Title: Time to Eat Study – Pilot

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1. Abstract:

Growing evidence shows that low-grade chronic inflammation, characterized by elevations in plasma C-reactive protein and particularly Interleukin-6 (IL-6), is an independent risk factor of disability, impaired mobility, and lower walking speed. Low-grade chronic inflammation is a modifiable risk factor. However, it is unknown whether interventions that reduce the levels of inflammatory markers *per se* improve mobility, or avert decline in mobility in older persons.

There also remains an important need for interventions that can improve unhealthy changes in body composition that occur during aging. Given the known loss of lean mass that occurs during both aging and continuous calorie restriction, alternative approaches are needed to help older adults lose unhealthy weight while retaining larger amounts of lean mass. Our recent review suggests that a specific form of intermittent fasting, time restricted feeding, results in weight and fat loss (even when caloric intake is not limited), without significant loss of lean mass. This would be an optimal outcome for overweight, older adults and should also result in a reduction in inflammation and improvement in mobility.

To date, no study has examined the effects of time restricted feeding in older adults. Preliminary data regarding feasibility need to be gathered before a full scale trial can be effectively designed and implemented. Thus, the primary aims of this pilot study are to evaluate the safety and feasibility of time restricted feeding in an older adult population. The Time to Eat Pilot Study will also assess the variance of inflammatory markers, walking speed, physical and cognitive function, grip strength, body measurements, perceived fatigability, and activity level. This allows the refinement of the design, recruitment yields, target population, adherence, retention, and tolerability of a larger scale study.

For this 4-week pilot study, we will recruit 10 overweight, older adults who are at risk for, or with, mobility impairment, as measured by slow gait speed and self-reported mobility difficulty and self-reported sedentary lifestyle to complete. All participants will receive the time restricted feeding intervention, which will consist of fasting for approximately 16 hours per day.

2. Specific Aims:

Growing evidence from our group and others shows that low-grade chronic inflammation, characterized by elevations in plasma C-reactive protein (CRP) and particularly interleukin-6 (IL-6) [1-7] is an independent risk factor for disability, impaired mobility, and slow walking speed [1, 6-11]. Low-grade chronic inflammation is a modifiable risk factor. However, it is unknown whether interventions that reduce the levels of inflammatory markers *per se* improve mobility, or avert decline in mobility in overweight, older persons

Conduct a pilot study of 10 overweight, older sedentary persons at risk for mobility decline to assess:

(i) Feasibility of the study intervention in the study population
   a. adherence and retention
   b. tolerability
   c. recruitment yields

(ii) Variance in the following:
   a. inflammation (IL-6 and CRP) level
   b. walking speed
   c. body mass index
   d. waist circumference
   e. grip strength
   f. fatigability level
   g. activity level
   h. cognitive functioning as measured by the MoCA
   i. perceived exertion during the 6 min walk
   j. health-related quality of life.

3. Background:

Aging is associated with a host of biological changes that contribute to a progressive decline in cognitive and physical function, ultimately leading to a loss of independence, and increased risk of mortality. To date, caloric restriction (i.e., a reduction in caloric intake without malnutrition) is the only non-genetic intervention that has consistently been found to extend both mean and maximal life span across a variety of species [12]. Although attenuated, these effects remain present even when moderate caloric restriction (20-40%) is implemented in middle-age mice [13]. Importantly, prolonged caloric restriction has also been found to delay the onset of age-associated disease conditions such as cancer and diabetes in rodents [14] and in nonhuman primates [15]. Thus, findings from animal studies, including recent primate studies, suggest prolonged caloric restriction has the potential to extend health-span and thereby increase quality of life.

In recent studies conducted in overweight humans, caloric restriction has been shown to improve a number of health outcomes including reducing several cardiac risk

factors [16-18], improving insulin-sensitivity [19], and enhancing mitochondrial function [20]. Additionally, prolonged caloric restriction has also been found to reduce oxidative damage to both DNA [21-23] and RNA, as assessed through white blood cells [23]. Thus, findings of initial human clinical trials appear to support the promise of caloric restriction demonstrated in animal studies, at least in overweight adults.

