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Study Protocol	
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1. ABSTRACT

Muscle loss with age is considered to be an important cause of disability in older adults1, 2. However, current tools frequently used and recommended to measure muscle mass in trials have limitations: For example, DEXA (Dual-energy X-ray absorptiometry) and BIA (Bioelectrical impedance analysis)- both widely used in clinical trials cannot distinguish muscle tissue from non-muscle elements such as subcutaneous and intramuscular fat, skin, water and connective tissue3-5

Deuterated creatine (D3Cr) dilution is a novel promising method that provides an accurate measure of the functional contractile tissue, without including the non-contractile elements, by measuring total body creatine. Our preliminary cross-sectional results in men aged >65 years show functional muscle mass (FMM) assessed by D3-Cr to be significantly associated with performance outcomes (SPPB, 400m walk, and chair stands) and long-term outcomes (incident mobility problems, serious injurious falls), while DEXA showed no associations. However, these are observational data and cannot determine a causative role of muscle per se on functional outcomes.

Hence, the objective of the pilot study is to assess the feasibility of our proposed future study to evaluate the role of muscle mass on functional outcomes. Specifically, the pilot study will randomize moderate to low-functioning older adults to a resistance training program or successful aging program to assess recruitment yields, participant adherence, retention, training program design aspects, sample size, and the cost of the main trial. Further, the impact of these changes in FMM on short-term performance outcomes, such as strength, walking speed, SPPB, self-reported measures, and balance will be examined.

2. BACKGROUND

It has now been well established that there is a progressive loss of muscle mass with aging. Prospective studies have shown this decline to be around 1% per year for both men and women[6-8]. This dramatic change in anatomy with age has prompted the coining of the term *sarcopenia (sarx: flesh; penia: loss)*[9] Although not the sole cause of strength loss in older adults, the loss of muscle mass is thought to be a major cause of the decline in strength observed with age. Consequently, the loss of muscle and strength has direct adverse consequences on physical function leading to disability and mobility impairments[1, 2]. The recent establishment of ICD-10 code by CDC recognizing sarcopenia as an "independent reportable condition" further emphasize the role of muscle and muscle loss as major medical concern that needs immediate attention[10].

But, current tools frequently used and recommended to measure muscle mass in trials have limitations[11-13].

- DEXA and BIA- both widely used in clinical trials and recommended to diagnose sarcopenia, cannot distinguish muscle tissue from non-muscle elements such as subcutaneous and intramuscular fat, skin, water and connective tissue[3, 14].

- Magnetic resonance imaging (MRI) and computed tomography (CT) can detect non-muscle tissue in body segments but are extremely expensive and cumbersome when the analysis involves total body skeletal muscle[15].

Deuterated creatine (D_3Cr) dilution is a novel promising method that provides an accurate measure of the functional contractile tissue, without including the non-contractile elements, by measuring total body creatine. Briefly, the method involves ingestion of deuterated non-radioactive, labeled-creatine in a capsule and subsequently measuring total creatine from a single urine sample, without any special diet restrictions; thus making the method quite safe and feasible. Considering there is no gold standard *in vivo* measurement of total body skeletal muscle mass, construct validation studies have shown a strong correlation with MRI in both men and women[16] (young and old) ($r = 0.87$) and in animal studies[17] ($r = 0.95$). More importantly, our preliminary cross-sectional results in community-dwelling men aged >65 years show functional muscle mass (FMM) assessed by D_3-Cr to be significantly associated with physical performance (SPPB, 400m walk, and chair stands) and long-term outcomes (incident mobility problems, serious injurious falls), while DEXA showed no associations.

However, these observational data cannot determine a causative role of muscle *per se* on functional outcomes. Additionally, there are no longitudinal or experimental studies that establish D_3-Cr as a standard measure of functional muscle mass (FMM). Our future study will use an experimental approach to evaluate the causative role and time course of FMM changes with D3-Cr on functional outcomes. The accomplishment of this future longitudinal study will also validate this promising method for characterizing FMM in response to muscle-targeted interventions for low functioning older adults (e.g., selective androgen receptor modulators). A pilot study will fill necessary gaps that include feasibility and within-person variability of the D3-Cr method that is currently unknown. Hence, the objective of the pilot study is to assess the feasibility of our proposed future trial.

Achieving the above aims will help in gathering feasibility information for our future study, but also provide novel insights into the time course of early FMM to resistance training and its relation to short-term outcomes. In addition, future studies can use D_3-Cr to evaluate the effectiveness of interventions to increase functional muscle mass.

3. SPECIFIC AIMS / OBJECTIVES

Primary objective

- To evaluate the feasibility of implementing the D3Cr method in a resistance training trial as it relates to participant adherence, retention, training program design aspects (intensity, volume, frequency & safety) sample size, and cost of the main study.

Secondary objectives

The secondary objectives are to:

- Evaluate and compare the time-course adaptations in skeletal muscle mass measured by D3-Cr and DEXA
- Correlate the **changes** in functional muscle mass with the change in walking speed, SPPB, strength, and balance.

