Study Protocol

Leucine and ACE inhibitors as therapies for sarcopenia: a two by two factorial randomised placebo controlled trial

Short lay title:

Perindopril and Leucine to improve muscle function in older people

Study Acronym	LACE trial
Sponsor	University of Dundee-NHS Tayside
Sponsor R&D Number	2013GR06
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PROTOCOL APPROVAL

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EudraCT number 2014-003455-61

Signatures

By signing this document I am confirming that I have read, understood and approve the protocol for the above study.

Dr Miles D Witham
Chief Investigator

Signature

27/5/19
Date

LIST OF ABBREVIATIONS
(including Study abbreviations)

AE	Adverse Event
AR	Adverse Reaction
BIA	BioImpedance Assay
CNORIS	Clinical Negligence and Other Risks Scheme
CRF	Case Report Form
DXA	Dual Energy X-ray Absorptiometry
EQ5D	EuroQol 5D
EWGSOP	European Working Group on Sarcopenia in Older People
FFQ	Food Frequency Questionnaire
GCP	Good Clinical Practice
IMP	Investigational Medicinal Product
ISF	Investigator Site File
MHRA	Medicines and Healthcare Products Regulatory Agency
NEADL	Nottingham Extended Activities of Daily Living
NRES	National Research Ethics Service
REC	Research Ethics Committee
SAE	Serious Adverse Event
SmPC	Summary of Product Characteristics
SOP	Standard Operating Procedure
SPPB	Short Physical Performance Battery
SUSAR	Suspected Unexpected Serious Adverse Reaction
TMF	Trial Master File
UAR	Unexpected Adverse Reaction
6MWD	Six Minute Walking Distance

SUMMARY

Hypothesis and aims: We hypothesise that supplementation with the essential amino acid leucine and/or the angiotensin converting enzyme inhibitor perindopril will improve muscle mass and function in older people with sarcopenia. Leucine supplementation has been shown to counter the resistance to muscle protein synthesis in older people, reduce inflammatory markers, improve insulin resistance, increase satellite cells (indicating muscle regeneration) and improve mitochondrial function in muscle. Perindopril (a common cardiovascular medication) has been shown to reduce inflammatory cytokines and myokines, improve insulin resistance and mitochondrial function in skeletal muscle.

We have previously found that perindopril improves exercise capacity (equivalent to that seen after 6 months of exercise training) in older people. While both interventions can positively affect many factors implicated in the development of sarcopenia their effect has not been tried in this group. We will study the effect of leucine and perindopril on muscle performance and muscle mass in sarcopenia. We will also investigate biomarkers which predict response to interventions and examine mechanisms of faction of these interventions on sarcopenia.

Design: Multicentre double blind randomised controlled 2x2 factorial trial

Population: >/=70 years with sarcopenia as per the EWGSOP definition (loss of both muscle mass and function). Excluding those on or with contraindications to perindopril.

Interventions: 4 groups receiving one of perindopril+leucine, perindopril+placebo, leucine+placebo or placebo+placebo for 12 months. Leucine dose will be 2.5g, 3 times/day and perindopril will be 4mg/day.

Outcomes: Primary outcome is the difference in Short Physical Performance Battery between leucine vs. placebo and perindopril vs placebo. Secondary outcomes are change in muscle mass, grip strength, quadriceps strength, 6 minute walking distance, quality of life (EuroQol)

Assessments: Performed at baseline, 6 and 12 months. Safety assessments will be done 3 monthly.

Sample size: 440 participants (n=110 per group) will have 90% power at 0.05 alpha to identify a change of 0.5 (SD 2.7) in primary outcome allowing for a 20% drop out rate.

Statistical analysis: Change in outcome measures between interventions will be analysed using repeated measures using mixed modelling.

Potential benefit: Sarcopenia is a major contributor to loss of mobility, falls, institutionalisation, morbidity and mortality in older people. Both leucine and perindopril are inexpensive. If found to be acceptable for older people and effective in improving sarcopaenia, these agents will be value for money, costing less than £300 per patient per year.

Biomarker substudy: Blood will be analysed for biomarkers that predict improvement in muscle mass and function. Changes in microRNA, myokines, cytokines and hormones in blood, plus selected gene polymorphisms will be measured to examine predictors of response to therapies and changes in biological pathways in sarcopenia in response to interventions.

Derby substudy:

A further substudy will be performed on participants at the Derby site; muscle mass will be estimated at baseline, 6 and 12 months using D3 creatine (a stable isotope tracer), and muscle turnover will be estimated using 3-methylhistidine as a stable tracer.

1 INTRODUCTION

1.1 BACKGROUND

Sarcopenia (the age related loss of muscle mass and function) is a major contributor to loss of mobility, falls, loss of independence, morbidity and mortality in older people. The mechanisms behind the development of sarcopenia are not fully understood, but accumulating evidence suggests that it is multifactorial in aetiology. Some of the factors involved are an altered turnover of muscle protein leading to a reduction in muscle protein synthesis (MPS) and a relative increase in muscle protein breakdown (MPB); chronic inflammation with increased cytokines including TNF and IL-6, inactivity, mitochondrial dysfunction, and altered neuromuscular junction structure and function. Currently, the intervention with the most evidence for efficacy in preventing and reversing sarcopenia is resistance exercise training. However, many older people are sedentary, and either cannot, or do not want to exercise.

Non-exercise interventions to prevent or counter the effects of sarcopenia are thus urgently required. A range of potential interventions have been proposed, including protein supplementation, myostatin inhibitors, testosterone and selective androgen receptor modifiers, growth hormone and novel interventions including activin ligands. Many of these approaches have either suffered from frequent side effects (e.g. testosterone, growth hormone) or are under evaluation in early-phase studies by the pharmaceutical industry. We propose to study the efficacy of two promising interventions (leucine and angiotensin converting enzyme inhibition) that can potentially improve muscle mass and function in people with sarcopenia as defined by the European Working Group on Sarcopenia (EWGSOP). Promising preliminary data exists from small trials for both interventions, but neither have yet been tested in adequately powered trials enrolling people with established sarcopenia.

1.2 RATIONALE FOR STUDY

We aim to investigate two interventions with potential to improve both muscle mass and function independently of exercise. Both have potential to improve factors contributing to sarcopenia, using different mechanisms.

Leucine
Muscle protein synthesis (MPS) in older people in response to protein ingestion is attenuated compared to younger people – i.e. there is anabolic resistance to protein supplementation. Increasing the amount of protein ingested is one way of overcoming this issue, but older, frail people typically already have suboptimal protein intakes and increasing protein intake may be challenging in practice.

Leucine, a branched-chain amino acid, is known to have important regulatory actions, mediated at least in part via the mTOR pathway. Leucine affects both aspects of protein turnover, by reducing proteolysis and enhancing protein synthesis in vitro. Studies in healthy older people confirm that addition of leucine to a protein meal enhances muscle protein synthesis; previous studies suggest that approximately 2.5g of leucine per meal is sufficient to generate the effect. In addition, leucine stimulates insulin release by pancreatic beta cells; insulin signalling not only improves glucose uptake by skeletal muscle, but is also an important anabolic signal for skeletal muscle.

Leucine-enriched amino acid supplements have been show to improve muscle strength and function in healthy older people, although one study of leucine alone failed to replicate this effect. However, no studies to date have examined the effect of leucine supplementation in people with diagnosed sarcopenia. This is precisely the group, with lower IGF-1 signalling, lower protein intake, and poor anabolic response to protein loading, who are most likely to demonstrate a response to intervention. Small trials of protein loading in frail older people show improvements in physical performance measured using the Short Physical Performance Battery test (1.0 point improvement at 24 weeks) but no improvement in skeletal muscle mass over this short follow-up period. In contrast, in older participants undergoing resistance training, protein supplementation did not enhance muscle strength or performance compared to placebo(15) suggesting that it is the non-exercising majority of older, frail people who are most likely to benefit from nutritional efforts to enhance muscle protein synthesis.

