(outside) 0 - cannot be performed 1 - performed partly 2 - performed faultlessly	
Adduction/Internal Rotation		
Elbow Extension		
Forearm Pronation		
III. Mixing Synergies		
Hand to Lumbar spine	0 - No specific action performed 1 - Hand passes anterior superior illiac spine 2 - Action is performed faultlessly	
Shoulder Flexion to 90°, elbow at 0°	0 - Arm immediately abducted or elbow flexes 1 - Abduction or elbow flexion occurs late in motion 2 - Faultless motion	
Pronation/Supination of forearm with elbow at 90° and shoulder at 0°	0 - Incorrect position and/or no pronation/supination 1 - Correct position with minimal pronantion/supination 2 - Correct position and complete pronation and supination	
IV. Out of Synergy		
Shoulder abduction to 90°, elbow at 0° and forearm pronated	0 - Initial elbow flexion or deviation from pronated forearm 1 - Motion performed partly or if during motion elbow is flexed or forearm not kept in pronation 2 - Faultless motion	
Shoulder flexion, 90° - 180°, elbow at 0°, and forearm in midposition	0 - Initial flexion of elbow or shoulder abduction occurs 1 - Elbow flexion or shoulder abduction, occurs during shoulder flexion 2 - Faultless motion	
Pronation/Supination of forearm elbow at 0° and shoulder between 30° - 90° of flexion	0 - Supination/Pronation not possible or elbow and shoulder position cannot be attained 1 - Elbow and shoulder properly positioned, pronation/supination limited 2 - Faultless motion	
V Normal Reflex Activity	***Only evaluated if stage IV has a score of 6***	
Biceps and/or finger flexors and triceps	0 - at least 2 of the 3 reflexes are hyperactive 1 - one reflex is hyperactive or 2 reflexes are lively 2 - no more than one reflex is lively and none are hyperactive	
VI. Wrist		
Stability, elbow 90°, shoulder 0°	0 - Cannot dorsiflex wrist to required 15° 1 - Dorsiflexion is accomplished, but no resistance is taken 2 - Position can be maintained with some resistance	
Stability, elbow 0°, shoulder 30°		
Flex/Ext elbow 90°, shoulder 30°	0 - Volitional movement does not occur 1 - Cannot actively move wrist joint through out total ROM 2 - Faultless smooth movement	
Flex/Ext elbow 90°, shoulder 0°		

Circumduction	0 - Cannot be performed 1 - Jerky or incomplete circumduction 2 - Complete motion with smoothness	
VII. Hand		
Finger mass flexion	0 - No flexion occurs 1 - Some flexion, but not full motion 2 - Complete active flexion (compared with unaffected hand)	
Finger Mass Extension	0 - No extension occurs 1 - Patient can release an active mass flexion grasp 2 - Full active extension	
G1: MP joints ext and PIPs & DIPs flexed.	0 - Required position cannot be performed 1 - Grasp is weak 2 - Grasp maintained against reasonable resistance	
G 2: Adduct thumb, IP & MP 0°	0 - Function cannot be performed 1 - Paper (can, ball) can be held in place but not against a tug 2 - Paper (can, ball) is held against tug	
G 3: Thumb opposes index finger		
G 4: Grasp can		
G 5: Grasp tennis ball		
VIII. Co-ordination/Speed		
Tremor - Finger to nose	0 - Marked tremor 1 - Slight tremor 2 - No tremor	
Dysmetria - Finger to nose	0 - Pronounced or unsystematic dysmetria 1 - Slight or pronounced dysmetria 2 - No dysmetria	
Speed - Finger to nose	0 - Activity is more than 6 seconds longer than unaffected hand 1 - 2 - 5 seconds longer than affected hand 2 - less than 2 seconds	
Total		

Appendix 6

Appendix 7

Modified Ashworth Scale (MAS)

Description	Grade
No increase in muscle tone	**0**
Slight increase in muscle tone, manifested by a catch and release or by minimal resistance at the end of the ROM when the affected part is moved in flexion or extension	**1**
Slight increase in muscle tone, manifested by a catch, followed by minimal resistance throughout the remainder (less than half) of ROM	**1+** **(or 1.5 for statistical purpose)**
More marked increase in muscle tone through most of ROM, but affected part easily moved	**2**
Considerable increase in muscle tone, passive movement difficult	**3**
Affect part rigid in flexion or extension	**4**