Despite these health promoting biological changes, most individuals have difficulty engaging in caloric restriction over the long-term [24]. Findings from weight loss interventions conducted over the past few decades indicate that the vast majority of individuals who lose weight regain this lost weight over the course of one to five years. An additional concern about implementing calorie restriction in older adults is that the weight loss is comprised of both fat and lean tissue; however, the weight that is frequently regained is primarily comprised of fat. This can lead to a situation where the individuals body composition is worse than when they initiated a weight loss program.

Due to poor long-term compliance and loss of lean tissue that occurs with calorie restriction, there is a need to explore the potential of alternative approaches for reducing body weight, specifically body fat, in overweight older adults. One alternative dietary approach that may produce similar biological changes as caloric restriction that has received increasing interest from the scientific community is Intermittent Fasting. In contrast to traditional caloric restriction paradigms, food is not consumed during designated fasting time periods but is typically not restricted during designated feeding time periods. The length of the fasting time period can also vary but is frequently 12 or more continuous hours.

Evidence that this approach may have beneficial effects on longevity first appeared several decade ago [25]. Since this time, a growing body of literature suggests that fasting periods and intermittent fasting regimens can trigger similar biological pathways as caloric restriction (i.e., increased autophagy and mitochondrial respiratory efficiency), which can result in host of beneficial biological effects including increased circulation and cardiovascular disease protection, modulation of reactive oxygen species and inflammatory cytokines, as well as antimutagenic, antibacterial, and anticarcinogenic effects [26].

We recently completed a review of the literature on the effects of intermittent fasting on changes in body composition in overweight individuals [27]. For the time restricted feeding approach, we identified four clinical trials that measured changes in participant's body composition before and after they received this intervention. An important finding of our review was that participants who engaged in time restricted feeding lost a significant amount of body fat without loss of lean tissue in three of the four clinical trials included in our review. These findings suggest that time restricted feeding results in weight and fat loss (even when caloric intake is not limited) without significant loss of lean mass.

Despite these promising findings, no study to date has examined the effects of time restricted feeding in overweight, older adults. Thus, there is a need to understand

whether a time restricted eating pattern could produce similar beneficial changes on the body composition of older adults, as well as reduce inflammation and improve mobility. Given the known loss of lean mass that occurs during both aging and continuous calorie restriction, time restricted feeding regimens may be an effective approach to help overweight, older adults lose unhealthy weight while retaining larger amounts of lean mass.

To date, no study has examined the effects of time restricted feeding in older adults. Preliminary data regarding feasibility need to be gathered before a full scale trial can be effectively designed and implemented. Thus, the primary aims of this pilot study are to evaluate the safety and feasibility of time restricted feeding in an overweight, older adult population. The Time to Eat Pilot Study will also assess the variance of inflammatory markers, walking speed, physical and cognitive function, grip strength, body measurements, perceived fatigability, and activity level. This allows the refinement of the design, recruitment yields, target population, adherence, retention, and tolerability of a larger scale study.

4. Research Plan:

4.1 Design

This is a pilot study of 10 overweight, older sedentary persons at risk for mobility decline to assess feasibility of an intermittent fasting intervention program within this study population and evaluate changes in inflammatory level, walk speed, physical and cognitive function, grip strength, body measurements, perceived fatigability, and activity levels.

4.2 Eligibility Criteria

Inclusion criteria

- Consent to participate in the study
- Men and women ≥ 65 years old
- Self-reported difficulty walking ¼ mile or climbing a flight of stairs
- Self-reported sedentariness (<30 minutes structured exercise per week)
- Walking speed <1 m/sec on the 4 m walk test
- Able to walk unassisted (cane allowed)
- Have a body mass index between 25 – 40 kg/m^2 (inclusive)

Exclusion criteria

- Current dietary habits
    - Fasting >12 hours per day
    - Actively trying to lose weight by participating in formal weight loss program or significantly restricting calorie intake
    - Weight loss > 5 lbs in the past month
- Medical history or conditions

