CAREES Research Study Protocol

Lifestyle, anatomical, metabolic and genetic influences on musculoskeletal, cardiovascular and neurocognative diseases and mortality in a cohort of elderly women

CAIFOS (Calcium intake Fracture Outcome Study) / CARES (CAIFOS Age Related Extension Study) Extension Study - CAREES

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1. Background

The Bone and Mineral Research Group of the School of Medicine and Pharmacology, The University of Western Australia and the Department of Endocrinology and Diabetes, Sir Charles Gairdner Hospital conducted the Calcium Intake Fracture Outcome Study (CAIFOS) from 1998 to 2003. This was a population based study of women aged over 70 years of age with the aim of determining the efficacy of calcium supplementation in preventing fracture. The treatment phase of this two-arm, randomised, double-blind, 5-year study of calcium supplementation (1200mg/d) versus placebo in 1,500 women has concluded in 2003. Of 24, 800 women aged over 70 on the electoral roll women 6.2% were recruited. The mean age of the population at baseline was 75.1 ± 2.7 years. These women had no illness likely to limit their involvement in the study for five years, and were not taking bone active agents. Although the subjects entering the study were weighted in favour of those in higher socio-economic categories they do not differ from the whole population in health resource utilisation [1].1275 (85%) of the study subjects remain active at the end of the CAIFOS study (2003).

The CARES study is a five-year (2003-2008) NHMRC supported extension of CAIFOS to collect further information regarding factors that influence osteoporosis and other diseases. 1136 (75%) patients from the original CAIFOS cohort enrolled in the CARES study. As of December 2007, 964 (85%) of the CARES cohort (64% of the CAIFOS cohort) was still active. Mortality accounts for 218 (14.5% of the original cohort), many of the remaining 21.5% are now in nursing facilities.

In this proposed project, we plan to continue to follow-up these subjects for another 5 years. This longitudinal study provides a unique opportunity for studying factors influence morbidity and mortality with aging, which is of great importance for the development of healthy aging policy with the aging of the Australian population. It is projected that by 2051 there will be a much greater proportion of people aged 65 years and over than in 2004. In 2004 people aged 65 years and over made up 13% of Australia's population. This proportion is projected to increase to between 26% and 28% in 2051 and to between 27% and 31% in 2101. While in 2004 there were just under 300,000 people aged 85 years and over in Australia (1.5% of the population), this group is projected to grow, to 2%–3% by 2021, to 6%–8% by 2051, and to 7%–10% by 2101 (Australian Bureau of Statistics: http://www.abs.gov.au).

1.1 Data set

The database currently consists of baseline and longitudinal measurements of phenotypic data related to bone health, cardiovascular health and neurocognitive clinical function, including: environmental factors (eg dietary intakes, physical activity); anatomical measures (eg bone mass and structure, anthropometry, body composition, carotid intimal medial thickness); physiological function (eg mobility, muscle strength); metabolic function (eg ApoE levels, hormonal status, homocysteine, beta-amyloid); and genetic polymorphisms (eg estrogen receptor, TGF beta, ApoE 4, Cyp19, MTHFR). These factors may predict increased risk of clinical events such as osteoporotic fracture, cardiovascular events, and neurocognitive function.

By the use of diaries and telephone calls we have maintained a complete set of adverse events and medication records throughout the past 9 years which are coded on line into an Access database using Real Beach event coding according to the second edition of the International Classification of Primary Care (ICPC2 Plus©) system [2]. This facilitates the determination of start and stop dates for adverse events and medications. Furthermore the participants have given approval for us to obtain a complete record of hospitalisation events and the cause of death from the Hospital Morbidity Data System (HMDS) should that occur. Thus we have a comprehensive record of the health and medications of these individuals.