Angiotensin Converting Enzyme inhibitor drugs (ACEi)
Renin-angiotensin-aldosterone system (RAAS) activity has been implicated in skeletal muscle dysfunction via multiple biological pathways. Angiotensin II impairs endothelial function and hence muscle blood supply; is associated with increased levels of inflammation, suppression of IGF-1 and has important effects on mitochondrial function. Aldosterone too has deleterious effects, including lowering serum potassium, impairment of endothelial function, and promotion of fibrosis.

Conversely, ACEi drugs have been shown to have multiple potentially beneficial effects on skeletal muscle function. ACEi improve endothelial function, angiogenesis and reduce inflammation, which translates to improved blood flow and exercise capacity in patients with peripheral vascular disease. They improve mitochondrial function, enhance IGF-1 levels, and suppress pro-inflammatory cytokine levels including IL-6; thought to be a key inflammatory mediator of sarcopenia. ACEi have also been shown to promote skeletal muscle glucose uptake, and promote a shift from type 1 to type 2 muscle fibres (the type preferentially affected by sarcopenia). Finally, there is evidence that ACEi may enhance neuromuscular junction activity, and selected ACEi may also have beneficial central CNS actions improving mood and motivation, which are likely to have important consequent benefits on physical activity levels.

There is some evidence that these multiple biological functions may indeed translate into clinical benefit. We have previously shown that the ACEi perindopril produces a significant improvement in physical function (31m improvement in 6 minute walk; improvement in quality of life of 0.09 points on the EQ5D tool) in functionally impaired older people with a mean age 79 years. This is comparable to that achieved by 6 months of exercise training. Observational studies report better muscle strength and larger lower extremity mass in older people taking ACE inhibitors; excitingly, centrally acting ACEi are associated with a slower decline in activities of daily living in people with dementia.

However studies of ACEi in fitter older people have not shown positive results which suggests that the effects of ACEi may be more relevant in frailer people. Similarly to studies with leucine, addition of ACEi to older people undergoing exercise training also appears not to have effects beyond those evident from exercise training; no additional improvement in six minute walk was seen with perindopril in our previous trial in which all participants were undertaking an exercise programme. Thus the existing evidence suggests that frail older people not engaged in exercise training are the target population most likely to benefit from ACEi therapy. ACEi have not yet been specifically studied in people with sarcopenia who have both poor muscle mass and function, and providing evidence of efficacy in this group with a well-defined specific deficit in muscle physiology is key to testing whether ACEi really have clinically relevant benefit in the target population for this commissioned call – i.e. people with sarcopenia.

2 STUDY OBJECTIVES

2.1 OBJECTIVES

2.1.1 Primary Objective

To determine the efficacy of leucine and perindopril in improving physical function in older people with sarcopenia diagnosed using the EWGSOP definition.

2.1.2 Secondary Objectives

To evaluate the effect of leucine and perindopril on muscle mass in older people

To evaluate biomarkers that can predict response to leucine and perindopril in patients with sarcopenia

2.2 OUTCOMES

2.2.1 Primary Outcomes

The primary outcome will be the between group difference in Short Physical Performance Battery (SPPB) score at 12 months.

2.2.2 Secondary Outcomes

Between group differences in:

- Appendicular muscle mass/height squared (measured by dual energy X-ray absorptiometry),
- Grip strength
- Quadriceps strength (handheld dynamometry)
- 6 minute walk test
- Gait speed (4m walk)
- Chair stands (Sit to stand test x 5)
- Activities of daily living (Nottingham extended ADL questionnaire) and quality of life (EuroQol 5D questionnaire).
- HOMA IR (Homeostatic Index of insulin resistance; measured from glucose and insulin levels)
- Hip Bone mineral density

Substudy outcomes (Derby participants only):

- Muscle mass measured by D3 creatine dilution
- Muscle protein turnover measured by 3-methylhistidine excretion

In addition, data will be collected on adverse events, medication adherence by tablet counting and leucine tub container weight, falls, hospitalisation, death and admission to institutional care; consent will be obtained for open-label follow up of participants for these outcomes collected from routine clinical data following the end of the 1 year study period. Data will be collected for diet at baseline and 12 months using the Scottish Collaborative Group Food Frequency Questionnaire (FFQ); this will allow the interaction between treatment and protein intake (total, plant vs animal protein) to be ascertained.

3 STUDY DESIGN

3.1 STUDY DESCRIPTION

LACE is a multicentre, double blind, placebo-controlled, 2x2 factorial randomised controlled trial. Participants will be randomised to receive either perindopril 4mg once daily or matching placebo, and to receive leucine 2.5g three times per day or matching placebo to mask treatment allocation to participants, investigators and usual healthcare providers. Intervention and follow up will be for 1 year. All trial outcomes will be measured and analysed masked to treatment allocation group. Randomisation will be performed via a central, web-based allocation system, using a minimisation algorithm to ensure balanced groups for key baseline factors (age, sex, SPPB, grip strength, comorbid disease, recruitment centre).

3.2 TRIAL FLOWCHART

Please see Appendix 1

3.3 STUDY MATRIX

Please see Appendix 2

3.4 SUB STUDY MATRIX

Please see Appendix 3

4 STUDY POPULATION

4.1 NUMBER OF PARTICIPANTS

The LACE trial aims to recruit a total of 440 participants, from a minimum of 15 centres across the UK. Recruitment will take place over an initial 18 month period, with extension if required due to low recruitment rates. We anticipate a dropout rate of 20% at 1 year, and thus aim to recruit a final evaluable sample of 352 patients. Dropout rates will be reviewed after 9 months and 12 months of recruitment, and the total recruitment target will be adjusted based on projected dropout rates at these time points to ensure a final evaluable sample size of 352. The substudy performed at Derby aims to recruit a minimum of 30 participants

4.2 INCLUSION CRITERIA

Age 70 and over

Sarcopenia criteria according to EWGSOP definition, based on:

- low height-adjusted total skeletal muscle mass on BioImpedance Assay (BIA) using the BIA 101 device and the Sergi equation. Cutoffs will vary with body mass index and sex as follows:

	Males	Females
BMI <18.5 kg/m^2	<=6.02 kg/m^2	<=5.25 kg/m^2
18.5 – 24.9	<=7.14	<=5.70
25.0 – 29.9	<=8.00	<=6.19
>=30 kg/m^2	<=8.77	<=6.72

- and either low gait speed (<0.8 m/s on 4m walk) or low handgrip strength (<20Kg for women, <30Kg for men)

4.3 EXCLUSION CRITERIA

a) Contraindications or existing indications to therapies or placebo

-Known clinical diagnosis of chronic heart failure (by European Society of Cardiology criteria)
-Confirmed LV systolic dysfunction on any imaging modality
-Known aortic stenosis (peak gradient >30mmHg)
-Systolic BP<90 mmHg (supine)
-Dizziness on standing associated with a postural drop of >20/10mmHg (asymptomatic orthostatic hypotension will not be a contraindication)
-Serum Creatinine >170 umol/L or eGFR<30ml/min by MDRD4 calculation
-K>5.0 mmol/L; Na<130 mmol/L
-Using ACEi, Angiotensin receptor blocker, aldosterone blocker or leucine already
-Previous adverse reaction to ACEi or leucine
-Current use of oral NSAIDs (aspirin is permitted, as are topical NSAIDs)
-Current use of potassium supplements, aliskiren, spironolactone or other potassium-sparing diuretics
-Hereditary or idiopathic angioedema
-Lactose intolerance

b) Contraindications to consent or undertaking study outcomes

-Implantable cardioverter defibrillator or pacemaker with atrial sensing lead (pacemakers with ventricular sensing lead only are allowed)
-Peripheral oedema present above knee level
-Unable to mobilise without human assistance (walking aids allowed)
-Unable to give written informed consent
-Currently enrolled in another intervention research study, or less than 30 days since completing another intervention research study. Concomitant enrolment in observational studies is permitted.