4. RESEARCH PLAN

Trial Design

- The pilot study will be single-blind, parallel group, randomized controlled trial of **24 moderate to low functioning** participants randomized in a 1:1 allocation ratio to a high-intensity Resistance Training group (RT) or a Health Education comparator group (HE) for **14 weeks.** We chose the specified population since they are at a greater risk of disability and is often excluded from trials[18].

Scehdule of Events

Table 1. Study visit and schedule

Schedule of Events				Training Intervention					
				Preparatory Phase		Training Phase			
	Phone scr.	Scr. Visit	Baseline	Week 1	Week 2	Week 8	Week 11	Week 14	Post-Test
Screening									
Basic eligibility phone screening	X								
Short consent		X							
Informed consent			X						
Physical exam, Medical History, Height			X						
Weight & Vitals			X			X			X
Interventions									
Resistance Training				←	—	—	—	→	
Successful Aging				←	—	—	—	→	
Assessments									
DEXA			X			X			X
D3Cr Dosing & Collection				X		X		X	
SPPB		X				X			X
Physical Functional Tests			X			X			X
Questionnaires			X			X			X

Participants

24 low functioning, both males and females, with 12 participants in each of the treatment arm.

Inclusion criteria

A participant must meet the following criteria to be eligible for inclusion in the study:

- men and women aged 70 or greater
- at high risk for mobility disability based on lower extremity functional limitations measured by Short Physical Performance battery (SPPB) with a score ≤ 8 out of 12
- could safely participate in the resistance training intervention as determined by medical history and physical examination.
- willing to give informed consent to be randomized to either the resistance training group or successful aging (Heath Education) comparison group and willing to follow the study protocol

Exclusion criteria

- Severe cardiac disease, including NYHA Class III or IV congestive heart failure, clinically significant aortic stenosis, history of cardiac arrest, use of a cardiac defibrillator, or uncontrolled angina
- Myocardial infarction, major heart surgery, stroke, deep vein thrombosis, or pulmonary embolus in the past 6 months
- Lung disease requiring either oral or injected steroids, or the use of supplemental oxygen
- Short, portable mental status questionnaire with 3 or more errors
- Severe arthritis (either osteoarthritis or rheumatoid arthritis) that severely limits mobility
- Severe lower back or shoulder pain that can worsen with weight lifting exercises.
- Cancer requiring treatment in the past 1 year (Melanomas excluded)
- Any present or recent history of severe psychiatric illness including depression that might preclude providing informed consent, safe participation, or compliance (self-report and investigator judgement
- Development of chest pain or severe shortness of breath on the 400 m self-paced walk test
- Parkinson's disease or other serious neurological disorders; renal disease requiring dialysis; other illness of such severity that life expectancy is considered to be less than 12 months
- Current diagnosis of schizophrenia, other psychotic disorders, or bipolar disorder
- Current consumption of more than 14 alcoholic drinks per week
- Uncontrolled hypertension (systolic blood pressure > 200 mm Hg and/or diastolic blood pressure >110 mm Hg)
- Currently on testosterone, Dehydroepiandrosterone (DHEA), or anabolic steroids.
- Undergoing physical therapy involving the lower extremities
- Currently enrolled in another randomized trial involving a pharmaceutical or lifestyle intervention. Observational studies are permitted
- Participation in progressive resistance exercise regimen (≥1 day/week) within the previous 6 months prior to screening.
- Weight change (intentional or not) over the last 6 months of > 5% of body or plan to lose or gain weight during the study
- Any other cardiovascular, pulmonary, orthopedic, neurologic, or other conditions that in the opinion of the local clinician would preclude participation and successful completion of the protocol

Temporary Exclusion Criteria

A person meeting any of the following temporary exclusion criteria at the time of screening will not be enrolled but may be re-screened later.

- Recent bacterial/viral infection (e.g. pneumonia) (< 2 weeks);
- Acute febrile illness in past 2 months;
- Severe hypertension, e.g., SBP > 200 mm Hg, DBP > 110 mm Hg
- Major surgery or fracture or hip/knee replacement (< 6 months);
- Hospitalization within the last 6 months (Not ER visits)

Premature Withdrawal from the Study: A participant has the right to withdraw from the study at any time, for any reason, and without repercussion. The investigators have the right to withdraw a participant from the study in the event of an intercurrent illness, AE, treatment failure, protocol deviation, and for administrative, or other reasons. An excessive rate of withdrawals would render the study uninterpretable; therefore, unnecessary withdrawal of participants should be avoided.

Sample Size: Our goal is to enroll a total of 24 participants. Since studies looking at changes in functional muscle mass in response to resistance training are not available, the sample size was arrived based on budgetary constraints and minimum number of participants required to meet feasibility goals. Assuming a conservative 20% loss to follow up, we estimate 12 participants per group (10 evaluable participants after the 20% loss due to attrition) would inform variance of the outcomes and feasibility for powering a future large-scale RCT.