Appendix 8

Arterial Elasticity Guideline Tables

MALE	C1 – Large Artery Elasticity Index Range			C2 – Small Artery Elasticity Index Range		
Age Range	Abnormal	Borderline	Normal	Abnormal	Borderline	Normal
15 - 19	< 10	10 – 17	> 17	< 6	6 – 9	> 9
20 - 29	< 9	9 – 16	> 16	< 6	6 – 8	> 8
30 - 39	< 8	8 – 14	> 14	< 6	6 – 8	> 8
40 - 49	< 7	7 – 12	> 12	< 5	5 – 7	> 7
50 - 59	< 6	6 – 11	> 11	< 5	5 – 7	> 7
60 - 69	< 5	5 – 10	> 10	< 4	4 – 6	> 6
> 70	< 5	5 – 9	> 9	< 4	4 – 5	> 5

FEMALE	C1 – Large Artery Elasticity Index Range			C2 – Small Artery Elasticity Index Range		
Age Range	Abnormal	Borderline	Normal	Abnormal	Borderline	Normal
15 - 19	< 9	9 – 15	> 15	< 6	6 – 8	> 8
20 - 29	< 8	8 – 14	> 14	< 5	5 – 7	> 7
30 - 39	< 7	7 – 12	> 12	< 4	4 – 6	> 6
40 - 49	< 6	6 – 10	> 10	< 4	4 – 6	> 6
50 - 59	< 5	5 – 10	> 10	< 3	3 – 5	> 5
60 - 69	< 4	4 – 9	> 9	< 3	3 – 5	> 5
> 70	< 4	4 – 8	> 8	< 2	2 – 4	> 4

Appendix 9

Checking multicollinearity: Correlation among variables of neuromuscular and cardiovascular function

Parameter	MAS	MAL	FMA	Grip Strength	CI	SI	VO_2	C_1
MAL	-0.646*							
FMA	-0.761*	0.782*						
Grip Strength	-0.643*	0.607*	0.744*					
CI	0.131	-0.083	-0.129	-0.124				
SI	0.148	-0.123	-0.120	-0.023	0.720*			
VO_2	-0.026	0.126	0.198	0.223	0.195	0.084		
C_1	-0.317	0.301*	0.119	0.326*	-0.151	0.218	-0.178	
C_2	-0.310*	0.299*	0.314*	0.489*	0.039	0.194	0.038	0.421*

There were significant relationships among the neuromuscular variables (MAL = Motor Activity Log; FMA = Fugl-Meyer motor assessment; CI = cardiac index; SI = stroke index; VO_2 = oxygen consumption; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant (Pearson's r or Spearman's rho, $p<0.05$).

Appendix 10

Multiple regression analyses for predicting areal bone mineral density (aBMD) of the ultradistal forearm

Predictor	F	R^2 change	B	95%CI	Beta	*P*
Model 1						
Age	**9.874**	**0.481**	**-0.005**	**-0.007, -0.003**	**-0.525**	**<0.001***
Sex			**-0.092**	**-0.132 , -0.053**	**-0.474**	**<0.001***
BMI			0.000	-0.005, 0.005	0.007	0.944
Post-stroke duration			0.000	0.000, 0.001	0.048	0.640
C_1		0.067	0.000	-0.003, 0.004	0.020	0.844
C_2			-0.002	-0.009, 0.005	-0.072	0.542
MAS			**-0.072**	**-0.123, -0.022**	**-0.289**	**0.005***
Model 2						
Age	**8.341**	**0.481**	**-0.005**	**-0.007, -0.003**	**-0.523**	**<0.001***
Sex			**-0.085**	**-0.129, -0.041**	**-0.436**	**<0.001***
BMI			0.000	-0.005, 0.006	0.016	0.870
Post-stroke duration			4.207E-5	0.000, 0.001	0.018	0.868
C_1		0.025	0.001	-0.003, 0.005	0.038	0.727
C_2			-0.002	-0.009, 0.005	-0.058	0.637
FMA			0.001	0.000, 0.002	0.192	0.099
Model 3						
Age	**8.689**	**0.481**	**-0.005**	**-0.007, -0.003**	**-0.522**	**<0.001***
Sex			**-0.091**	**-0.132, -0.049**	**-0.467**	**<0.001***
BMI			0.001	-0.004, 0.006	0.035	0.724
Post-stroke duration			2.751E-5	0.000, 0.001	0.012	0.909
C_1		0.035	0.000	-0.004, 0.004	-0.017	0.878
C_2			-0.002	-0.008, 0.005	-0.057	0.639
MAL			**0.012**	**0.000, 0.024**	**0.214**	**0.048***
Model 4						
Age	**8.933**	**0.481**	**-0.005**	**-0.006, -0.003**	**-0.494**	**<0.001***
Sex			**-0.068**	**-0.116, -0.019**	**-0.348**	**0.007***
BMI			0.000	-0.006, 0.005	-0.010	0.916
Post-stroke duration			6.378E-5	0.000, 0.001	0.028	0.793
C_1		0.042	0.000	-0.004, 0.004	0.008	0.943
C_2			-0.002	-0.009, 0.005	-0.077	0.530
Grip strength			**0.003**	**0.000, 0.005**	**0.299**	**0.030***