c) Overlap with other myopathic conditions or important confounders

-Currently enrolled in a time-limited exercise-based rehabilitation programme
-Any progressive neurological or malignant condition with life expectancy <6 months
-Severe COPD (GOLD stage IV)
-Known myositis or other established myopathy
-Self-reported weight loss of >10% in last 6 months (to exclude significant cachexia)
-Known uncontrolled thyrotoxicosis
-7.5mg/day or greater prednisolone use (or equivalent)

5 PARTICIPANT SELECTION AND ENROLMENT

5.1 IDENTIFYING PARTICIPANTS

A four-pronged approach to recruitment will be used, informed by current evidence and best practice guidelines:

1) Via UK Primary Care Research Network
2) Via recruitment from secondary care clinics
3) Via targeted advertising to the older population
4) Via patient research databases e.g. SHARE (Scottish Health Research Register)

A telephone pre-screening phase will be included. This will allow rapid, efficient selection of participants most likely to pass a screening visit, and represents the best available strategy for practical screening in sarcopenia trials. This will be based on a brief telephone conversation conducted by the research nurse exploring any contraindications (e.g. heart failure, taking ACEi) and on administration of the SARC-F tool (see Appendix 4); a threshold score will be used to denote a high probability of having sarcopenia, and would prompt a screening visit. The threshold score will initially be set at 4 points or above (out of 10). This threshold will be reviewed regularly in the light of screening information, and will be adjusted up or down to maximise efficiency of recruitment. Patients who are not suitable to attend for a screening visit will be asked if they agree to be contacted if in the future the SARC-F threshold changes and they may be then suitable to attend for a screening visit. Full details of the method of adjustment will be given in an accompanying working practice document. Tokens of appreciation (e.g. mugs, fridge magnets) will be given to participants as reminders about participation in the trial.

See Appendix 1 for the trial screening flowchart

5.2 CONSENTING PARTICIPANTS

Obtaining consent will be carried out by the research nurse (RN) at each centre, local PI or other local delegated individual. Potential participants will be provided with a study information brochure either at the clinic or by post with a stamped addressed FREEPOST envelope to reply. After receiving a positive response from a potential participant, either orally or by return of a reply slip, a pre-screening telephone call will be used to check provisional eligibility, then a screening visit will be arranged where participants will be able to ask any further questions about the study. A full participant information sheet (PIS) will be posted to the patient and all participants will be given at least 48 hours to consider their participation. If they decide to

participate, informed consent will be taken by the RN or other local delegated individual and eligibility by all the criteria confirmed.

Where a participant requests to speak with a physician from the study team the consent process will not be completed until the participant had spoken to the physician and had all their questions answered to their satisfaction.

5.3 SCREENING FOR ELIGIBILITY

Participants will attend a screening visit. Written informed consent will be obtained at the screening visit, and the following assessments performed to check eligibility:

- Medical and medication history
- Blood sample for U+Es (if none available in the last 4 weeks)
- Handgrip measurement
- Short Physical Performance Battery
- Blood pressure
- Bioimpedance measurement to estimate muscle mass

Participants will proceed to randomisation if they are eligible from the screening visit. U+E results do not require repeating at the screening visit if results from within 4 weeks prior to the screening visit are available and meet the eligibility criteria.

If the participant is deemed ineligible following one of the above assessments, data will still be collected on all assessments listed above apart from bloods.

5.4 INELIGIBLE AND NON-RECRUITED PARTICIPANTS

The reason(s) for ineligibility will be explained to participants and any questions they have will be answered. They will be thanked for their participation and any relevant information from this will be added to their hospital notes and be communicated to their GP where the patient consents for this to happen

5.5 RANDOMISATION

5.5.1 Randomisation

Randomisation will be via a centrally controlled web-based GCP compliant randomisation system, TRuST, run by Tayside Clinical Trials Unit (TCTU). Randomisation will be stratified by site. To ensure balanced assignment across critical variables, a minimisation algorithm will be employed, using baseline age, sex, SPPB score, grip strength and Charlson comorbidity score.

5.5.2 Treatment Allocation

The starting dose of perindopril will be 2mg daily which will be up-titrated to 4mg after 2 weeks if tolerated and renal function / potassium levels remain within defined limits (see Section 5.5.4 and 6.4). Participants in the placebo group will also undergo a ‘mock’ up-titration. If the 4 mg daily dose is not tolerated, participants will be down-titrated to the original 2mg daily. If 2mg is also not tolerated, medication will be withdrawn.

Medication will be dispensed by the Clinical Trials Pharmacist on receipt of a prescription with the participant number. Perindopril/placebo will be delivered to the participants on the day dispensed in sealed bottles with child proof caps. If the participant is unable to open child proof caps, a spare ordinary cap will be issued. Participants will be instructed to take one capsule every morning. This instruction is also printed on the bottle label.

Adherence will be checked and documented using tablet counts at each visit. If non-adherent, they will be encouraged to continue taking medication. If they persist as non-adherent, they will stay in the study, in order to do an “intention to treat” analysis.

Leucine and placebo will be dispensed as 400g tamper-evident tubs of identical powder (sufficient for 1 month) with a scoop to measure 4.5mls of powder (3 levelled scoops, 1.5ml each). Three level scoops will be taken with each meal. Instructions will be printed on the tub. Adherence will be checked by weighing the tubs at each study visit, with non-adherence treated as for perindopril.

5.5.3 Emergency Unblinding Procedures

Treatment allocation will be carried out by each centre's hospital pharmacy using the randomisation sequence provided by TCTU. TCTU will use an approved web-based randomisation program and will securely backup both the randomisation seed and the randomisation allocation. TCTU will provide each PI with a login to the IWRS, TRuST, for 24 hour emergency unblinding. If unblinding is required the PI or delegate should use the web-based unblinding system. In addition a paper copy of the allocation will be stored securely by Tayside Pharmaceuticals and an additional copy will be stored in NHS Tayside Clinical Trials Pharmacy. Unblinding will only be carried out where a physician considers that it is necessary for clinical safety. The unblinding procedure will be carried out according to the Sponsor SOP for Randomisation, Blinding and Code Breaking in Clinical Trials of Investigational Medicinal Products.

5.5.4 Withdrawal procedures

Participants will not receive further doses of perindopril medication or placebo if:

- They exhibit serum potassium levels of >5.5mmol/L,
- Serum sodium < 130mmol/L,
- Serum creatinine rises from baseline by >60 umol/L or serum creatinine is >180 umol/L
- If systolic BP falls to <90mmHg (lying) when on 2mg perindopril/placebo.

They will however remain in the study and undergo their remaining follow-up visits to preserve intention to treat analysis in these cases.

Participants will not receive further doses of leucine if:

- They undergo an adverse reaction (e.g. allergic reaction) attributable to leucine or leucine placebo

Withdrawal from one medication/placebo does not affect administration of the other medication/placebo, which will continue until the end of the trial.

The reasons for withdrawal will be noted in the participant's CRF and casenotes. If withdrawal is due to an AE it will be logged as such on the Adverse Event Log.
Participants are free to withdraw from the study at any time. Data collected up to the point of withdrawal will be kept on file unless the participant requests that it is removed. Tissue will also be kept in storage unless the patient participant requests that it is removed.

Although a participant is not obliged to give reason(s) for withdrawing prematurely, if the participant appears lost to follow up, the CI will make a **reasonable** effort to ascertain the reason(s), while fully respecting the individual's rights, and will demonstrate that everything possible was done in an attempt to find any participant lost to follow-up. Those lost to follow-up or withdrawn will be identified and a descriptive analysis of them provided, including the reasons for their loss and its relationship to treatment and outcome.