1.2 Skeletal disease

In the first 5 years (1998-2003) follow-up of the CAIFOS population, a 5-year incident fracture rate of 16.2% has been observed. In the first 3.5 years follow-up (2004-2007) of the CAIFOS extension study, 9.9% subjects had incident fracture. According published fracture incident data [3], the 5-

year incident fracture rate is likely to be 20-30% for the next 5 years. Environmental and metabolic factors, namely hormonal status, genetics, medications use, co-morbidity and lifestyle factors affect the pathogenesis of osteoporosis. In the CAIFOS study, we have showed that in patients who took 80% of more of their assigned medication, 1200 mg/day calcium could reduce the 5-year fracture risk by 34% [4]. In cross-sectional analysis, we also found that lifestyle factors such as calcium intake, protein intake, physical activity and intakes of tea and chocolate were associated with bone structure in these women [5-9]. An association between hormonal factors such as estradiol [10-11] and genetic factors and bone density and short-term incidence of fracture has also been found in this cohort [12-16]. Bone structure data of CAIFOS/CARES participants have been collected at baseline, 1, 5, 7 years and will be collected at year 10. In the proposed project, we plan to collected bone structure data at year 12 and 15. Thus, the CAIFOS/CARES/CAREES study provides a unique opportunity to study the influence of environmental and genetic factors on the change in bone structure and strength with aging and fracture risk.

1.3 Cardiovascular disease

Hospitalization rates from coronary heart disease and stroke are 3.5 times and 8 times higher respectively in women aged 75 compared with women aged 55. The prevalence of cardiovascular disease or condition is 15.5% in the total population, the reason why this is a National Health Priority Area (NHPA) [17]. In the first 5 years follow-up of the CAIFOS population, 12.3% of subjects reported incident ischaemic heart disease, peripheral vascular disease or stroke. The intima medial thickness measurements undertaken on the CAIFOS population were compared to one of the most quoted study's of IMT predictive value, the ARIC study [18]. This study which compared <1mm vs ≥1mm mean baseline IMT, found that women with IMT≥1mm ("extreme mean IMT") had a HR 5.07 for coronary heart disease incidence over 4 year follow-up when compared to those with IMT <1mm ("non-extreme mean IMT"). 4.9% of the CAIFOS women had an IMT ≥1mm placing them at increased risk of coronary heart disease. The CAIFOS study provides a unique opportunity to assess the interactions between evidence of early atherosclerosis, novel cardiovascular risk factors and cardiovascular outcomes in postmenopausal women. The women in the CAIFOS/CARES study have had their cardiovascular risk factors as well as their genomic profile and hormonal status well characterised, are now likely to develop cardiovascular events at an increasing rate.

1.4 Neurocognative function

Another NHPA is mental disorders where the prevalence is 5.8% of the total population [17]. Four percent of CAIFOS/CARES subjects have reported dementia, loss of memory or have been diagnosed with AD. Cognitive function of subjects has also been assessed by the Abbreviated Mini Mental State Score during the study.

1.5 Links between metabolic systems

Some of the parameters we have collected relate to all three-focus research areas and can be advantageous when determining links between metabolic systems. For example, the lipid transport protein apolipoprotein ε4 (APOE-ε4) is an important risk factor for cardiovascular disease and has been associated with increased plasma cholesterol levels [19]. Other studies have shown that patients suffering from cardiovascular disease and those with high plasma cholesterol exhibit increased neuropathology characteristic of Alzheimer's Disease (AD) [20]. In addition in the CAIFOS population we have associated reduced bone density in subjects with the APOE-ε4 genotypes [21]. Findings from the Study of Osteoporotic Fracture has shown that women with rapid hip bone loss were more likely to develop cognitive decline than those with who had slower bone loss [22]. The mechanism of the association is still unclear but suggestive of an APOE-ε4 phenotype.