Randomization: Randomization will be executed via a computer algorithm using random block lengths and stratified by gender. To ensure concealment of the allocation, the randomization procedures will be performed by an investigator who are not involved in the assessments

Recruitment

Participant recruitment will be coordinated in conjunction with the OAIC Clinical Research Core. Potential participants will be recruited from the general population in the North/Central Florida region through:

- IRB-approved advertisements (postcards, brochures, social media, newspaper ads) providing basic information about the study and inviting potential participants to call for additional information;
- Contact of potential participants who are enrolled in the Claude D. Pepper Recruitment Registry (IRB#417-2007) by Registry staff;
- We will also use existing entities of UF Health that include the Consent2Share program. Individuals who screen out of other studies may also be approached and asked about their interest in this study;

Screening process

We will employ a staged screening process where we will verify eligible participants over a series of contacts.

- Interested participants are first screened by phone. The phone interview is designed to exclude individuals who are ineligible due to safety concerns or not interested after learning about the study.
- After the phone screening, at the first in-person screening visit, we will consent participants with a short form and perform the SPPB test. Participant with SPPB score of ≤ 8 will be invited for the baseline visit.

Interventions

Resistance Training Intervention: The 14-week supervised resistance training intervention will include a full body, resistance training performed three days per week. The intensity, volume, tempo, and progression will be based on the Federal Physical Activity guidelines[19] as shown in Table 2.

Exercises: The resistance training exercises will include mainly multi-joint exercises using selectorized machines to minimize training duration and maximize the benefit. Each session will include upper body exercises (Chest Press, Shoulder Press, Seated Rows) and lower body exercises (Leg Press, Leg Extension and Leg Curls, and Calf raise) that will be tailored to the particpant needs. Inerventionists will suprvise all exercise sessions.

Warm up & cool down: Each training session will begin with 1 set of warm-up at a lighter load for 8–10 repetitions on 1 multi-joint upper body and 1 lower body machine. Each session will end with a 5 min cool-down involving stretching exercises and light calisthenics

Preparatory phase: Considering the participants are low functioning; we will use a 2-week *preparatory phase*, which will ensure sufficient time for connective tissue adaptations and for the participants to learn proper lifting form and breathing technique.

Training Phase: The training phase will last for 12 weeks and will be divided into 2 phases as shown in Table 2.

Intensity: The intensity will be maintained at 6-8 (moderate to vigorous RPE) as recommended by ACSM for older adults[20] and the sets will be done until volitional fatigue. The CR-10 BORG's scale ranges from 1-10 will be used to document perceived exertion during resistance training[21].

Progression: The load will increased when participants can perform all repetitions in all sets .

Table 2. Resistance training parameters

	Preparatory Phase	Training Phase	
	Week 1-2	Week 3-5	Week 5-14
Days/wk	3	3	3
Sets * Reps	1-2* 12-15	3 *12-15	3* 8-10
RPE	6 - 8	6 - 8	6 - 8
Rest (min)	1	1	1
Progression		Can do 3 sets of 15	Can do 3 sets of 10
Note: All values are subject to change based on participant availability and ability of the participant to conduct the number of sets and reps prescribed.			

Successful Aging: The comparator group will meet every 2 weeks. Weekly workshops will include flexibility exercises and presentation topics related to older adults, such as preventive services and screenings, finding credible health information, and so forth. We instruct all control participants not to change their PA levels and the importance of remaining weight stable throughout the study period. The rationale for the health education group in place of a sedentary control is to minimize the social interaction effects and to minimize participant drop-outs[22].

Outcomes & Questionnaires

All outcomes and questionnaires will be conducted at the Institute on Aging by trained and certified clinical site staff.

Medical history: A questionnaire that asks about health conditions will be administered prior to performing activities. Medical staff will evaluate participants with any health conditions (cardiovascular disease, falls, metabolic diseases, etc.) that are exacerbated with physical exertion.

Medication inventory: Many adults use both prescription and non-prescription pharmaceutical products. The use of these products is of interest for several reasons. All participants are asked to bring all prescription and non-prescription medications taken in the past two weeks to their first visit.

Short, portable mental status questionnaire: The questionnaire will be administered to identify potential participants with cognitive dysfunction who would pose a potential safety risk

PROMIS: The NIH developed Patient-Reported Outcomes Measurement Information System (PROMIS) physical function questionnaire uses item response theory (IRT) and computer adaptive tests (CAT) to improve precision and minimize respondent burden. The test has been shown to be reliable and valid across a range of population, including high-functioning and low functioning individuals [23] The following PROMIS tests will be used: physical Function, upper extremity, and mobility. All measures are administered via computer. Time to complete each measure ranges from 1-3 minute

Photograph Series of Daily Activities Scale - Short Electronic Version (PHODA-SeV) – The PHODA-SeV is an assessment of pain-related fear in the context of chronic back pain. The PHODA-SeV includes 40 photographs depicting common activities ranging from household chores to physical exercise. Using a 0–100 scale, respondents indicate their concern of harming their back when engaging in each depicted movement[25].

Five Facet Mindfulness Questionnaire (FFMQ) – The FFMQ is a reliable and valid comprehensive instrument for assessing different aspects of mindfulness. The FFMQ is based on a factor analytic study of five independently developed mindfulness questionnaires[26].