MAS, MAL, and grip strength were independently associated with ultradistal forearm aBMD after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 11

Multiple regression analyses for predicting areal bone mineral density (aBMD) of mid-forearm

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**16.220**	**0.592**	**-0.006**	**-0.008, -0.003**	**-0.468**	**<0.001***
Sex			**-0.134**	**-0.178, -0.089**	**-0.530**	**<0.001***
BMI			0.000	-0.006, 0.006	-0.008	0.918
Post-stroke duration			0.000	0.000, 0.001	0.135	0.128
C_1		0.073	0.002	-0.002, 0.006	0.081	0.364
C_2			0.002	-0.006, 0.009	0.046	0.648
MAS			**-0.091**	**-0.147, -0.035**	**-0.281**	**0.002***
Model 2						
Age	**13.720**	**0.592**	**-0.006**	**-0.008, -0.003**	**-0.467**	**<0.001***
Sex			**-0.124**	**-0.173, -0.074**	**-0.491**	**<0.001***
BMI			2.609E-5	-0.006, 0.006	0.001	0.993
Post-stroke duration			0.000	0.000, 0.001	0.099	0.299
C_1		0.035	0.002	-0.002, 0.007	0.099	0.299
C_2			0.002	-0.006, 0.010	0.059	0.582
FMA			0.001	0.000, 0.002	0.191	0.059
Model 3						
Age	**13.538**	**0.592**	**-0.005**	**-0.008, -0.003**	**-0.464**	**<0.001***
Sex			**-0.133**	**-0.181, -0.086**	**-0.529**	**<0.001***
BMI			0.001	-0.006, 0.007	0.017	0.847
Post-stroke duration			0.000	0.000, 0.001	0.088	0.343
C_1		0.033	0.001	-0.003, 0.006	0.051	0.592
C_2			0.002	-0.005, 0.010	0.064	0.547
MAL			0.012	-0.001, 0.026	0.171	0.073
Model 4						
Age	**14.674**	**0.592**	**-0.005**	**-0.007, -0.003**	**-0.438**	**<0.001***
Sex			**-0.102**	**-0.157, -0.048**	**-0.406**	**<0.001***
BMI			-0.001	-0.007, 0.005	-0.025	0.767
Post-stroke duration			0.000	0.000, 0.001	0.116	0.205
C_1		0.051	0.002	-0.003, 0.006	0.069	0.458
C_2			0.002	-0.006, 0.009	0.041	0.695
Grip strength			**0.003**	**0.001, 0.006**	**0.293**	**0.014***

MAS and grip strength were independently associated with mid-forearm aBMD after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 12

Multiple regression analyses for predicting areal bone mineral density (aBMD) of 1/3 forearm

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**14.998**	**0.573**	**-0.004**	**-0.007, -0.002**	**-0.337**	**<0.001***
Sex			**-0.161**	**-0.211, -0.112**	**-0.585**	**<0.001***
BMI			0.001	-0.006, 0.008	0.025	0.763
Post-stroke duration			0.000	0.000, 0.001	0.127	0.163
C_1		0.075	0.004	-0.001, 0.009	0.153	0.098
C_2			0.001	-0.008, 0.009	0.013	0.904
MAS			**-0.093**	**-0.155, -0.030**	**-0.260**	**0.005***
Model 2						
Age	**13.034**	**0.573**	**-0.004**	**-0.007, -0.002**	**-0.336**	**0.001***
Sex			**-0.152**	**-0.206, -0.097**	**-0.549**	**<0.001***
BMI			0.001	-0.006, 0.008	0.034	0.700
Post-stroke duration			0.000	0.000, 0.001	0.102	0.293
C_1		0.042	0.004	-0.001, 0.009	0.170	0.082
C_2			0.001	-0.008, 0.010	0.024	0.823
FMA			0.001	0.000, 0.003	0.178	0.082
Model 3						
Age	**12.290**	**0.573**	**-0.004**	**-0.007, -0.002**	**-0.331**	**0.001***
Sex			**-0.164**	**-0.217, -0.111**	**-0.594**	**<0.001***
BMI			0.002	-0.005, 0.009	0.045	0.613
Post-stroke duration			0.000	0.000, 0.001	0.062	0.513
C_1		0.028	0.004	-0.002, 0.009	0.136	0.172
C_2			0.001	-0.007, 0.010	0.036	0.744
MAL			0.008	-0.008, 0.023	0.097	0.318
Model 4						
Age	**13.914**	**0.573**	**-0.004**	**-0.006, -0.002**	**-0.309**	**0.002***
Sex			**-0.129**	**-0.189, -0.068**	**-0.466**	**<0.001***
BMI			0.000	-0.006, 0.007	0.009	0.919
Post-stroke duration			0.000	0.000, 0.001	0.112	0.230
C_1		0.058	0.004	-0.001, 0.009	0.141	0.136
C_2			0.000	-0.008, 0.009	0.007	0.949
Grip strength			**0.003**	**0.001, 0.006**	**0.280**	**0.021***