In the area of neurocognitve health, APOE-ε4 is one of the major genetic risk factors for AD [23-25]. Some studies have demonstrated that polymorphisms in the APOE promoter also promote risk of AD, which is independent of the APOE-ε4 allele. In particular the AA polymorphism at position -491 of the APOE promoter is associated with an increased risk of AD and its presence is consistent with increased expression of the apoE protein [26, 27]. It is interesting to note that the

levels of apoE may be regulated by estrogen. There is considerable interest in the role of estrogen on cognitive performance and the CAIFOS study has allowed for such studies to examine the association between endogenous estrogen levels with cognitive performance. Hormone levels may have important regulatory effects, directly or indirectly, that modify the complex genetic mechanism of regulation of apoE expression. It has been reported that estradiol up-regulates apoE gene expression by increasing levels of apoE mRNA in the polysomal translating pool [28] and region specific up-regulation of apoE by estrogen (although in non physiological concentration) has also been reported [29]. Additionally, the neuro-protective role for estrogen in global ischaemia is apoE-dependent [30]. These studies also suggest that each of these mechanisms are largely mediated by estrogen receptors (ERα and ERβ). As such the effect of polymorphisms within ERα would be of interest to investigate in correlation with cognitive performance – especially in light of recent publications linking such polymorphisms with an increased risk of developing AD in females [31].

In the area of skeletal and cardiovascular medicine, estrogen has long been recognized as a protective factor in women prior to menopause. Its action after menopause is pronounced in maintaining bone density [32]. In the CAIFOS/CARES cohort, we have recently shown that the women in the lowest tertile of free estradiol index had twice the 5-year risk of sustaining a clinical vertebral fracture compared to those in the highest tertile [10]. Estrogen's protective action against cognitive decline involves regulation of cholesterol metabolism analogous to its action against cardiovascular disease. Thus it may be predicted that identification of factors that modulate estrogen's action against cognitive decline may be applicable to its action against cardiovascular disease.

Another metabolic factor that is considered a risk factor cerebrovascular disease and may play an important role in the pathogenesis of AD is total plasma homocysteine (tHcy) levels [33]. High values of plasma tHcy and low levels of vitamin B12 and folate are frequently present in AD patients [34, 35] and CVD patients [36]. Moreover, the homozygous mutation (C677T) of the methylene tetrahydrofolate reductase (MTHFR) gene, related to a thermolabile type of the encoded enzyme, causes hyperhomocysteinemia by reducing the 5-methyltetrahydrofolate availability. As such there is interest in determining the validity of the association of this mutation with both AD and pre-dementia cognitive impairment [37]. Homocysteine and folate levels, which have been measured, can be examined in relation to ongoing assessments of cardiovascular and cognitive endpoints, which include time to event.

1.6 Mortality data

Another important endpoint collected during the CAIFOS/CARES follow-up is death data. All cause of death data have been obtained from the HMDS mortality data and have been ascertained from death certificates from the Registrar of Births Marriages and Deaths. So far 218 women of the original CAIFOS had deceased. In a recent report, we have found that daily back pain, present in 25% of the patients, is associated with greater overall 5-year mortality risk (hazard ratio: 2.03 95% CI 1.14-3.60) compared to those who experienced back pain less than once a month [38]. The death rate is expected to increase over the next 5 years, and it would be important to understand factors associated with mortality risk to development strategies to increase the life expectancy of the elderly.

Therefore, we would like to continue to follow-up these subjects for another 5 years by obtaining subjects' self-reported adverse events data and their hospital admission and mortality data through the Hospital morbidity data scheme (HMDS) and to collect data on anthropometry, bone structure, physical function, dietary intake and metabolic function at year 2 and 5. This follow-up study will continue to provide valuable insight into the prevalence, incidence, prognosis, and predisposing risk factors for mortality and diseases in three focus areas: osteoporotic fracture, cardiovascular and cerebrovascular disease and neurocognitive clinical function in a cohort of elderly women.

2. Study Aim

The aim of this five-year prospective epidemiological study is to determine health outcomes in a cohort of elderly postmenopausal women who in 2008 will have completed the five-year extension

of a five-year population based clinical trial of a public health intervention commenced in 1998. The dietary intervention study was a five-year study of an increased oral administration of calcium in the prevention of fracture.