Tampa Scale for Kinesiophobia (TSK) – The TSK is a reliable and valid self-report checklist that uses a 4-point scale to measure of fear of movement or (re)injury. The scale is based on the model of fear avoidance, fear of work related activities, fear of movement and fear of re-injury. The TSK is considered as one of the most suited questionnaires to evaluate pain beliefs and fear of movement/(re) injury in chronic low back pain[27].

Borg Category Ratio (CR) 10 pain/RPE scale – The Borg CR10 pain/RPE scale is a reliable, valid, and frequently used quantitative measure of perceived exertion and pain during each exercise session and after the physical function tests. The Borg CR10 uses a category ratio scale that ranges from 0 (no pain/exertion) to 10 (maximal pain/exertion)[28].

Gait Analysis: We will ask participants to walk at their usual pace and a rapid pace two times across a carpet that contains sensors arranged in a grid pattern (48 X 288) (GAITRite Inc.). The system collects temporal (step time, gait cycle, single support time, double support time, stance time and swing time) and spatial (step length and stride length) parameters.

Vital signs: During screening, resting blood pressure and heart rate, height, weight will be measured. Blood pressure and heart rate are measured to assess eligibility criteria and for safety purposes and others are collected for descriptive purposes.

Oxygen Consumption: Oxygen consumption will be measured using a portable metabolic unit (Cosmed K4b2) for three daily tasks for 6 minutes each. Oxygen consumption will be measured breath-by-breath through a mask that is fitted to the participants face.

Mopping: Mop a 200 sqft room using mop and water
Lifting: Lift chairs and transfer across a 200 sq ft room

Muscle mass measurements:

- **D_3-Cr method:** The participant will ingest a 60-mg dose of deuterated creatine (D_3-Cr), a non-radioactive isotope, at the baseline assessment. They will return morning to provide a urine sample 3-5 days later. The sample will be frozen and deidentified for D_3-creatinine analysis. urine samples (-20^0) will be shipped to University of California, Berkeley where unlabeled creatinine and creatine will be measured using high-performance liquid chromatography(HPLC) and mass spectrometry (MS). The details of methods have been published previously.[16]
- **DEXA:** Total and regional body composition will be measured using at UF site Dual-energy X-ray absorptiometry (DEXA, QDR-4500w; Hologic, Waltham, MA, USA). DEXA is widely used in trials, is safe and is currently recommended to diagnose sarcopenia[11, 12].

Walk Tests: The 400m walk test is a feasible, objective, reliable, well-validated and used in large clinical trial. Participants will be asked to walk 400 m at their usual pace, without overexerting, on a 20-m course for 10 laps (40 m/lap)[29, 30]. The 6 min walk test will include walking at a rapid pace around 2 cones 20 m apart wearing the portable metabolic unit.

Physical performance will be measured using the Short Physical Performance Battery (SPPB), which is based on a timed 4-m walk, balance, and chair stand tests. This scale is reliable and valid for predicting institutionalization, mortality and disability[31, 32, 2933, 34] The score ranges from 0 (worst performers) to 12 (best performers).

Muscle strength will be measured using tests of isometric grip strength and isokinetic leg extension test. Grip strength is frequently used of upper body skeletal muscle function and has been widely used as a general indicator of functional status[35]. Grip strength will be measured using the Jammar dynamometer. Leg strength will be assessed using Isokinetic dynamometry (System 3 pro; Biodex Medical Systems, Shirley, NY, USA) at velocities of 60º and 120º per second[36].[2429]

Participants will perform 2 sets of 5 maximal isokinetic leg extensions at each velocity with one-minute of rest between each set for the dominant leg.

Dynamic balance will be measured using the Timed Up and Go test which is widely used to examine mobility and dynamic balance. Also, the test is recommended as a screening test for falls by the American Geriatric Society and the British Geriatric Society[37, 38]. Standing or static balance (narrow, tandem and semi-tandem) will also be assessed as part of the SPPPB test.

Adherence

Adherence involves both attendance to scheduled sessions as well as the quality of the training that occurs during each session. Adherence to the exercise protocols are affected by personal, social and environmental factors. At a personal level, it is important to ensure that the goals of each participant are realistic. We recognize that social interactions between staff and participant as well as the broader social environment surrounding the study influence adherence. All staff will be instructed to engage with and be supportive toward all participants. We will also adopt a number of other strategies to improve attendance and quality of training sessions. Some strategies include, having trained interventionists to supervise training, including a preparatory phase to educate the participants about proper lifting technique, tailoring the training program based on the physical needs of the participant, maintaining a workout log to help motivation, monitoring missed visits so that the study coordinator can take prompt action and so forth

The physical environment and accessibility influence adherence. This includes such factors as ease of parking, transportation, and accommodations made for friends or significant others who may come to

training facilities to support participants via transportation, etc. At the time of baseline assessments, staff discuss these topics with each participant to be certain there are no significant barriers in this domain and/or to problem solve potential challenges that exist.