MAS and grip strength were independently associated with 1/3 forearm aBMD after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 13

Multiple regression analyses for predicting areal bone mineral density (aBMD) of the total forearm

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**16.908**	**0.600**	**-0.005**	**-0.007, -0.003**	**-0.455**	**<0.001***
Sex			**-0.130**	**-0.171, -0.089**	**-0.553**	**<0.001***
BMI			0.000	-0.005, 0.006	0.009	0.913
Post-stroke duration			0.000	0.000, 0.001	0.128	0.143
C_1		**0.075**	**0.002**	**-0.002, 0.006**	**0.089**	**0.312**
C_2			**0.001**	**-0.006, 0.008**	**0.022**	**0.823**
MAS			**-0.087**	**-0.138, -0.035**	**-0.286**	**0.001***
Model 2						
Age	**14.148**	**0.600**	**-0.005**	**-0.007, -0.003**	**-0.453**	**<0.001***
Sex			**-0.121**	**-0.167, -0.075**	**-0.514**	**<0.001***
BMI			0.001	-0.005, 0.006	0.018	0.831
Post-stroke duration			0.000	0.000, 0.001	0.100	0.290
C_1		0.034	0.002	-0.002, 0.007	0.107	0.256
C_2			0.001	-0.006, 0.008	0.036	0.737
FMA			0.001	0.000, 0.002	0.193	0.054
Model 3						
Age	**13.910**	**0.600**	**-0.005**	**-0.007, -0.003**	**-0.450**	**<0.001***
Sex			**-0.130**	**-0.174, -0.086**	**-0.553**	**<0.001***
BMI			0.001	-0.005, 0.007	0.034	0.692
Post-stroke duration			0.000	0.000, 0.001	0.077	0.399
C_1		0.030	0.001	-0.003, 0.006	0.060	0.527
C_2			0.001	-0.006, 0.009	0.042	0.694
MAL			0.011	-0.001, 0.024	0.167	0.077
Model 4						
Age	**15.274**	**0.600**	**-0.005**	**-0.007, -0.003**	**-0.424**	**<0.001***
Sex			**-0.100**	**-0.150, -0.050**	**-0.425**	**<0.001***
BMI			0.000	-0.006, 0.005	-0.009	0.917
Post-stroke duration			0.000	0.000, 0.001	0.110	0.224
C_1		0.052	0.002	-0.002, 0.006	0.076	0.403
C_2			0.001	-0.006, 0.008	0.017	0.871
Grip strength			**0.003**	**0.001, 0.005**	**0.302**	**0.011***

MAS and grip strength were independently associated with total forearm aBMD after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 14

Multiple regression analyses for predicting compressive bone strength index (CBSI) of the radius epiphysis

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**14.675**	**0.605**	**-0.006**	**-0.009, -0.003**	**-0.342**	**<0.001***
Sex			**-0.223**	**-0.291, -0.155**	**-0.596**	**<0.001***
BMI			0.004	-0.005, 0.013	0.078	0.354
Post-stroke duration			2.725E-5	-0.001, 0.001	0.006	0.946
C_1		0.038	0.001	-0.006, 0.007	0.021	0.820
C_2			0.004	-0.007, 0.015	0.072	0.494
MAS			**-0.097**	**-0.183, -0.011**	**-0.200**	**0.028***
Model 2						
Age	**14.859**	**0.605**	**-0.006**	**-0.009, -0.003**	**-0.346**	**<0.001***
Sex			**-0.201**	**-0.273, -0.130**	**-0.537**	**<0.001***
BMI			0.004	-0.005, 0.013	0.082	0.331
Post-stroke duration			0.000	-0.001, 0.001	0.025	0.790
C_1		0.041	0.002	-0.005, 0.008	0.043	0.644
C_2			0.004	-0.007, 0.015	0.069	0.506
FMA			**0.002**	**0.000, 0.004**	**0.228**	**0.022***
Model 3						
Age	**13.614**	**0.605**	**-0.006**	**-0.009, -0.003**	**-0.339**	**0.001***
Sex			**-0.222**	**-0.292, -0.152**	**-0.592**	**<0.001***
BMI			0.005	-0.004, 0.014	0.097	0.262
Post-stroke duration			-9.818E-5	-0.241, 0.810	-0.022	0.810
C_1		0.021	0.000	-0.007, 0.007	-0.003	0.975
C_2			0.004	-0.007, 0.016	0.083	0.439
MAL			0.015	-0.005, -0.035	0.138	0.144
Model 4						
Age	**16.735**	**0.605**	**-0.005**	**-0.009, -0.002**	**-0.310**	**0.001***
Sex			**-0.161**	**-0.238, -0.083**	**-0.429**	**<0.001***
BMI			0.003	-0.006, 0.011	-0.049	0.545
Post-stroke duration			0.000	-0.001, 0.001	0.038	0.663
C_1		0.068	0.000	-0.006, 0.006	0.006	0.945
C_2			0.003	-0.008, 0.013	0.046	0.645
Grip strength			**0.006**	**0.002, 0.010**	**0.364**	**0.002***