The specific aims are:
1. Through collecting self-reported adverse events and HMDS hospital admission and mortality data, evaluate the effects of parameters already collected including:

- Environmental factors (eg dietary intakes, physical activity)
- Anatomical measures (eg bone mass and structure, anthropometry, body composition, carotid intimal medial thickness)
- Physiological function (eg mobility, muscle strength)
- Metabolic function (eg ApoE levels, hormonal status, homocysteine, beta-amyloid)
- Genetic polymorphisms (eg estrogen receptor, TGF beta, ApoE 4, Cyp19, MTHFR)

on predicting risk of clinical events specifically osteoporotic fracture, cardiovascular events, neurocognitive function and death.

2. Study the changes in bone structure, body composition, quality of life, physical function and neurocognitive endpoints with aging and to investigate factors influencing these changes.

3. Study the relationship between aging and resource consumption through collecting data on hospital admission and length of stay.

3. Study Design

This is a prospective five-year follow-up trial of a cohort of elderly women who were participants in a five-year study of the effect of calcium supplementation on fracture prevention and its five years extension.

3.1 Subjects

As this is an extension to an existing study currently ongoing, new study participants will therefore not be recruited. All consenting participants of the CAIFOS/CARES trial will be eligible. Subjects will be aged over 80 years at the beginning of this extension study.

3.2 Withdrawals

Withdrawals will be handled as missing values in the statistical analysis. Data for those subjects who withdraw will be used up to the time of withdrawal. Hospital morbidity data (HMDS) will be obtained from 2009-2013 on all CAIFOS/CARES subjects (withdrawn and active) unless the subject withdraws their consent for retrieval of HMDS data.

3.3 Drugs and Interventions

Nil.

3.4 Study Plan and Schedule of Assessment

3.4.1 End of CARES study

During 2008 all active subjects will be reviewed within a four-week period of their baseline anniversary date. Each subject will undergo a repeat of all baseline measurements. The 10-year change in bone density will be calculated and reported to each subject and reported to her GP. The data analysis will take place at the end of 2008 and letters relating the findings of the study will be communicated to all subjects.

During the 2008 visit, each subject will be asked whether they wish to continue with the extension study. Those subjects that wish to continue will be required to sign a Patient Information and Consent Form entitled "**CAIFOS/CARES Addendum 7: Lifestyle, anatomical, metabolic and genetic influences on musculoskeletal, cardiovascular and neurocognative diseases and**

mortality in a cohort of elderly women".

3.4.2 Contact 1-3 and 5-9

The subjects will be reviewed every 6 months after their clinic visit in 2008 using a short telephone interview. The data collection will include:

Falls and incident fracture

Each subject will be asked whether they have had any fall and/or fracture in the previous 6 months. All incident fractures will be verified by a radiological report.

Adverse event data

Adverse events have been monitored during CAIFOS (years 1-5) using a diary system that is completed by the subject, their GP, other health professional or specialist and CARES (years 6-10) through telephone interview. In this study, adverse event collection will continue to be administered by the clinical research assistant at the time of the telephone interview and recorded in the subject diary kept in the subjects source data. The adverse event review will entail review of any significant adverse events and changes in medication use. This will include hospitalisation for any reason, change in state or severity of chronic or present disease, and an acute condition that required medication, new diagnosis of a condition not previously known in addition to any fall, fracture or/and death. In addition, patient medical records will be obtained via the Data Linkage Unit of the Health Department of Western Australia utilizing the Hospital Morbidity Data System (HMDS). Adverse data will then be coded according to the second edition of the International Classification of Primary Care (ICPC2 Plus©) system [2], a primary care data base in which commonly used lay terms are classified under predetermined headings by experienced staff, and then entered into assess database. The codes used in the ICPC2 Plus© system corresponded directly to specific ICD 10 codes (the Tenth Revision of the International Classification of Diseases). Hospital admission and length of stay in hospital will also be collected through HMDS.

All cause of death data will continue to be obtained from the HMDS mortality data. The causes of death will be classified according to ICD10 criteria.