The comparator group will not participate in a resistance raining intervention and hence pose a greater risk of dropping out. We expect, based on our previous experiences, that the flexibility exercises and health education classes offered every 2 weeks will serves an effective “active control” that will help to promote retention in the study. We will also provide an exercise prescription and education materials at the end of the study duration so that they can start a resistance training program outside the site.

Retention

The following guidelines promote compliance to the protocol and retention

- Participant-staff relationship. A key element contributing to participants’ continued commitment to the study involves fostering positive, respectful relationships between study participants and individual members of the staff.
- Continuity of care. In general, participants appointments should be scheduled so that they can be seen by the same clinic staff members during each visit.
- Clinic environment. A clinic environment that is warm and pleasant, and oriented to the comfort of the participant.
- Participant-staff communications. Good and consistent communication is essential. Instructions are clear and interactions are friendly and individualized. The participant is reminded of the benefits of study participation. Written reminders about appointments further enhance communication efforts.
- Convenience and accessibility. An easily accessible clinic location, availability of transportation, and convenient clinic hours all serve to facilitate study adherence. We will make study visits as easy as possible for participants, a critical factor to the success of the study. All sites take steps to ensure that clinic attendance is not compromised by a lack of transportation, unsuitable hours of clinic operation, or any similar circumstance. If necessary, participants are reimbursed for or are provided transportation to the clinic assessment visits.
- Time in clinic. Total clinic visit time is kept to a minimum, consistent with maintaining quality. If waiting is necessary, the situation is explained to the participant and, if possible, an offer is made for the participant to see another staff member, or to reschedule the visit. On the other hand, participants are not rushed or made to feel unwelcome. Study personnel trained to take time to visit with participants.
- Appointment reminders. Appointment reminders are used to prompt participants to come for clinic visits. These written reminders are mailed to participants so that they receive them before their scheduled visit date.
- Compensation: Participant will be compensated $100.00 for being in the study. The remuneration is given for attendance at clinic assessments, regardless of the level of participation in intervention activities

Statistical plan

Data analysis will be conducted at the Biostatistics and Data Management Core under the direction of Peihua Qiu, Ph.D. Our primary analysis will use an intention to treat approach in which participants are grouped according to randomization assignment without consideration of their adherence to the exercise or successful aging health education programs. We will also perform “per protocol” analyses to examine muscle adaptations in those individuals with adherence rates > 80%.

Although pilot studies are not powered for hypothesis testing, we will use a mixed effects model to assess changes in the outcomes with baseline measure serving as covariate. Associations between muscle mass and other outcomes will be explored using a correlational analysis

Data Safety and Monitoring Board

This project is funded as a Pilot Study through the UF Claude D. Pepper Older Americans Independence Center (OAIC) grant (NIH P30 AG028740) and thus uses the OAIC Data Safety Monitoring Board (DSMB).

The OAIC DSMB consists of an established board which has reviewed all studies conducted within the UF OAIC during for the past seven years. This board meets annually. DSMB Reports will be provided to the UF IRB-01 with annual Continuing Review submissions.

Confidentiality

Confidentiality of data is maintained by using research identification numbers or alphanumerics that uniquely identify each individual. Phone pre-screeners will be de-identified by replacing identifiable information with numbers or alphanumerics. Collected data will be used only for research purposes, and publications will not contain any individual identifiers Safeguards are established to ensure the security and privacy of participants' study records. The research records are kept in a locked room in locked computer files and file cabinets. Storage Areas for all records containing PHI or other restricted information is physically secure and environmentally controlled, to protect the records from unauthorized access and damage or loss from temperature fluctuations, fire, water damage, pests, and other hazards. Only study personnel have access to these files. After the study is completed, local data are stored with other completed research studies in a secured storage vault. According to the HIPAA rules, consent forms signed by the study participants, the study's research records containing identifiable data will be stored for a minimum of 6 (six) years after the study is completed and closed with IRB.

In compliance with the Health Insurance Portability and Accountability Act (HIPAA) and the Standards for Privacy of Individually Identifiable Health Information of the Department of Health and Human Services, study staff accesses personal health information and medical records only after receiving informed consent (verbal for phone screener and signed for short form and main consent).

4. POSSIBLE DISCOMFORTS AND RISKS

400-m walk: The 400-meter test may be associated with the risk of falling or development of chest discomfort due to coronary ischemia or dyspnea due to heart failure or lung disease. Rarely, falling during the 400-meter walk test may result in a fracture. Similar to the six-minute walk test, completion of the SPPB may be associated with the risk of falling or development of chest discomfort due to coronary ischemia or dyspnea due to heart failure or lung disease. Rarely, falling during the SPPB test may result in a fracture. In our experience conducting thousands of 400 m walk tests on individuals within various studies, the risk of falling is less than 1/500 and the risk of a fracture-associated fall is less than 1/5000. In the LIFE study we have performed approximately 10,000 400 m walk tests with no safety concerns raised by the DSMB.

Questionnaires: There are some risks associated with questionnaire administration. Sensitive information collected for this study includes lifestyle factors (e.g., alcohol use) and personal health information (PHI) and are used predominantly in screening for eligibility and safety. Participation includes a risk of loss of confidentiality of this information.