  - Resting heart rate of >120 beats per minute, systolic blood pressure > 180 mmHg or diastolic blood pressure of > 100 mmHg
  - Unstable angina, heart attack or stroke in the past 3 months
  - Continuous use of supplemental oxygen to manage a chronic pulmonary condition or heart failure
  - Rheumatoid arthritis, Parkinson's disease or currently on dialysis
  - Active treatment for cancer in the past year
  - Insulin dependent diabetes mellitus
- Taking medications that preclude fasting for 16 hours (e.g. must be taken with food at least 12 hours apart)
- Any condition that in the opinion of the investigator would impair ability to participate in the trial

4.3 Screening, assessments, and follow-up

Table 1. Schedule of Assessments

	Phone screen (phone)	Screen/ Baseline (clinic)	Week 1 (phone)	Week 2 (phone)	Week 3 (phone)	Week 4 (clinic)
Verbal consent & phone screening	X					
Consent		X				
Review of medical history*		X				
4 m walk*		X				
Height*, weight*, waist circumference		X				X
Blood pressure*, pulse*, temperature		X				X
6 minute walk		X				X
Grip strength		X				X
Fasting blood collection		X				X
Montreal Cognitive Assessment (MoCA)		X				X
Fatigability Questionnaire		X				X
SF-12		X				X
Distribute / collect accelerometer		X				X
Distribute food intake time and sleep diaries		X				
Review food intake time diary			X	X	X	X
AE review			X	X	X	X
Satisfaction survey & exit interview						X

* Must be completed to confirm eligibility prior to completing other baseline measurements.

Recruitment and Telephone Screening

Potential participants will be recruited from the general population in the North/Central Florida area through general mailings and advertisements and targeted outreach to those who have consented to participate in the Claude D. Pepper Recruitment Registry (IRB#417-2007).

Potential participants who verbally consent to complete the telephone screening are assessed of the main eligibility criteria, including:

- Age
- Self-reported sedentariness
- Self-reported mobility difficulty
- Self-reported height and weight
- Medical history
- Current dietary pattern

Screening/Baseline Clinic Visit

Those who qualify based on the telephone screening are invited to take part in the in person clinic visit. During this visit, informed consent is obtained and the following assessments are completed:

- Measurement of height and weight to determine body mass index
- 4 m walk test

If the participant is eligible to participate based on these assessments, he/she will then continue with the remainder of the baseline assessments, including:

- Measurement of waist circumference
- 6 minute walk test
- Grip strength test
- Collection of blood sample for the measurement of inflammation biomarkers (IL-6 and CRP)
- Montreal Cognitive Assessment
- Fatigability Questionnaire
- SF-12
- Distribute food intake and sleep time record diaries
- Distribute activity monitor

Weekly Phone Contacts

Participants will be contacted by phone at the end of weeks 1, 2, and 3. The following will be reviewed:

- Time of first and last food or drink consumption over the past week
- Changes in health (adverse events)

Study staff will provide participants counseling if he/she is experiencing difficulty in adhering to the study intervention.

Additional phone calls may be made as needed, if the participant requires additional support or guidance in adhering to the study intervention.

Week 4 Follow-up Clinic Visit

Following 4 weeks of the study intervention, participants will complete a follow-up clinic visit. At this visit, baseline assessments are repeated and participants will be asked to report any changes in health since initiating the study. Additionally, participants will be asked to complete a satisfaction questionnaire and an exit interview about their experience during the study.

4.4 Intervention

Intermittent Fasting

Study participants will be asked to fast for a target of 16 hours per day for a period of 4 weeks. The first week will involve of a ramp up to a full 16-hour fasting period (Days 1-3 fast for 12-14 hours per day, Days 4-6 fast for 14-16 hours per day, Days 7-28 fast for 16 hours per day). Participants will be allowed to consume calorie-free beverages, tea, black coffee, sugar-free gum, and they will be encouraged to drink plenty of water throughout the entire intervention period. Participants will be asked to record the time of first and final food/drink consumption each day.