6 INVESTIGATIONAL MEDICINAL PRODUCT

6.1 STUDY DRUG

Tayside Pharmaceuticals will be manufacturing, labelling and blinding the study drugs. Tayside pharmaceuticals will deliver batches of study medication to individual trial site pharmacies, who will be responsible for local storage, dispensing, accountability and return of study drugs.

6.1.1 Study Drug Identification

Perindopril 2 mg tablets: over encapsulated in hard capsule
Perindopril 4 mg tablets: over encapsulated in hard capsule

Leucine as powder; supplied bulk in 400g pots

6.1.2 Study Drug Manufacturer

Tayside Pharmaceuticals, Ninewells Hospital & Medical School, Dundee will over encapsulate Perindopril tablets which have Marketing Authorisation.

6.1.3 Marketing Authorisation Holder

Tayside Pharmaceuticals, Level 5, Ninewells Hospital, Dundee DD1 9SY. MA 14076

6.1.4 Labelling and Packaging

Labelling and packaging will be performed by Tayside Pharmaceuticals as detailed above. Tablets will be included in packs containing the adequate number of units required for each treatment period. Both the IMP and placebo will be packaged for clinical use.

Labelling will be done in compliance with GMP (GMP; annex 13.) A description of the core text of the IMP labels is displayed below:

- Manufacturer name, address
- Pharmaceutical dosage form, route of administration, quantity of dose units, product name and strength
- Batch number
- Study number
- Study pack number
- Name of investigator
- Directions for use
- “For clinical trial use only” or similar wording
- Storage conditions
- Period of use (MM/YYYY format)
- “Keep out of reach and sight of children”

The PI, or designee, only will administer IMP/Placebo to patients included in this study. Each patient will only be given the IMP/placebo carrying his/her number. The administration for each patient will be documented in the CRF.

Patients will be supplied with study drug as detailed in the Study Operations Manual.

6.1.5 Storage

Study medication will be stored as each site’s Pharmacy/Clinical Trials Pharmacy as detailed in the IMP Management Plan. Study medications will be stored securely at between 15 ^{o}C and 25^{o}C, away from direct sunlight

6.1.6 IMP Safety Information

The most up to date version of the Summary of Product Characteristics (SmPC) for perindopril and the IMP Dossier will be held in the Pharmacy Site File (PSF), Trial Master File (TMF) and Investigator Site File(s) (ISF).

6.1.7 Accountability procedures

Drug accountability procedures will be overseen by each site's designated Clinical Trials Pharmacist or identified Pharmacist. Accountability Logs will be held on TRuST (Tayside web based randomisation and stock control system) and will include date, number and dose of capsules issued; date and number of capsules returned by the participant. Similarly, date of issue of each tub of leucine or placebo, date and weight of leucine/placebo returned by the participant will be recorded. The accountability procedures will be detailed in the IMP management plan adhering to TASC Policy on Drug Accountability for CTIMPs.

6.2 STUDY COMPARATOR

6.2.1 Comparator Identification

Placebo capsules (with an identical appearance to the active perindopril) and containing lactose.

Lactose powder, identical in appearance to leucine, supplied bulk in 400g pots

6.2.2 Comparator Manufacturer

Tayside Pharmaceuticals, Ninewells Hospital, Dundee DD1 9SY.

6.2.3 Labelling and Packaging

Will be performed by Tayside Pharmaceuticals, Ninewells Hospital, Dundee.

6.2.4 Storage

Placebo medication, as with study medication, will be stored as each site's Pharmacy/Clinical Trials Pharmacy as detailed in the IMP Management Plan. Study medications will be stored securely at between 15 and 25^{o}C, away from direct sunlight

6.2.5 Accountability procedures

As for active study medication, this will be under the supervision of the local Clinical Trials Pharmacist at each trial site as per the drug accountability and pharmacovigilance guidelines from TASC.

6.3 DOSING REGIME

a) Perindopril / placebo

Perindopril 2mg daily or placebo will be issued on the first convenient day following the initial screening/baseline visit and up to four weeks after screening visit.

Two weeks after this, a visit will be performed, with blood pressure, renal function and electrolytes checked. If medication is tolerated, Perindopril 4mg daily or placebo will be issued. This will be continued for 50 weeks.

b) Leucine / placebo

Leucine / placebo will be issued on the first convenient day following the initial screening/baseline visit and up to four weeks after screening visit. Three months supply at a time will be issued. The initial dose (2.5g three times per day) will be continued throughout the 1 year study period.

6.4 DOSE CHANGES

If there are significant increases in serum potassium (>5.0mmol/l but ≤5.5mmol/l), or in creatinine (>30 umol/L but ≤60 umol/l from baseline) participants will not be uptitrated but will be maintained on the Perindopril 2mg or placebo.

Participants who tolerated the initial dose of Perindopril 2mg daily or placebo but were unable to tolerate the higher dose of 4mg, due to significant increases in serum potassium (>5.0mmol/l but ≤5.5mmol/l), in creatinine (>30 umol/L but ≤60 umol/l from baseline) or drop in systolic blood pressure (<90 mmHg) will be downtitrated back to Perindopril 2mg or placebo.

Where there is down titration to the lower 2mg dose or discontinuation of perindopril or placebo these patients will be reviewed as guided by the local PI

Subjects will not receive further doses of medication or placebo if:

- They exhibit serum potassium levels of >5.5mmol/L,
- serum sodium < 130mmol/L,
- serum creatinine rises by >60 umol/L or serum creatinine is >180 umol/L
- if systolic BP falls to <90mmHg when on 2mg perindopril/placebo.

No changes to leucine dose are anticipated; intolerance will lead to leucine/placebo being discontinued as per section 5.5.4

6.5 PARTICIPANT ADHERENCE

Adherence to perindopril or placebo will be checked by tablet counting, and of leucine or placebo by weighing container tubs, at each safety visit. In addition, serum ACE activity will be measured as a further test of adherence to perindopril therapy; this has been shown to have good sensitivity and specificity to adherence to ACE inhibitors.

6.6 OVERDOSE

Overdose with perindopril can produce hypotension and impaired renal function which in severe cases may cause renal failure requiring dialysis. In the event of reported minor overdose (one extra capsule taken by mistake), participants will be assessed by the research team at home, GP surgery or local research unit, whichever is the more convenient. They will be given verbal instructions when seen by the research nurse to contact him/her should this occur. If haemodynamically stable, they will asked to stay at home for that day and drink plenty of fluids. If the participant is unwell or has taken a larger overdose, the RN will refer them to their GP who will refer them to A&E or else directly to A&E for admission and monitoring of blood pressure and renal function.

Overdose with leucine has not been associated with ill effects and requires no specific action. If the participant feels unwell after taking greater than the required dose, symptomatic treatment should be advised by the research team, with referral to the participants GP or hospital in case of persisting or worsening symptoms.

6.7 OTHER MEDICATIONS

6.7.1 Permitted Medications

We expect our target population to already be on multiple medications however they will be recruited only after assessment of risk by the local research team, such that the study medications are unlikely to interfere with their current medications.

Perindopril is a common drug that is used for cardiovascular conditions and it usually compatible with most other medications in older people. However patients will be monitored by the research team if they are started on concurrent diuretics, antihypertensive agents or

potassium sparing diuretics as these may interfere with renal function and blood pressure control.

6.7.2 Prohibited Medications

Potassium supplements: If potassium supplements are prescribed or taken in error, participants will be advised to stop the potassium supplements and trial medication will be withheld. Blood samples will be taken to measure serum potassium on the day of receiving this information. Further samples will be taken according to clinical indication depending upon the serum potassium levels. Medication will be reinstated when felt clinically appropriate.