To minimize risk, questionnaire data are collected in secure spaces where the interview cannot be overheard. Participants are reminded that they can decline to answer questions that are uncomfortable or concerning. All research staff members complete annual HIPPA for Researchers training as required by UF.

DXA: DXA scans to determine body composition involve small amounts of radiation exposure. The amount of radiation exposure from each whole body DXA scan is <2.5 mRem per scan. Although the potential long-term risk from these radiation doses is uncertain, such doses are exceedingly low and have never been associated with any definite adverse effects.

Resistance Training: Potential risks associated with RT will be explained to each participant by appropriately trained clinical site staff during the initial informed consent, screening, and study orientation.

- Difficulty breathing and/or shortness of breath
- Muscle, joint strains and soreness
- Soft tissue injury, falls and fractures
- Increasing intraocular and systemic pressures associated with use of the Valsalva maneuver to levels that may cause injury (detached retina, hernia, conjunctival hemorrhage), which is greatly reduced by instruction in proper exercise technique.
- Hernia
- Exacerbation of arthritis or other joint conditions
- Lumbar disk hernia
- A small increase in the risk of sudden death, stroke, and acute MI occurring during a bout of
- vigorous exercise has been reported, especially in previously sedentary individuals.
- Orthostatic hypotension
- Triggering of an asthma episode

These risks are minimized in all exercise sessions by having supervised and trained exercise interventionists who instruct participants in the proper exercise techniques, including instructions to breathe normally during the exercise. Procedures to minimize injury include warm-up and cool-down activities that include large muscle movements and stretching. The preparatory phase is designed so that participants can learn proper exercise technique and gradually increase training volume and intensity.

The entire exercise intervention is conducted on-site and all sessions are supervised by trained interventionists, who will monitor potential adverse experiences and symptoms. During the exercise sessions, a defibrillator and on-site trained staff are available to deal with medical emergencies.

To minimize these risks, research staff are trained in the conduct of all physical performance tests and certified by before they work with study participants. Study staff are instructed <u>not</u> to perform these tests if they feel that testing is unsafe for an individual participant or if the participant is concerned about safety. If safety concerns are identified by either the study staff or the participant during the testing procedures, testing is halted and the participant is not allowed to complete the test. In either case, the participant is assessed to determine the need for medical intervention and the cause for concern is evaluated. All study staff are trained in activating the emergency response system at The University of Florida's Institute on Aging facility.

All sessions are conducted by research staff trained to monitor potential adverse experiences and symptoms during the testing sessions. All personnel associated with the study have completed Basic Life Support (BLS) training and education on management of acute events including syncope, chest pain, acute dyspnea, focal neurological symptoms and abnormal vital signs. Additionally, a study physician is available on call and contact numbers for emergency services are posted. Institutional and community EMS services will be activated if needed.

Participants will be instructed to talk with the investigators about any discomforts that occur during the study. If a participant reports an injury, chest pain, leg swelling, excessive shortness of breath, palpitations, or dizziness, he/she will be referred to medical attention (his/her doctor, or Emergency Department).

All research staff complete human participants training required by The University of Florida institutional review board (IRB) and National Institutes of Health (NIH).

Adverse events (AE) and serious adverse events (SAE)

The study will collects and reports only Serious Adverse Events (SAEs) and (nonserious) AEs associated with the intervention or study procedures. Pre-existing conditions and preplanned procedures (surgeries or therapies) scheduled prior to signing the Informed Consent are not considered AEs/SAEs.

Adverse events (AE): For this study's purposes, an AE or experience is defined as any health-related unfavorable or unintended medical occurrence that happens after randomization. Examples of AEs include, but are not limited to, the following:

- A clinically significant laboratory or clinical test result at follow up assessments
- An event that requires a visit to a physician because it alters the participant's ability to exercise
- An event that occurs as a result of a study procedure which is not listed in the Risks section of the consent

Serious adverse events (SAE): SAEs are defined as events that may be harmful to the participant and/or may be serious enough to warrant either temporary or permanent discontinuation of the study intervention or study procedures,
either because they are intolerable or because they are judged to be potentially harmful. Consistent with NIH guidelines, SAEs are AEs that meet any of the following criteria:

- results in death
- is life-threatening
- requires inpatient hospitalization
- prolongs an existing hospitalization
- results in a persistent or significant disability/incapacity
- important medical events that investigators judge to represent significant hazards or harm to
    - participants, and may require medical or surgical intervention to prevent one of the other SAEs
    - listed in this definition (e.g., hospitalization, death, persistent disability)

Relationship to study intervention: The classification of potential relationship to the intervention is as follows.

- Not Related – AE is clearly not related to the intervention or procedure (e.g., temporal sequence of events is not consistent with administration of intervention or procedures, other causes are more plausible or causal relationship is implausible.
- Possibly Related – AE that follows reasonable sequence from administration of intervention or procedure but that could readily have been produced by several other factors.
- Definitely Related – AE is clearly related to the intervention or procedure (e.g., follows reasonable temporal sequence from administration of the study intervention or procedure, confirmed by improvement after stopping intervention or procedure, and reappearance on repeated exposure to intervention or procedure, and cannot be explained by participants clinical or health status).