4.5 Outcomes

Adherence and Retention

Adherence to the study intervention is measured using food intake time diary. Participants are considered compliant to the study intervention if he/she fasts between 14 – 18 hours per day during weeks 2 - 4. Retention is measured by drop-out rate.

Tolerability

At each follow-up contact (either by phone or in person), participants are asked about any changes to their health or physical function since the previous contact. The changes are documented on the adverse event log. Participants are also asked about problems or challenges in following the study intervention. Any reported difficulties will be recorded on a progress note.

6 Minute Walk Test

The 6 Minute Walk test measures the amount of distance the participant can complete on a standard walking course in six minutes without running or overexerting themselves. Contraindications for test administration and stopping rules based on American Thoracic Society will be followed [28]. The walk test is completed at baseline and Week 4.

Grip Strength

Isometric hand grip strength is a commonly used measure of upper body skeletal muscle function and has been widely used as a general indicator of functional status [29, 30] and is completed at baseline and Week 4.

Montreal Cognitive Assessment (MoCA)

The Montreal Cognitive Assessment is a 30-point assessment of mild cognitive impairment, which assesses the domains of attention and concentration, executive functions, memory, language, visuoconstructional skills, conceptual thinking, calculations, and orientation [31]. This test is completed at baseline and Week 4. Different versions of the test are given at the two time points to avoid learning effects between repeated administrations of the test.

Fatigability Questionnaire

A self-administered, 26-item questionnaire to measure perceived mental and physical fatigability [32] is completed at baseline and Week 4.

Short Form (SF) -12 Health Survey

Self-administered, 12-item questionnaire to measure health-related quality of life is completed at baseline and Week 4.

Anthropometric Measurements

Body weight will be measured following the removal of excess clothing and shoes with calibrated scales.

Waist circumference is taken at the mid-point between the participant's lowest rib and the top of his/her hip bone. Both body weight and waist circumference measurements are completed at screening/baseline and Week 4.

Activity Monitor

A wrist activity monitor accelerometer to measure daily activity level is worn for the duration of the 4-week study intervention.

Systemic Inflammatory biomarkers. Inflammatory biomarkers (CRP and IL-6) are measured by ELISA at Screening/Baseline and the Week 4 visit.

Satisfaction Survey and Exit Interview

At the conclusion of the study, participants are asked to complete a self-administered survey of their level of satisfaction and experience with his/her study participation.

Participants will also complete an exit interview with study staff to allow sharing of additional feedback regarding study participation and the design of the full scale study protocol.

4.6 Statistical Analysis

Sample size estimates for pilot studies are inherently difficult because by intent, a pilot study is meant to include a smaller sample than a fully powered study. The enrollment sample of 10 participants is intended to provide sufficient data to indicate feasibility of the intermittent fasting (i.e., time restricted feeding) intervention in this study population and to provide descriptive estimates of effects.

Feasibility and acceptability of the proposed intervention in overweight, older adults will be evaluated by process variables including number of participants completing the study; adherence to study intervention, and number and severity of adverse events. We will also evaluate the participants' responses on the satisfaction survey as part of the feasibility and acceptability assessment.

4.7 Compensation

For time and travel, participants will receive compensation of $25 for the Screening/Baseline clinic visit and $25 for the Week 4 follow-up clinic visit. The maximum compensation for completing the study is $50.

4.8 Data Safety Monitoring Plan and Data Safety Monitoring Board

Data Safety Monitoring Plan

Safety of the study participants is always our major concern. The study coordinator will record and monitor all adverse events reported at in person visits, and the study interventionist will record and monitor all adverse events reported by telephone. The Study Physician will determine the seriousness of any reported adverse events and whether or not the event is related to the study intervention.

An **adverse event** is any untoward medical occurrence associated with the use of a drug in humans, whether or not considered study related.

The event is **serious** if it results in death, is life-threatening, requires inpatient hospitalization or prolongs an existing hospitalization, results in a persistent or significant disability/incapacity, or congenital anomaly/birth defect.

Safety related events are reported in a timely fashion, as required by the DSMB and the IRBs that are responsible for study oversight.