Angiotensin receptor blockers, ACE inhibitors, aliskiren, spironolactone, other potassium-sparing diuretics: if any of these medications are introduced, the perindopril/placebo trial medication will be withdrawn. Participants will be encouraged to attend the follow up assessments and to continue the leucine/placebo trial medication.

The GP information letter and PIS will contain information on all the above points.

6.7.3 Concomitant Medications

Details of all concomitant medications will be recorded in the Case Report Form (CRF).

7 STUDY ASSESSMENTS

7.1 STUDY ASSESSMENTS

Study assessments are detailed in the study matrix (Appendix 2)

7.2 SAFETY ASSESSMENTS

Safety assessments will be conducted at each study visit. Reported or observed side effects will be recorded.

ACE inhibitors have been used for cardiovascular diseases for almost two decades now. They have been well tolerated by older people and are known to reduce the risk of cardiovascular events even in this population. The participants being recruited to this study are likely to suffer from multiple comorbidities in the form of chronic diseases including cardiovascular diseases.

Severe side effects are uncommon. The list of side effects is given in the SmPC. The most commonly encountered side effects are cough, hypotension, worsening renal function and hyperkalemia. Participants will be monitored at 2 weeks, 5 weeks, 3, 6, 9 and 12 months to detect any deterioration in renal function and blood pressure will be measured at these time points.

Medication will be withdrawn if serum potassium increases >5.5mmol/l, serum sodium falls <130 mmol/l, serum creatinine increases by >60 umol/l from baseline or is >180umol/l. Medication will be withdrawn if systolic blood pressure drops <90mmHg on the lower dose of 2 mg Perindopril or placebo.

Some participants may develop a dry cough which can disappear on continuation of the medication. However in those with persistent dry disabling cough, medication will be discontinued.

Participants who withdraw medication will be encouraged to maintain the follow up home visit assessments.

Leucine is an amino acid present in the diet at levels similar to those administered in this trial. No specific side-effects or risks have been identified with the use of leucine at this dose.

Safety follow-up:

Any adverse events will be followed up until resolution. As safety assessments will be carried out at the final visit, last day of study medication, no further safety assessments will be carried out beyond those outlined above unless felt clinically necessary. Where any of the safety assessments at the final visit are thought to be of clinical importance these will be followed up beyond the study period by the study team. Where appropriate this may include referral to other hospital specialities or for GP follow up.

8 DATA COLLECTION & MANAGEMENT

8.1 DATA COLLECTION

It is the CI responsibility to ensure the accuracy of all data entered and recorded in the CRFs and the database. The Delegation of Responsibilities Log will identify all trial personnel responsible for data collection, entry, handling and managing the database.

The data will be collected by the RN, the Site PI and/or or other local delegated individual either directly onto a paper CRF with subsequent transcription to the eCRF, or direct data entry onto the web based eCRF. Where there is electronic storage of non-identifiable data it will be on a password protected device and/or database.

The study questionnaires will be completed at each study visit by the patient with the assistance of the research nurse, directly onto a paper format with subsequent transcription to the eCRF.

NHS laboratory derived blood tests and DXA results will be held on local NHS clinical systems databases in an identifiable format and for an indefinite time frame which can be assessed by primary and secondary care practitioners for future health care of patients. All research data and data established from the NHS tests will be stored in an unidentifiable format on password protected disaster recovery formatted OpenClinica database on a University of Dundee server. Quality control of data will be maintained by the Data Monitoring Committee. Patients will be informed of data storage and consent will be sought.

All research blood samples (link-anonymised) will be processed at each site (as per Working Practice Guidelines in the Study Operations Manual) and transported to the relevant laboratory for analysis at the end of the study for batch analysis. Depending upon volume and composition of each blood sample additional/surplus blood serum/plasma samples will be stored (with consent) and transported at the end of the study to the clinical laboratory at University of Dundee under the custodianship of the CI for future research which will be scientifically and ethically reviewed.

Samples for myostatin analysis will be batched.and stored for future analysis.

The medical notes can act as source data for past medical history, subsequent medical conditions, hospital admissions, diagnostic reports and blood and urine results.

8.2 DATA MANAGEMENT SYSTEM

A data management system will be provided by TCTU using OpenClinica. The study system will be based on the protocol and case report form (CRF) for the study and individual requirements of the investigators. Development and validation of the study database and QC and extraction of data will be done according to TCTU procedures and TASC SOPs. Extracts for analysis will be based on the dummy data tables provided by the study team.

9 STATISTICS AND DATA ANALYSIS

9.1 SAMPLE SIZE CALCULATION

We have taken a deliberately conservative approach and used the minimum clinically important difference (MCID) for SPPB to ensure we have proof of efficacy. In order to detect the MCID in SPPB of 0.5 points (anticipated SPPB of 8 in placebo group and 8.5 in intervention group, SD

= 2.7 points) with a power of 90% at alpha 0.05, and assuming a correlation between time points of 0.7 as seen from our previous work, we would require a total of 352 participants for each of the 4 groups (88 per group). Assuming 20% dropout at 12 months, we would therefore need to recruit 440 patients. This sample size would also have 90% power to detect a 5% difference in muscle mass at 12 months, assuming a baseline value of 19kg (SD 2.8)

Biomarkers substudy: Assuming an effect size (F2) of 0.1 (equivalent to a 15% difference between groups with a standard deviation equivalent to half the baseline mean) for each component in a multivariable linear regression, and 20 variables (including baseline demographic, functional and biochemical variables), to provide a model with 90% power at alpha = 0.05 would require 278 participants; 20% dropout would require that 350 participants were enrolled. With a total sample size of 440 participants, 90% power would be achieved for up to 35 variables.

The smaller numbers utilised in analyses including angiotensin fragments and microRNAs will still deliver adequate power for both univariate and multivariate analyses. Studies by our group of microRNA changes in COPD have shown a greater than twofold difference between patients and controls. Hence an effect size (F^2) of 0.2, or 40% difference between groups for each marker, would comfortably detect this difference in a multivariable linear regression model of 20 variables; such a model would have 90% power to detect this effect size with a sample size of 148 participants.

The Derby substudy is powered on finding a correlation coefficient (r) of 0.5 or greater between DEXA muscle mass and D3 creatine measured muscle mass. To detect r=0.5 with 80% power, assuming alpha=0.05 requires 29 participants. We will aim to recruit a minimum of 30 participants into the Derby substudy.

9.2 PROPOSED ANALYSES

Analyses will be according to Intention-to-Treat and comply with the ICH E9 'Statistical Principles for Clinical Trials'. A 2 sided p value of <0.05 will be taken as significant for all analyses. The primary analysis will be a repeated measures mixed model between-group comparison of SPPB utilising all available data points during follow-up. Initially, a test for treatment interaction will be carried out and if not significant the main analysis will proceed using the full power of the factorial design. Between-group differences for all primary and secondary outcomes will be adjusted for baseline values, age, sex and minimisation variables.

Secondary outcomes will be analysed with similar methodology using repeated measures mixed model between-group comparisons. Unadjusted and adjusted analyses will be presented as above. Sensitivity analyses will be performed to further test the effect of missing data by assigning worst possible result status to missing datapoints. A supplementary per-protocol analysis will be performed to examine adverse events in those taking at least 80% of study medication.

No interim analyses are planned. Exploratory subgroup analyses will be performed, including examining differences in treatment effects by age (above and below median), SPPB >8 or 8 and below, by sex, and by body mass index. A statistical analysis plan will be prepared and finalised prior to trial database lock.

Biomarkers substudy: Univariate analyses examining the interaction between a) individual baseline biomarker values and treatment effect for muscle mass and SPPB at 12 months, and b) biomarker change between baseline and 3 months and treatment effect for muscle mass and SPPB at 12 months will be conducted. In addition, multivariable analysis for a) baseline values including age, sex, SPPB, grip, muscle mass, albumin, creatinine, all measured biomarkers and treatment group will be conducted to examine whether a) baseline values and b) change at 3 months, predicts SPPB change and muscle mass change at 12 months.