  Expected AE's: Expected AEs are defined as anticipated events based on the prior experience with the intervention and listed in the participant consent and protocol; these can be attributed to an underlying condition or the patient population being studied. These conditions that may be unpleasant and bothersome to the participant, such as sore muscles, muscle or joint pain, or breathing problems, do not require discontinuing the study intervention or components of the intervention.

  Unexpected Adverse Events: Unexpected adverse events (UAEs) are defined as events that occur during participation in the study but do not commonly occur in the study population and are

not listed in the informed consent. An AE is not defined as a UAE unless it meets all three defining criteria:

- unexpected given the research procedures described in the informed consent and characteristics of the study population;
- definitely related or possibly related to participation in the study intervention or procedures;
- suggests that the research intervention or procedures place participants or others at greater risk of harm than was previously realized.

5. POSSIBLE BENEFITS

- Participant will receive health and medical screening examinations at baseline.
- Participant will have the opportunity to participate in a strength training program or health education program with professional supervision. Participation in these activities has the potential to improve participant's health, body composition, and quality of life.
  - Also, all participants will receive an **individualized report** of your strength, muscle mass, fat percentage, bone density and physical function in comparison with other older adults in your age group and gender at the end of the intervention.
  - The results of the study will help validate a new tool to measure muscle mass that will have a number of potential uses, ranging from detecting muscle loss in both health and disease (cachexia) to evaluating the efficacy of behavioral and pharmacological interventions to increase muscle mass.

6. CONFLICT OF INTEREST

None

1. Visser M, Goodpaster BH, Kritchevsky SB, et al. Muscle mass, muscle strength, and muscle fat infiltration as predictors of incident mobility limitations in well-functioning older persons. *J Gerontol A Biol Sci Med Sci*. 2005;60:324-333.

2. Schaap LA, Koster A, Visser M. Adiposity, muscle mass, and muscle strength in relation to functional decline in older persons. *Epidemiol Rev*. 2013;35:51-65.

3. Proctor DN, O'Brien PC, Atkinson EJ, Nair KS. Comparison of techniques to estimate total body skeletal muscle mass in people of different age groups. *Am J Physiol*. 1999;277:E489-95.

4. Overend T, Cunningham D, Paterson D, Lefcoe M. Thigh composition in young and elderly men determined by computed tomography. *Clinical Physiology and Functional Imaging*. 1992;12:629-640.

5. Rice C, Cunningham D, Paterson D, Lefcoe M. Arm and leg composition determined by computed tomography in young and elderly men. *Clinical Physiology and Functional Imaging*. 1989;9:207-220.

6. Frontera WR, Reid KF, Phillips EM, et al. Muscle fiber size and function in elderly humans: a longitudinal study. *J Appl Physiol (1985)*. 2008;105:637-642.

7. Hughes VA, Frontera WR, Wood M, et al. Longitudinal muscle strength changes in older adults: influence of muscle mass, physical activity, and health. *J Gerontol A Biol Sci Med Sci*. 2001;56:B209-17.

8. Delmonico MJ, Harris TB, Visser M, et al. Longitudinal study of muscle strength, quality, and adipose tissue infiltration. *Am J Clin Nutr*. 2009;90:1579-1585.

9. Rosenberg IH. Sarcopenia: Origins and Clinical Relevance. *Clin Geriatr Med*. 2011;27:337-339.

10. Cao L, Morley JE. Sarcopenia Is Recognized as an Independent Condition by an International Classification of Disease, Tenth Revision, Clinical Modification (ICD-10-CM) Code. *J Am Med Dir Assoc*. 2016;17:675-677.

11. Morley JE, Abbatecola AM, Argiles JM, et al. Sarcopenia with limited mobility: an international consensus. *J Am Med Dir Assoc*. 2011;12:403-409.

12. Studenski SA, Peters KW, Alley DE, et al. The FNIH Sarcopenia Project: Rationale, Study Description, Conference Recommendations, and Final Estimates. *The Journals of Gerontology Series A: Biological Sciences and Medical Sciences*. 2014.

13. Cruz-Jentoft AJ, Baeyens JP, Bauer JM, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. *Age Ageing*. 2010;39:412-423.

14. Lee RC, Wang Z, Heymsfield SB. Skeletal muscle mass and aging: regional and whole-body measurement methods. *Canadian Journal of Applied Physiology*. 2001;26:102-122.

15. Boutin RD, Yao L, Canter RJ, Lenchik L. Sarcopenia: current concepts and imaging implications. *Am J Roentgenol*. 2015;205:W255-W266.

16. Clark RV, Walker AC, O'Connor-Semmes RL, et al. Total body skeletal muscle mass: estimation by creatine (methyl-d3) dilution in humans. *J Appl Physiol (1985)*. 2014;116:1605-1613.

17. Stimpson SA, Turner SM, Clifton LG, et al. Total-body creatine pool size and skeletal muscle mass determination by creatine-(methyl-D3) dilution in rats. *J Appl Physiol (1985)*. 2012;112:1940-1948.