3.4.3 Contacts 4 and 10

For contact 4 and 10, at the 2nd (2010) and 5th (2013) year of the extension study (CAIFOS Year 12 and 15) subjects will attend a clinic visit at the Sir Charles Gairdner Hospital. For subjects who cannot come to the hospital, the clinical review will be conducted at subjects' home by research assistants. Fasting blood and urine samples will be collected from all participants. The data collection will include:

a) Falls and Incident fracture

Each subject will be asked whether they have had any fall and/or fracture in the previous 6 months. All incident fractures will be verified by radiological reports.

b) Adverse event data

Review of any significant adverse events and changes in medication use. This will include hospitalisation for any reason, change in state or severity of chronic or present disease, and an acute condition that required medication, new diagnosis of a condition not previously known.

c) Anthropometry

Standing height will be measured by a wall-mounted stadiometer to the nearest 0.1cm and body weight will be measured by an electronic scale to the nearest 0.1 kg. Mid upper arm triceps skin fold will be measured by a caliper to the nearest 0.01cm. Waist and hip girth will be measured by a tape measurer to the nearest 0.1 cm. Body mass index will be calculated. These data will allow us to assess the change in anthropometric measures with aging.

d) Bone structure

Measurements of bone parameters will be ascertained at the visit. These will include:

Quantitative ultrasound measurements (QUS): QUS of the calcaneus of the left foot will be measured in duplicate using a Lunar Achilles Ultrasound machine (Lunar Corp., Madison, WI, USA). The manufacturer's quality assurance methods will be employed. The average measurement of the Speed of Sound (SOS), Broadband Ultrasound Attenuation (BUA) and Stiffness will be determined. The CV for SOS and BUA determined on the patients in our laboratory are 0.43% and 1.59%, respectively.

***Dual energy X-ray absoprtiomtery (DXA)*:** Bone mineral density will be measured on a Hologic 4500A bone densitometer (Hologic Corp., Waltham, MA, USA) at **the hip and whole body** with CVs under 2% in our laboratory. Lean and fat body mass will be derived from the whole body scan. **Morphometric X-ray absorptiometry (VFA) will also be made**.

Peripheral quantitative computed tomography (pQCT): Scans will be undertaken using a Stratec XCT 2000 (Stratec Medizintechnik GmbH, Pforzheim Germany). The voxel size will be set at 150μ in the x and y direction and 1000μ in the z direction, which will increase the scan time to five minutes. **At radius, the distal radius (4% site) and the 15% site will be measured. At tibia, the distal tibia (4% site) and the 15% site will be measured**. The CV error in our laboratory for total BMD is 4.6%, for trabecular BMD is 4.0% and for cortical BMD is 8.0%. A previously validated biomechanical parameter, the Stress Strain Index (SSI), will be calculated as the product of the section modulus and cortical density normalized to the maximal physiological cortical density of human bones (1200 mg/cm^3) for the polar moment (CV is 5.8%) and the bending moments in the x (CV is 3.3%) and y (CV is 2.6%) directions, where the y direction is the widest part of the radius and the x direction is perpendicular to this.

Central QCT spine and hip: QCT scans of spine and hip will be undertaken using a Phillips 64 slice scanner.

QCT Hip

Scans parameters: Peak voltage 120 kV, 100 mAs, 1mm slice thickness, pitch 1.

The patient will be positioned on top of the Mindways calibration phantom and will be positioned according to the Mindways manual. The scan will commence 1 cm below the lesser trochanter and continue to 5mm above the left femoral head.

Reconstruction parameters:

First reconstruction, reconstruction field of view: 40cm, reconstruction increment: 1 mm, using kernel B.

Second reconstruction the **left hip only** reconstruction field of view: 15 cm, reconstruction increment 1 mm, kernel B, the calibration phantom will not be visible in this FOV.

The images will be saved in DICOM format and will be sent to Endocrinology, SCGH.

QCT Spine

Scans parameters: Peak voltage 120 kV, 50 mAs, 1mm slice thickness, pitch 1

The patient will be positioned on top of the Mindways calibration phantom according to the Mindways manual. The scan will commence at the xiphoid level and scan down a length of approx 10 cm to include T12, L1 and L2. Patients can breathe normally during the scan.