Derby substudy: Correlational analysis (Pearson's correlation coefficient, and also Bland-Altman plots) will be derived to compare muscle mass measured by DEXA at each timepoint with D3 creatine measures of muscle mass. The effect of the study interventions on muscle

mass (D3 creatine) and muscle protein turnover (3-methylhistidine) will be analysed as for the main study outcomes using repeated measures mixed-model methods.

9.3 MISSING DATA

Missing data will be handled in the analyses via two sensitivity analyses; a) by multiple imputation, and b) by assigning the worst possible value to missing data

9.4 TRANSFER OF DATA

Data transfer will occur via the secure, web-based electronic CRF hosted by Tayside Clinical Trials Unit. Data that are not part of the CRF (e.g. batched biomarker results analysed at the end of the trial) will be analysed via their unique trial number, and results without personal identifiers will be transferred to TCTU via encrypted spreadsheet for amalgamation with the main trial database.

10 ADVERSE EVENTS

10.1 DEFINITIONS

An **adverse event** (AE) is any untoward medical event affecting a clinical trial participant. Each initial AE will be considered for severity, causality or expectedness and may be reclassified as a serious event or reaction based on prevailing circumstances.

An **adverse reaction** (AR) is where it is suspected that an AE has been caused by a reaction to a trial drug

A **serious adverse event** (SAE), **serious adverse reaction** (SAR) or **suspected unexpected serious adverse reaction** (SUSAR) is any AE, AR or UAR that at any dose:

- results in death
- is life threatening
- requires hospitalisation or prolongation of existing hospitalisation
- results in persistent or significant disability or incapacity
- is a congenital anomaly or birth defect
- Or is otherwise considered serious

Note: Hospitalisations for treatment planned prior to randomisation and hospitalisation for elective treatment of a pre-existing condition will not be considered as an SAE. However any adverse events occurring during such hospitalisation will be recorded.

10.2 RECORDING AND REPORTING AES AND SAES

All AEs and SAEs will be recorded from the time a participant consents to join the study until the last study visit. Participants with unresolved AEs at the last study visit will be followed up until resolution or 30 days after last visit of that participant, whichever is sooner. SUSARS will be followed until resolution.

The CI, PI or delegate will ask about the occurrence of AEs and hospitalisations at every visit during the study. AEs will be recorded on the AE Log in the CRF. Immediately reportable SAEs will be submitted on an SAE form to the Sponsor Safety Section (pharmacovigilance.tayside@nhs.net) within 24 hours of becoming aware of the SAE. SAEs will be assessed for expectedness and causality by the Investigator. The evaluation of expectedness will be made based on the knowledge of the reaction and the relevant product information (SmPC or IMP Dossier). Refer to TASC SOP 11 “Identifying, Recording and Reporting Adverse Events for CTIMPs”.

Due to the large amount of comorbid disease and very high levels of illness and adverse events that we expect to be present in this population, we will record as Adverse Events, but not report as SAEs to Sponsor, in the following categories:

- Any death or hospitalisation due to new cardiovascular event (with the exception of a) angioedema, and b) symptomatic hypotension as a primary cause, which will be reportable as an SAE)
- Any death or hospitalisation due to new diagnosis or treatment of cancer
- Any death or hospitalisation due to fall or fracture
- Any death or hospitalisation due to infection
- Any death or hospitalisation due to exacerbation of an existing medical condition.
- Any admission for elective or planned investigation or treatment
- Any death or hospitalisation for deteriorating renal function, high or low potassium levels
- Any hospitalisation due to nausea, vomiting, constipation or diarrhoea

10.3 REGULATORY REPORTING REQUIREMENTS

The Sponsor, together with the CI, is responsible for reporting SUSARs to the competent authority, the MHRA, the Research Ethics Committee (REC) and any other competent authorities. Fatal or life threatening SUSARs will be reported within 7 days and non-fatal and non-life threatening SUSARs within 15 days.

10.4 ANNUAL REPORTING REQUIREMENTS

The following reports will be submitted each year as a condition of the authorisation to undertake a clinical trial or as a condition of a favourable opinion from a REC.

The Development Safety Update Report (DSUR) will be prepared jointly by the TASC Safety Section and CI and submitted to the MHRA on the anniversary of date of Clinical Trial Authorisation (CTA).

An NRES CTIMP Safety Report Form will be sent to REC along with the DSUR. Reports of SUSARs in the UK, urgent safety measures and any other safety reports submitted, for example, reports of a data monitoring committee, will also be accompanied by a Safety Report Form.

A NRES Annual Progress Report for CTIMPs will be prepared and submitted by the CI to REC, and copied to Sponsor, on the anniversary date of the REC favourable opinion.

10.5 URGENT SAFETY MEASURES

The CI, PI or other clinician may take appropriate immediate urgent safety measures in order to protect the participants of a CTIMP against any immediate hazard to their health or safety. The MHRA, REC and Sponsor will be notified in writing within three days.

11 PREGNANCY

Pregnancy is not considered an AE or SAE, unless there is a congenital abnormality or birth defect. Any unexpected pregnancy occurring during the clinical study and the outcome of the pregnancy, will be recorded on a TASC Pregnancy Notification Form and submitted to the TASC Safety Section (pharmacovigilance.tayside@nhs.net) within 24 hours of becoming aware of the pregnancy. The pregnancy will be followed up until the end of the pregnancy. If the study participant is a male, informed consent for follow up must be sought and obtained from his female partner.

Pregnancy is considered an unlikely event in the context of this trial, although pregnancy in the partner of a male participant is possible.

12 TRIAL MANAGEMENT AND OVERSIGHT ARRANGEMENTS

12.1 TRIAL MANAGEMENT GROUP

The trial will be co-ordinated by a Trial Management Group, consisting of the grant holders and Trial Manager. Local external PIs and research nurses will be invited to attend the Trial Management Group, as will selected Clinical Trials Unit staff as required.

12.2 TRIAL MANAGEMENT

A Trial Manager will oversee the study and will be accountable to the CI. The Trial Manager will be responsible for checking the CRFs for completeness, plausibility and consistency. However, this will remain the overall responsibility of the CI. Any queries will be resolved by the CI or delegated member of the trial team.

A study-specific Delegation Log will be prepared for each site, detailing the responsibilities of each member of staff working on the trial.

12.3 TRIAL STEERING COMMITTEE

A Trial Steering Committee (TSC) will be established to oversee the conduct and progress of the trial a lay representative will be included in the TSC. The terms of reference of the TSC and the draft template for reporting will be held in the TMF.

12.4 DATA MONITORING COMMITTEE

An independent Data Monitoring Committee (DMC) will be established to oversee the safety of trial participants. The terms of reference of the DMC will be detailed in the Trial Master File.

12.5 INSPECTION OF RECORDS

The CI, PIs and all institutions involved in the study will permit trial related monitoring, audits, REC review, and regulatory inspection(s). In the event of an audit, the CI will allow the Sponsor, representatives of the Sponsor or regulatory authorities direct access to all study records and source documentation.

12.6 RISK ASSESSMENT

A study risk assessment was carried out by the TASC Research Governance Manager prior to Sponsorship approval being granted.

12.7 STUDY MONITORING

The Sponsor has determined the appropriate extent and nature of monitoring for the trial and will appoint appropriately qualified and trained monitors.

12.7.1 Potential Risks

ACE inhibitors have been used for cardiovascular diseases for almost two decades now. They have been well tolerated by older people and are known to reduce the risk of cardiovascular events even in this population. The participants being recruited to this study are likely to suffer from multiple comorbidities in the form of chronic diseases including cardiovascular diseases.

Severe side effects are uncommon. The list of side effects is given in the SmPC. The most commonly encountered side effects are cough, hypotension, worsening renal function and

hyperkalemia. Some participants may develop a dry cough which can disappear on continuation of the medication. However in those with persistent dry disabling cough, medication will be discontinued.