MAS, FMA, and grip strength were independently associated with CBSI of the radius epiphysis after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 15

Multiple regression analyses for predicting polar stress-strain index (p-SSI) of the radius diaphysis

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**22.842**	**0.715**	**-1.672**	**-2.799, -0.545**	**-0.233**	**0.004***
Sex			**-111.992**	**-135.792, -88.193**	**-0.731**	**<0.001***
BMI			1.688	-1.467, 4.843	0.077	0.289
Post-stroke duration			-0.028	-0.310, 0.254	-0.015	0.843
C_1		0.022	2.236	-0.057, 4.529	0.154	0.056
C_2			0.431	-3.531, 4.393	0.019	0.828
MAS			-10.310	-40.418, 19.798	-0.052	0.496
Model 2						
Age	**22.888**	**0.715**	**-1.680**	**-2.807, -0.553**	**-0.234**	**0.004***
Sex			**-109.528**	**-134.723, -84.333**	**-0.715**	**<0.001***
BMI			1.706	-1.444, 4.856	0.078	0.283
Post-stroke duration			-0.017	-0.307, 0.272	-0.010	0.905
C_1		**0.022**	**2.322**	**0.018, 4.626**	**0.160**	**0.048***
C_2			0.412	-3.549, 4.373	0.019	0.836
FMA			0.235	-0.397, 0.868	0.062	0.459
Model 3						
Age	**22.595**	**0.715**	**-1.655**	**-2.786, -0.523**	**-0.230**	**0.005***
Sex			**-112.920**	**-137.014, -88.825**	**-0.737**	**<0.001***
BMI			1.741	-1.427, 4.910	0.079	0.276
Post-stroke duration			-0.068	-0.348, 0.212	-0.038	0.627
C_1		0.020	2.252	-0.081, 4.585	0.155	0.058
C_2			0.599	-3.372, 4.570	0.027	0.764
MAL			-0.360	-7.295, 6.579	-0.008	0.918
Model 4						
Age	**28.507**	**0.715**	**-1.518**	**-2.556, -0.479**	**-0.211**	**0.005***
Sex			**-89.023**	**-115.104, -62.942**	**-0.581**	**<0.001***
BMI			1.017	-1.913, 3.947	0.046	0.490
Post-stroke duration			0.098	-0.162, 0.358	0.054	0.453
C_1		0.063	2.064	-0.047, 4.174	0.142	0.055
C_2			0.362	-4.027, 3.304	-0.016	0.844
Grip strength			**2.009**	**0.795, 3.224**	**0.304**	**0.002***

Grip strength was independently associated with p-SSI of the radius diaphysis after adjusting for relevant factors. C_1 was also a significant determinant in model 2 (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 16

Checking multicollinearity: Correlation of percent side-to-side difference of grip strength with other impairment variables

Parameter	CI	SI	VO_2	C_1	C_2	MAS	MAL	FMA
%Δ grip strength	-0.164	-0.146	0.085	0.148	**0.346***	**-0.736***	**0.680***	**0.820***

The percent side-to-side difference in grip strength was significantly correlated with MAS, MAL, FMA and C_2 (%Δ = percent side-to-side difference; CI = cardiac index; SI = stroke index; VO_2 = oxygen consumption; C_1 = large artery elasticity index; C_2 = small artery elasticity index; MAS = Modified Ashworth Scale; MAL = Motor Activity Log**;** FMA = Fugl-Meyer motor assessment).
*Statistically significant (Pearson's r or Spearman's rho, $p<0.05$)

Appendix 17

Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (BMD) of the ultradistal forearm