The images will be saved in DICOM format and sent to Endocrinology, SCGH.

Dose reduction

The maximum mAs will be set at 100 for hip scan and 50 for spine scan. Dose reduction functions of ACS (automatic current selection) and DOM (dose modulation) will be used to reduce the radiation dose according to the size of patient.

e) Questionnaires

- ***Dietary intake**:* Dietary intake will be reassessed using the **Cancer Council Victoria Food Frequency Questionnaire** administered at the CAIFOS/CARES study. The process of collection will be identical where by a dietitian supervises the completion of the questionnaire in small groups. Food models, cup and spoons and charts for frequency of seasonal fruits are used. The machine readable answer sheets are checked and sent to the Cancer Council

Victoria for processing. These data are received in a spreadsheet format and include individual food frequency data and mean nutrient intake for 31 nutrients. Beverage intakes will be assessed by a **Beverage Frequency Questionnaire**.

- ***Quality of life and depression:*** Quality of life will be assessed by a symptom related **quality of life (SF36) questionnaire**. Depression will be assessed by the **Patient Health Questionnaire (PHQ-9)**. These data will allow us to assess how quality of life changes with aging and co-morbidity.
- ***WOMAC:*** The **WOMAC questionnaire** (Western Ontario & Mcmaster University Osteoarthritis Index) will be used to assess the pain and stiffness subjects experienced at knees and hips and the physical function of their knees and hips.
- ***Cognitive function:*** Cognitive function will be assessed using the **abbreviated mini mental state test (AMS)**.
- ***Activity of daily living:*** Activities of daily living will be assessed using the **Barthel questionnaire**.

f) Physical function

- ***Muscle strength:*** hand grip strength will be assessed by a hand dynamometer. Subjects will be given a practice attempt then given three attempts, where the greater of these will be recorded.
- ***Mobility functioning:*** mobility functioning will be measured by the ***Timed Up and Go Test (TUAG)***. The TUAG test requires the participants to be timed while getting up, walking 3 meters, turning, returning to chair and sitting down again. The test will be practised once and then timed.
- ***Balance function:*** Balance function of subjects will be assessed by the ***Romberg test.***
- ***Balance reaction and applied muscle force***: will be assessed using a Balance–X–sensor (Soehnle Professional, Germany).

g) Biochemistry

Fasting blood and urine samples collection will be taken at contacts 4 and 10. The venous blood sample will be collected in the morning after an overnight fast from 10 pm. The serum and plasma will be rapidly separated from the blood after collection.

Bone metabolism related biochemistry: Serum calcium, phosphate and alkaline phosphatase and urine calcium, phosphate, creatinine, biochemical markers of bone resorption (NTX) and formation (P1NP), 25OHD, PTH

Hormone: estrodiol

Cardiovascular disease related biochemistry: Fasting blood lipids, glucose, C-reactive protein, homocysteine, apolipoprotein E.

***Cognitive function related biochemistry*:** Plasma beta-amyloid.

Renal function related biochemistry: Plasma creatinine and calculated GFR.

3.4.4 Summary of Assessment Schedule

	Data collected or will be collected during CAIFOS/CARES	CAIFOS year				
		11 (2009)	12 (2010)	13 (2011)	14 (2012)	15 (2013)
		Phone	Clinic	Phone	Phone	Clinic
Adverse events	Baseline – year 10	**X**	**X**	**X**	**X**	**X**
Anthropometry	Baseline and year 5, 7 and 10		**X**			**X**
Physical function						
Muscle strength, TUAG, Romberg	Baseline and year 5, 7 and 10		**X**			**X**
Balance reaction and applied muscle force	Year 10		**X**			**X**
Bone structure						
Heel QUS	Baseline and year 5, 7 and 10		**X**			**X**
DXA hip and total body	Year 1, 5, 7 and 10		**X**			**X**
VFA	Year 1, 5, 7 and 10		**X**			**X**
pQCT distal radius and distal tibia	Year 5, 7 and 10		**X**			**X**
QCT hip and spine	Year 9 (in 100 subjects)		**X**			**X**
Questionnaires						
Dietary intake	Baseline and year 5, 7 and 10		**X**			**X**
SF-36 quality of life	Baseline and year 5, 7 and 10		**X**			**X**
PHQ-9			**X**			**X**
WOMAC	Year 4, 7 and 10		**X**			**X**
Mini Mental State	Baseline and year 5, 7 and 10		**X**			**X**
Barthel activity of daily living	Baseline and year 3, 5, 7 and 10		**X**			**X**
Demographic questionnaire	Baseline and year 5, 7 and 10		**X**			**X**