ACE inhibitors are sometimes stopped in people with falls due to postural hypotension since they can lower blood pressure. However recent evidence suggests that these medications may not actually increase the risk of postural hypotension. The study investigators have previously used ACE inhibitors in a similar population with no increased risk. This study will monitor both postural blood pressure and falls.

Participants who are withdrawn will be encouraged to attending the follow up assessments.

The participant group in this trial are old, frail and may have many co-morbidities. Participants will be expected to attend hospital on 3-4 occasions with an additional 5 visits which may be carried out in the patient's home / GP surgery/ hospital and undergo blood tests.

Participants will be asked to keep a diary of falls, hospital and GP visits and record any noticeable change in health status.

Reference Safety Information for Leucine:

Leucine is an essential dietary amino acid and forms part of a normal balanced diet. It is found in most foodstuffs, and daily ingestion of leucine in a normal balanced diet is similar to the quantity to be ingested in this trial (e.g. wild salmon contains 2g/100g of meat).

Ingestion of leucine as 5% of total diet for 13 weeks caused no adverse effects in any organ system in rats[59]. Human studies revealed no gastrointestinal, metabolic or hepatic side-effects in adult males given 500mg/Kg/day of leucine (a dose three times higher than we propose in this trial)[60,61]

No adverse effects are known or expected to arise from ingestion of leucine at the doses proposed in this trial, hence no expected suspected adverse reactions are listed.

12.7.2 Minimising Risk

We will exclude participants at a higher risk of side effects: Already in receipt of ACE inhibitor or ARB; contraindication to ACE inhibitor use (significant aortic outflow obstruction; eGFR <30ml/hr; serum potassium >5.0mmol/l; serum creatinine >170 umol/L; systolic BP <90mmHg)

Participants will be monitored at 2 weeks, 5 weeks, 3, 6 and 9 months and final visit at 12 months to detect any deterioration in renal function and blood pressure will be measured at these time points. See section 6.4 for criteria for dose changes and withdrawal.

Careful monitoring of patients throughout the trial with dedicated care and adverse event monitoring will be undertaken as required.

Patients will receive a card containing all study staff contact details to enable patients to contact staff with any concerns.

All patients will have access to taxi transport for each visit to aid accessibility and comfort. Where the participant agrees, visits 3, 4, 5, 6 and 8 may be carried out in the participants home or GP surgery to minimise the burden of hospitals visits for the participant.

Risks of venepuncture will be discussed with the patients.

Members of the research team carrying out home visits will work according to local guidelines for lone working, transportation of blood and transportation of IMP.

13 GOOD CLINICAL PRACTICE

13.1 ETHICAL CONDUCT OF THE STUDY

The study will be conducted in accordance with the principles of good clinical practice (GCP). In addition to Sponsorship approval, a favorable ethical opinion will be obtained from an appropriate REC. Authorisation from an appropriate competent authority(s) and appropriate NHS R&D permissions(s) will be obtained prior to commencement of the study.

13.1.1 Confidentiality

All laboratory specimens, evaluation forms, reports, and other records will be identified in a manner designed to maintain participant confidentiality. All records will be kept in a secure storage area with limited access to study staff only. Clinical information will not be released without the written permission of the participant, except as necessary for monitoring and auditing by the Sponsor, its designee or Regulatory Authorities. The CI and study staff involved with this study will not disclose or use for any purpose other than performance of the study, any data, record, or other unpublished, confidential information disclosed to those individuals for the purpose of the study. Prior written agreement from the Sponsor or its designee will be obtained for the disclosure of any said confidential information to other parties.

13.1.2 Data Protection

The CI and study staff involved with this study will comply with the requirements of the Data Protection Act 1998 with regard to the collection, storage, processing and disclosure of personal information and will uphold the Act's core principles. The CI and study staff will also adhere, if appropriate, to the current version of the NHS Scotland Code of Practice on Protecting Patient Confidentiality. Access to collated participant data will be restricted to the CI and appropriate study staff.

Computers used to collate the data will have limited access measures via user names and passwords.

Published results will not contain any personal data that could allow identification of individual participants.

13.1.3 Insurance and Indemnity

The University of Dundee and NHS Tayside Health Board are Co-Sponsoring the trial under the aegis of Tayside Academic Science Centre (TASC).

Insurance. The University of Dundee will obtain and hold Professional Negligence Clinical Trials Insurance cover for legal liabilities arising from the study.

Tayside Health Board will maintain its membership of the Clinical Negligence and Other Risks Insurance Scheme ("CNORIS") which covers the legal liability of Tayside in relation to the study.

Where the study involves University of Dundee staff undertaking clinical research on NHS patients, such staff will hold honorary contracts with Tayside Health Board which means they will have cover under Tayside's membership of the CNORIS scheme.

Indemnity. The Co-Sponsors do not provide study participants with indemnity in relation to participation in the Study but have insurance for legal liability as described above.

14 STUDY CONDUCT RESPONSIBILITIES

14.1 PROTOCOL AMENDMENTS

The CI will seek Sponsor approval for any amendments to the Protocol or other study documents. Amendments to the protocol or other study docs will not be implemented without approval from the Sponsor and subsequent approval from the appropriate REC and/or Regulatory Authority, as appropriate, and NHS R&D Office(s). Refer to TASC SOP 26 “Amendments to CTIMPs”

14.2 PROTOCOL DEVIATIONS, BREACHES AND WAIVERS

The CI will not implement any deviation from the protocol without agreement from the Sponsor, except where necessary to eliminate an immediate hazard to trial participants.

In the event that the CI needs to deviate from the protocol, the nature of and reasons for the deviation will be recorded in the CRF, documented in a TASC Deviation & Breach Log and notified to the Sponsor. If this necessitates a subsequent protocol amendment, this will be submitted to the Sponsor for approval and then to the appropriate REC, Regulatory Authority and local NHS R&D for review and approvals as appropriate. It is Sponsor policy that waivers to the Protocol will not be approved.

In the event that a serious breach of the protocol or GCP is suspected, this will be reported to the Sponsor immediately using the form “Notification to Sponsor of Potential Serious Breach or Serious Deviation”. Refer to TASC SOP 25 “Escalation and Notification of Serious Breaches of GCP or the Trial Protocol for CTIMPs”.

14.3 STUDY RECORD RETENTION

Archiving of study documents will be carried out as specified in TASC SOP 13: Archiving Clinical Research Data for Clinical Trials of Investigational Medicinal Products. All study documentation will be kept for at least 15 years.

14.4 END OF STUDY

The end of study is defined as last patient last visit. The Sponsor, CI and/or the TSC have the right at any time to terminate the study for clinical or administrative reasons.

The end of the study will be reported to the Sponsor, REC, Regulatory Authority and NHS R&D Office(s) within 90 days, or 15 days if the study is terminated prematurely. The CI will ensure that any appropriate follow up is arranged for all participants.

A final report of the study will be provided to the Sponsor, REC and Regulatory Authority within 1 year of the end of the study.

14.5 CONTINUATION OF DRUG FOLLOWING THE END OF STUDY

The decision as to whether to continue either perindopril or leucine after the end of each participants study participation will reside with the primary and secondary care team providing usual care to the participant. No provision for continuation will be made by the trial team or Sponsor.

15 REPORTING, PUBLICATIONS AND NOTIFICATION OF RESULTS

Regular progress reports will be sought by the funder as outlined in its programme policy. Advice on the scheduled dates will be provided by the NIHR EME Programme.

15.1 AUTHORSHIP POLICY

Ownership of the data arising from this study resides with the study team and their respective employers. On completion of the study, the study data will be analysed and tabulated, and a clinical study report will be prepared.