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

Definite - Temporal pattern + Known or expected AE response pattern + Confirmed by stopping the intervention + Reappearance of AE on re-challenge

Possible - Temporal pattern + Known or expected AE response pattern + Could have been produced by a number of other factors

Not related - AE for which sufficient information exists to indicate that the cause is

unrelated to the study intervention

Time to Eat study participants are comprised of an overweight, moderately functioning population expected to be at high risk for acute and chronic comorbid health events.

A Data and Safety Monitoring Plan is implemented to ensure the safety of all participants involved in the study and to ensure the validity and integrity of the data. The Principal Investigator with the advice of the Study Physician monitor all aspects of safety.

Data Safety Monitoring Board

This project supported by the UF Claude D. Pepper Older Americans Independence Center (OAIC) grant (NIH P30 AG028740) and will utilize the OAIC Data Safety Monitoring Board (DSMB).

The OAIC DSMB consists of an established board which has reviews all studies conducted within the UF OAIC. This board meets bi-annually and consists of the following individuals: (1) Stephen Kritchevsky, Ph.D., Chair, an epidemiologist who has been involved in research for many years, (2) Jing Cheng, Ph.D., a biostatistician who has been involved in numerous clinical trials, and (3) John Meuleman, M.D., a physician who has been involved in the conduct of clinical research for many years. DSMB Reports will be provided to the UF IRB-01 with annual Continuing Review submissions.

5. Possible Discomforts and Risks:

Intermittent Fasting

Only a limited number of human clinical trials have been published to date evaluating intermittent fasting dietary interventions [33, 34]. Of the adverse effects reported in these and other studies, undesirable weight loss and hunger were the two most prominent. It is likely that it will take participants several days to start adjusting to a time restricted eating pattern. During these times, participants may experience gastrointestinal discomfort, feelings of being more or less hungry than usual, and have more or less energy at certain times of the day than they usually have. While adhering to time restricted feeding, participants may feel their blood sugar levels fluctuate up and down more rapidly than they are typically used to, which may affect their energy levels and how hungry or full they feel. Also, there is a small risk that their blood sugar levels will go too high or too low. If their blood sugar levels drop too low, they may experience dizziness, sweating, sleepiness, anxiety, hunger, or weakness. Participants will be encouraged to contact the study physician and/or PI if they become concerned while adhering to this eating pattern.

Risks associated with the blood draw. The risks of drawing blood from a vein include discomfort at the site of puncture; possible bruising and swelling around the puncture site; rarely an infection; and, uncommonly, faintness from the procedure. To minimize these risks, study staff are trained to properly draw blood and use measures that help prevent complications.

Risks associated with blood pressure measurement. The risks of placing a blood pressure cuff on a participant's arms include pinching or slight bruising. To minimize these risks, study staff are trained to appropriately administer blood pressure tests.

Risks associated with physical performance tests. There is a risk of losing balance and falling associated with the physical performance-based tested (e.g., 4 meter walk and 6 minute walk tests). Falling includes the risk for bone fracture and soreness or injury to muscles or tendon/ligaments. To lessen these risks, participants will be safely escorted to chairs located along the walking course should he/she become unsteady. A Study Coordinator will follow at a close distance during the walking tests and will continuously monitor the participant for safety.

The 6 minute walk test will be stopped immediately if any of the following signs and symptoms are reported or observed: chest pain, intolerable dyspnea, leg cramps, staggering, diaphoresis, and pale or ashen appearance.

Risks associated with loss of confidentiality
Lack of maintenance of confidentiality is a potential risk. Data are used only in aggregate and no identifying characteristics of individuals will be published or presented.

Confidentiality of data is maintained by using research identification numbers that uniquely identify each individual. Safeguards are established to ensure the security and privacy of participants' study records. Appropriate measures are taken to prevent unauthorized use of study information. The research ID number is used. The research records are kept in a limited access, secured storage area. Only trained and certified study personnel has access to these files.

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, we access personal health information and medical records only after receiving signed informed consent.

6. Possible Benefits:

All study participants will take part in the intermittent fasting study intervention (time restricted feeding), which may or may not result in various health benefits.

7. Conflict of Interest:

None

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