Predictor	F	R^2 change	B	95%CI	Beta	*P*
Model 1						
Age	2.373	0.118	-0.322	-0.740, 0.095	-0.187	0.128
Sex			-1.335	-10.348, 7.678	-0.036	0.768
BMI			-0.360	-1.649, 0.929	-0.069	0.578
Post-stroke duration			-0.071	-0.168, 0.043	-0.164	0.215
MAS		0.049	-11.678	-24.217, 0.861	-0.247	0.067
Model 2						
Age	3.239	0.118	-0.298	-0.704, 0.108	-0.173	0.147
Sex			0.302	-8.596, 9.236	0.009	0.943
BMI			-0.0245	-1.493, 1.002	-0.047	0.695
Post-stroke duration			-0.054	-0.165, 0.056	-0.125	0.329
MAL		**0.097**	**3.713**	**0.962, 6.464**	**0.351**	**0.009***
Model 3						
Age	2.774	0.118	-0.344	-0.756, 0.068	-0.200	0.100
Sex			1.763	-7.815, 11.341	-0.048	0.714
BMI			-0.322	-1.590, 0.947	-0.061	0.614
Post-stroke duration			-0.056	-0.171, 0.059	-0.128	0.337
FMA		**0.072**	**0.296**	**0.037, 0.555**	**0.326**	**0.026***
Model 4						
Age	3.640	0.118	-0.401	-0.804, 0.003	-0.233	0.051
Sex			2.339	-6.807, 11.486	0.064	0.611
BMI			-0.600	-1.851, 0.651	-0.114	0.341
Post-stroke duration			-0.042	-0.153, 0.069	-0.096	0.455
%ΔGrip strength		**0.117**	**19.354**	**6.489, 32.218**	**0.417**	**0.004***

FMA, MAL, and percent side-to-side difference in grip strength were independently associated with percent side-to-side difference in ultradistal forearm aBMD after adjusting for relevant factors ((%Δ = percent side-to-side difference; B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 18

Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (aBMD) of the mid-forearm

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**6.763**	**0.259**	**-0.303**	**-0.536, -0.071**	**-0.298**	**0.011***
Sex			-4.408	-9.383, 0.567	-0.203	0.081
BMI			-0.092	-0.745, 0.562	-0.030	0.779
Post-stroke duration			-0.047	-0.105, 0.012	-0.182	0.1114
C_2		0.153	-0.268	-1.085, 0.548	-0.085	0.513
MAS			**-12.253**	**-18.579, -5.928**	**-0.438**	**<0.001***
Model 2						
Age	**6.824**	**0.259**	**-0.320**	**-0.552, -0.088**	**-0.315**	**0.008***
Sex			-1.872	-7.115, 3.371	-0.086	0.478
BMI			-0.045	-0.696, 0.606	-0.014	0.891
Post-stroke duration			-0.040	-0.100, 0.019	-0.157	0.180
C_2		0.155	-0.244	-1.057, 0.570	-0.078	0.551
FMA			**0.258**	**0.126, 0.390**	**0.480**	**<0.001***
Model 3						
Age	**5.579**	**0.259**	**-0.288**	**-0.529, -0.048**	**-0.283**	**0.020***
Sex			-3.882	-9.102, 1.338	-0.179	0.142
BMI			0.018	-0.660, 0.695	0.006	0.959
Post-stroke duration			-0.054	-0.115, 0.006	-0.211	0.078
C_2		0.107	-0.257	-1.106, 0.591	-0.082	0.546
MAL			**2.321**	**0.835, 3.808**	**0.372**	**0.003***
Model 4						
Age	**11.693**	**0.259**	**-0.402**	**-0.610, -0.195**	**-0.396**	**<0.001***
Sex			-1.625	-6.134, 2.883	-0.075	0.473
BMI			-0.282	-0.861, 0.297	-0.091	0.333
Post-stroke duration			-0.027	-0.079, 0.025	-0.105	0.299
C_2		0.289	-0.471	-1.193, 0.251	-0.150	0.197
%ΔGrip strength			**18.202**	**12.208, 24.196**	**0.663**	**<0.001***

MAS, FMA, MAL, and percent side-to-side difference in grip strength were independently associated with percent side-to-side difference in mid-forearm aBMD after adjusting for relevant factors ((%Δ = percent side-to-side difference; B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 19

Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (aBMD) of the 1/3 forearm

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**3.714**	**0.241**	**-0.385**	**-0.721, -0.049**	**-0.291**	**0.025***
Sex			-6.955	-14.048, 0.137	-0.246	0.054
BMI			-0.262	-1.202, 0.678	-0.065	0.579
Post-stroke duration			-0.056	-0.140, 0.028	-0.167	0.189
C_1		0.072	0.345	-0.338, 1.028	0.129	0.316
C_2			-0.431	-1.612, 0.749	-0.106	0.467
MAS			**-9.919**	**-18.891, -0.947**	**-0.273**	**0.031***
Model 2						
Age	**3.603**	**0.241**	**-0.390**	**-0.728, -0.052**	**-0.294**	**0.024***
Sex			-4.989	-12.538, 2.561	-0.177	0.191
BMI			-0.239	-1.183, 0.705	-0.059	0.614
Post-stroke duration			-0.051	-0.138, 0.035	-0.153	0.241
C_1		0.066	0.416	-0.274, 1.106	0.155	0.232
C_2			-0.428	-1.615, 0.759	-0.105	0.473
FMA			**0.197**	**0.007,0.386**	**0.281**	**0.042***
Model 3						
Age	**2.859**	**0.241**	**-0.375**	**-0.724, -0.027**	**-0.283**	**0.035***
Sex			-7.276	-14.700, 0.148	-0.258	0.055
BMI			-0.181	-1.158, 0.795	-0.045	0.711
Post-stroke duration			-0.080	-0.166, 0.006	-0.239	0.069
C_1		0.019	0.302	-0.416, 1.021	0.113	0.403
C_2			-0.325	-1.549, 0.899	-0.080	0.597
MAL			0.024	-0.048, 0.095	0.087	0.510
Model 4						
Age	**4.927**	**0.241**	**-0.462**	**-0.787, -0.138**	**-0.349**	**0.006***
Sex			-4.467	-11.461, 2.526	-0.158	0.206
BMI			-0.445	-1.351, 0.461	-0.110	0.309
Post-stroke duration			-0.036	-0.117, 0.046	-0.106	0.384
C_1		0.136	0.424	-0.228, 1.077	0.158	0.198
C_2			-0.638	-1.775, 0.499	-0.156	0.266
%ΔGrip strength			**15.498**	**6.238, 24.758**	**0.434**	**0.001***

MAS, FMA, and percent side-to-side difference in grip strength were independently associated with percent side-to-side difference in 1/3 forearm aBMD after adjusting for relevant factors ((%Δ = percent side-to-side difference; B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 20

Multiple regression analyses for predicting percent side-to-side difference in areal bone mineral density (aBMD) of the total forearm

Predictors	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	**5.619**	**0.273**	**-0.285**	**-0.520, -0.049**	**-0.285**	**0.019***
Sex			-4.548	-9.519, 0.423	-0.213	0.072
BMI			-0.085	-0.745, 0.574	-0.028	0.796
Post-stroke duration			-0.049	-0.108, 0.010	-0.193	0.102
C_1		0.135	0.069	-0.410, 0.548	0.034	0.774
C_2			-0.238	-1.066, 0.589	-0.077	0.566
MAS			**-11.281**	**-17.571, -4.992**	**-0.411**	**0.001***
Model 2						
Age	**6.272**	**0.273**	**-0.294**	**-0.524, -0.064**	**-0.294**	**0.013***
Sex			-1.821	-6.967, 3.325	-0.085	0.481
BMI			-0.066	-0.710, 0.577	-0.022	0.837
Post-stroke duration			-0.037	-0.096, 0.022	-0.146	0.218
C_1		0.162	0.164	-0.307, 0.634	0.081	0.488
C_2			-0.261	-1.070, 0.548	-0.085	0.521
FMA			**0.260**	**0.131, 0.389**	**0.492**	**<0.001***
Model 3						
Age	**5.123**	**0.273**	**-0.284**	**-0.524, -0.044**	**-0.284**	**0.021***
Sex			-4.066	-9.172, 1.040	-0.191	0.116
BMI			0.050	-0.622, 0.721	0.016	0.883
Post-stroke duration			-0.055	-0.114, 0.004	0.217	0.069
C_1		0.113	-0.065	-0.559, 0.430	-0.032	0.794
C_2			-0.199	-1.041, 0.642	-0.065	0.637
MAL			**0.079**	**0.030, 0.128**	**0.385**	**0.002***
Model 4						
Age	**11.337**	**0.273**	**-0.378**	**-0.579, -0.177**	**-0.378**	**<0.001***
Sex			-1.533	-5.859, 2.792	-0.072	0.481
BMI			-0.308	-0.868, 0.252	-0.101	0.276
Post-stroke duration			-0.023	-0.073, 0.027	-0.090	0.367
C_1		0.309	0.164	-0.240, 0.568	0.081	0.419
C_2			-0.494	-1.197, 0.210	-0.160	0.165
%ΔGrip strength			**18.526**	**12.798, 24.254**	**0.687**	**<0.001***