18. Ferrucci L, Guralnik JM, Studenski S, Fried LP, Cutler GB, Walston JD. Designing randomized, controlled trials aimed at preventing or delaying functional decline and disability in frail, older persons: a consensus report. *J Am Geriatr Soc*. 2004;52:625-634.

19. Physical Activity Guidelines Advisory Committee. Physical activity guidelines advisory committee report, 2008. *Washington, DC: US Department of Health and Human Services*. 2008;2008:A1-H14.

20. Nelson ME, Rejeski WJ, Blair SN, et al. Physical activity and public health in older adults: recommendation from the American College of Sports Medicine and the American Heart Association. *Med Sci Sports Exerc*. 2007;39:1435-1445.

21. Sweet TW, Foster C, McGuigan MR, Brice G. Quantitation of resistance training using the session rating of perceived exertion method. *J Strength Cond Res*. 2004;18:796-802.

22. Fielding RA, Rejeski WJ, Blair S, et al. The Lifestyle Interventions and Independence for Elders Study: design and methods. *The journals of gerontology.Series A, Biological sciences and medical sciences*. 2011;66:1226.

23. Fries JF, Witter J, Rose M, Cella D, Khanna D, Morgan-DeWitt E. Item response theory, computerized adaptive testing, and PROMIS: assessment of physical function. *J Rheumatol*. 2014;41:153-158.

24. McHorney CA, Ware Jr JE, Raczek AE. The MOS 36-Item Short-Form Health Survey (SF-36): II. Psychometric and clinical tests of validity in measuring physical and mental health constructs. *Med Care*. 1993:247-263.

25. Leeuw M, Goossens ME, van Breukelen GJ, Boersma K, Vlaeyen JW. Measuring perceived harmfulness of physical activities in patients with chronic low back pain: the Photograph Series of Daily Activities--short electronic version. *J Pain*. 2007;8:840-849.

26. Baer RA, Smith GT, Hopkins J, Krietemeyer J, Toney L. Using self-report assessment methods to explore facets of mindfulness. *Assessment*. 2006;13:27-45.

27. Goubert L, Crombez G, Van Damme S, Vlaeyen JW, Bijttebier P, Roelofs J. Confirmatory factor analysis of the Tampa Scale for Kinesiophobia: invariant two-factor model across low back pain patients and fibromyalgia patients. *Clin J Pain*. 2004;20:103-110.

28. Borg G. *Borg's Perceived Exertion and Pain Scales.* Human kinetics; 1998.

29. Newman AB, Simonsick EM, Naydeck BL, et al. Association of long-distance corridor walk performance with mortality, cardiovascular disease, mobility limitation, and disability. *JAMA*. 2006;295:2018-2026.

30. Espeland MA, Gill TM, Guralnik J, et al. Designing clinical trials of interventions for mobility disability: results from the lifestyle interventions and independence for elders pilot (LIFE-P) trial. *The Journals of Gerontology Series A: Biological Sciences and Medical Sciences*. 2007;62:1237-1243.

31. Guralnik JM, Ferrucci L, Pieper CF, et al. Lower extremity function and subsequent disability: consistency across studies, predictive models, and value of gait speed alone compared with the short physical performance battery. *J Gerontol A Biol Sci Med Sci*. 2000;55:M221-31.

32. Guralnik JM, Simonsick EM, Ferrucci L, et al. A short physical performance battery assessing lower extremity function: association with self-reported disability and prediction of mortality and nursing home admission. *J Gerontol*. 1994;49:M85-94.

33. Guralnik JM, Ferrucci L, Pieper CF, et al. Lower extremity function and subsequent disability: consistency across studies, predictive models, and value of gait speed alone compared with the short physical performance battery. *J Gerontol A Biol Sci Med Sci*. 2000;55:M221-31.

34. Guralnik JM, Simonsick EM, Ferrucci L, et al. A short physical performance battery assessing lower extremity function: association with self-reported disability and prediction of mortality and nursing home admission. *J Gerontol*. 1994;49:M85-94.

35. Rantanen T, Harris T, Leveille SG, et al. Muscle strength and body mass index as long-term predictors of mortality in initially healthy men. *The Journals of Gerontology Series A: Biological Sciences and Medical Sciences*. 2000;55:M168-M173.

36. Drouin JM, Valovich-mcLeod TC, Shultz SJ, Gansneder BM, Perrin DH. Reliability and validity of the Biodex system 3 pro isokinetic dynamometer velocity, torque and position measurements. *Eur J Appl Physiol*. 2004;91:22-29.

37. Kenny R, Rubenstein LZ, Tinetti ME, et al. Summary of the updated American Geriatrics Society/British Geriatrics Society clinical practice guideline for prevention of falls in older persons. *J Am Geriatr Soc*. 2011;59:148-157.

38. Lusardi MM, Fritz S, Middleton A, et al. Determining Risk of Falls in Community Dwelling Older Adults: A Systematic Review and Meta-analysis Using Posttest Probability. *J Geriatr Phys Ther*. 2017;40:1-36.