3.5 Statistical Analysis

All primary analyses of osteoporotic fracture, cardiovascular events (ie stroke, TIA, myocardial infarction, unstable angina and sudden cardiac death using ICD definitions), neurocognitive impairment and death will use time-to-event methods. For a given outcome, the time of event is determined from study commencement (during 1998) to the first diagnosis as determined by reporting in the adverse event diary retained by the centre. Participants without a diagnosis will be censored for that event at the time of last follow up. Participants who withdraw from the study will be censored at the time of withdrawal. Primary outcome comparisons will be presented as hazard ratios (HRs) and 95% confidence intervals from Cox proportional hazards analyses after adjustment for age, years since menopause, body weight, metabolic and environmental factors.

These data will also be analysed using logistic regression at time of last follow up. Being a prospective follow up study we can calculate the predictive risk of osteoporotic fracture,

cardiovascular events, and neurocognitive impairment based on their baseline characteristics, randomisation status (placebo or treatment) and prior disease and medication use.

To determine the lifestyle, anatomical, metabolic and genetic predictors of changes in bone structure, body composition, quality of life, physical function and neurocognitive function with aging, stepwise multiple regression will be used to determine which factors play a significant role.

The significance level for test statistics will be set at $P < 0.05$. All data were will be analyzed by SPSS (version 15; SPSS Inc, Chicago, IL)

3.6 Data Management and Reporting

All data from telephone interviews, HMDS and clinic visits will be entered in to the same relational database as utilised for CAIFOS/CARES study ie. Real Beach which runs in parallel to Site Safe system, that front ends Microsoft Access. This utilises ICPC2 Plus© a database of disease coding and CAPS©, a database of pharmaceutical's, developed and supplied by the Family Medicine Research Unit, Department of General Practice, University of Sydney [2].

4. Compliance with Good Clinical Practice & Ethical Considerations

This study will be conducted in compliance with the conditions stipulated by the Research Institute Ethics Committee, informed consent regulations and NH&MRC Guidelines. All amendments to the study will be placed before the Research Institute Ethics Committee for approval. Any information that may affect either committee's decision to continue approval for the trial will be forwarded to the committees without delay. An annual report will be issued, in accordance with the guidelines provided by the Research Institute Ethics Committee.

5. Informed Consent

The investigator, or a person designated by the investigator will explain the benefits and risks of participation in the study to each subject and obtain written informed consent prior to the subject entering the study by signing a consent form approved by HREC. Each subject's original consent form, signed and dated by the subject will be retained by the investigator. All subjects will receive a copy of the document.

6. Record retention

Records will be retained for 15 years in a locked facility, in accordance with NH&MRC guidelines. Access to the records will be limited to treating clinician, chief investigator and his/her nominated co-investigators, and authorised representatives of drug regulatory bodies. Patient confidentiality will be maintained in accordance with local requirements.

7. Withdrawal criteria

Participants will be withdrawn from the study under the following condition:
Patient request.

8. Liability & Indemnity

The risks of participating in this study are low. In the unlikely event of liability for an adverse event related to a per protocol event the University of Western Australia will carry liability.

9. Documentation of study findings

The results of this study will be published in peer reviewed journals and presented at scientific meetings. This will be after all investigators have agreed to the manuscript. Authorship will be determined by mutual agreement in relation to normal standards relating to having played a substantial role in the study.

10. Study dates

Starting date: August 2008

Finish date: December 2013

11. References

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