Authorship eligibility for each manuscript arising from this study will be determined according to the criteria laid out in the Working Practice Document on Authorship filed in the Study Operations Manual.

15.2 PUBLICATION

The clinical study report will be used for publication and presentation at scientific meetings. Trial investigators have the right to publish orally or in writing the results of the study.

Summaries of results will also be made available to Investigators for dissemination within their clinical areas (where appropriate and according to their discretion).

15.3 PEER REVIEW

Peer review of the protocol is via the UoD-NHST Joint Sponsorship Committee. The resulting publication will be reviewed by the referees of the journal to which the paper (and its protocol) will be submitted.

The trial is funded by the NIHR Efficacy and Mechanisms Board, which has peer reviewed the grant application.

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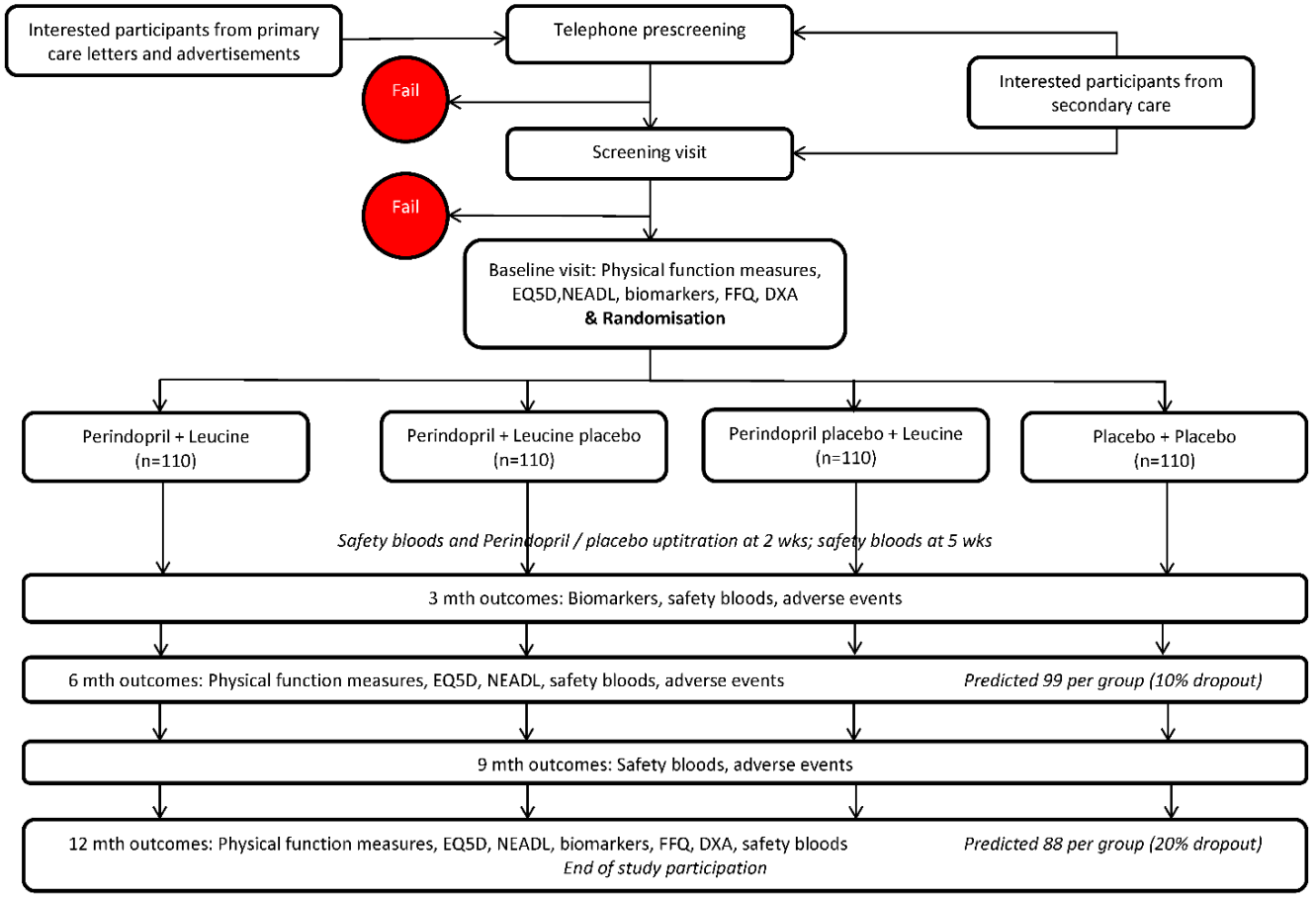
APPENDIX 1: STUDY FLOW CHART



APPENDIX 2: STUDY VISIT MATRIX

	Visit 1[c]	Visit 2[cf]	Visit 3[h]	Visit 4[h]	Visit 5[h]	Visit 6[hf]	Visit 7	Visit 8[h]	Visit 9[f]
	Screening	Baseline Up to 4 weeks after screening	2wks + 1 week	Visit 3 + 0-5 days	5wks +/- 1 week	3mths +/- 2 weeks	6 mths +/- 2 weeks	9 mths +/- 2 weeks	12 mths +/- 2 weeks
Consent	X								
Inclusion exclusion	X	X							
Demographics	X								
Medical History	X								
Review/record concomitanmeds	X	X	X	X	X	X	X	X	X
Height	X								X
Weight	X						X		X
Blood pressure	X		X		X	X	X	X	X
Bloods (safety): Sodium, potassium, creatinine, urea, eGFR, albumin,	X		X		X	X	X	X	X
Bloods (safety): glucose		X				X			X
Bloods (IMP compliance) – serum ACE							X		X
Bloods(biomarkers)		X				X			X
BIA	X						X		X
SPPB	X						X		X
Handgrip	X						X		X
Quads strength		X					X		X
6MWD		X					X		X
NEADL		X					X		X
EQ-5D		X					X		X
DXA		X							X
FFQ		X							X
Charlson comorbidity score	X								
Falls diary		X				X	X	X	
Randomisation		X							
Supply meds [g]		X		X		X	X	X	
Adverse events		X	X	X	X	X	X	X	X
Adherence			X	X	X	X	X	X	X

c – screening and baseline visit may be combined where eligibility bloods are known within 4 weeks
h- visit may be in participants home or GP surgery if convenient
f – patients will be asked to fast from midnight prior to attending for a morning appointment and after a light breakfast for an afternoon appointment
g – IMP can be issued within 24 hours of the visit taking place

APPENDIX 3: SUB STUDY MATRIX

	Day before baseline visit	Baseline visit	Baseline + 2 days	Day before Visit 7	Visit 7 6 months	Visit 7 + 2 days	Day before Visit 9	Visit 9 12 months	Visit 9 + 2 days
	Extra visit to centre		*Post the urine sample*	*Extra visit to centre*		*Post the urine sample*	*Extra visit to centre*		*Post the urine sample*
*Blood samples**	X	X		X	X		X	X	
Urine sample	X		X	X		X	X		X
Tracer Drink +	X			X			X		
24 hour urine collection	X			X			X		

* Biomarkers for muscle protein
+30mg D3-creatine and 10mg 3-MH

APPENDIX 4: THE SARC-F QUESTIONNAIRE

Strength: How much difficulty do you have in lifting and carrying 10 pounds / a bag of shopping?

None:	0
Some:	1
A lot or unable:	2

Assistance in walking: How much difficulty do you have walking across a room?

None:	0
Some:	1
A lot, use aids, or unable:	2

Rise from a chair: How much difficulty do you have transferring from a chair or bed?

None:	0
Some:	1
A lot or unable without help:	2

Climb stairs: How much difficulty do you have climbing a flight of 10 stairs?

None:	0
Some:	1
A lot or unable:	2

Falls: How many times have you fallen in the past year?

None:	0
1 to 3 falls:	1
4 or more falls:	2