MAS, FMA, MAL, and percent side-to-side difference in grip strength were independently associated with percent side-to-side difference in total forearm aBMD after adjusting for relevant factors ((%Δ = percent side-to-side difference; B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 21

Multiple regression analyses for predicting percent side-to-side difference of compressive bone strength index (CBSI) at the radius epiphysis

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	3.960	0.247	-0.392	-1.085, 0.302	-0.142	0.263
Sex			-7.900	-22.752, 6.952	-0.134	0.291
BMI			0.636	-1.315, 2.587	0.075	0.516
Post-stroke duration			**-0.216**	**-0.390, -0.042**	**-0.309**	**0.016***
C_2		0.043	-0.011	-2.448, 2.426	-0.001	0.993
MAS			-17.674	-36.558, 1.210	-0.233	0.066
Model 2						
Age	5.635	0.247	-0.449	-1.104, 0.207	-0.162	0.176
Sex			-0.984	-15.783, 13.814	-0.017	0.895
BMI			0.678	-1.161, 2.516	0.080	0.463
Post-stroke duration			-0.160	-0.328, 0.008	-0.229	0.061
C_2		0.121	-0.120	-2.415, 2.176	-0.014	0.917
FMA			**0.619**	**0.247, 0.992**	**0.423**	**0.002***
Model 3						
Age	5.585	0.247	-0.374	-1.029, 0.281	-0.135	0.257
Sex			-5.164	-19.354, 9.026	-0.088	0.469
BMI			0.844	-0.998, 2.686	0.100	0.363
Post-stroke duration			**-0.177**	**-0.342, -0.013**	**-0.254**	**0.035***
C_2		0.119	-0.229	-2.536, 2.078	-0.027	0.843
MAL			**6.648**	**2.608, 0.688**	**0.391**	**0.002***
Model 4						
Age	9.026	0.247	-0.653	-1.255, -0.051	-0.236	0.034*
Sex			-0.191	-13.294, 12.913	-0.003	0.911
BMI			0.094	-1.588, 1.776	0.011	0.911
Post-stroke duration			-0.125	-0.275, 0.025	-0.179	0.102
C_2		0.236	-0.686	-2.785, 1.413	-0.080	0.516
%ΔGrip strength			**44.697**	**27.277, 62.117**	**0.599**	**<0.001***

FMA, MAL, and percent side-to-side difference in grip strength were independently associated with percent side-to-side difference in CBSI of the radius epiphysis after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)

Appendix 22

Multiple regression analyses for predicting percent side-to-side difference of polar stress-strain index (p-SSI) at the radius diaphysis

Predictor	F	R^2 change	B	95%	Beta	*P*
Model 1						
Age	3.611	0.221	-0.191	-0.450, 0.069	-0.176	0.147
Sex			-0.585	-6.494, 5.324	-0.025	0.844
BMI			-0.071	-0.143, 0.000	-0.260	0.050
Post-stroke duration			**-0.074**	**-0.146, -0.001**	**-0.269**	**0.047***
C_1		0.051	0.496	-0.066, 1.057	0.226	0.082
FMA			0.088	-0.069, 0.244	0.153	0.267
Model 2						
Age	3.734	0.221	-0.190	-0.448, 0.069	-0.175	0.147
Sex			-1.161	-6.657, 4.336	-0.050	0.674
BMI			-0.312	-1.088, 0.464	-0.094	0.404
Post-stroke duration			**-0.073**	**-0.141, -0.004**	**-0.265**	**0.037***
C_1		0.058	0.400	-0.166, 0.967	0.182	0.163
MAL			1.144	-0.560, 2.848	0.172	0.184
Model 3						
Age	4.693	0.221	-0.222	-0.473, 0.029	-0.205	0.083
Sex			-0.539	-5.027, 6.105	-0.023	0.847
BMI			-0.507	-1.269, 0.254	-0.153	0.187
Post-stroke duration			-0.053	-0.121, 0.015	-0.194	0.123
C_1		0.106	0.496	-0.043, 1.034	0.226	0.070
%ΔGrip strength			**9.459**	**1.783, 17.135**	**0.323**	**0.017***

Percent side-to-side difference in grip strength was independently associated with percent side-to-side difference in p-SSI of the radius diaphysis after adjusting for relevant factors (B = Unstandardized regression coefficient; Beta = Standardized regression coefficient; 95%CI = 95% confidence interval; BMI = body mass index; MAS: Modified Ashworth Scale; MAL = Motor Activity log; FMA = Fugl-Meyer motor assessment; C_1 = large artery elasticity index; C_2 = small artery elasticity index).
*Statistically significant ($p<0.05$)