1.0 Objectives

Obesity significantly contributes to the risk of developing postmenopausal breast cancer, particularly estrogen receptor-positive (ER+) breast cancer.[3] For every 5 kg/m^2 increase in body mass index (BMI), the risk of postmenopausal breast cancer increases by 12%, with the risk increased by 17% for ER+ breast cancer.[4] However, BMI is an imprecise indicator of body composition because it does not differentiate between fat mass and lean mass.[5] New research conducted by our group shows that postmenopausal women with BMI in the normal range (18.5- 24.9 kg/m^2) who have high adiposity have approximately a doubling in the risk of developing ER+ breast cancer, compared to those in the lowest quartile of body fatness.[1] This increased risk may be explained by both local changes in the breast, e.g., elevated levels of aromatase, the rate-limiting enzyme for estrogen biosynthesis, and systemic abnormalities, i.e., changes in circulating levels of metabo-inflammatory factors.[6]

Our long-term research goal is to determine if diet and exercise is an effective breast cancer prevention strategy in this population of women who are indeed at higher risk of breast cancer and often missed as candidates for lifestyle interventions and breast cancer prevention programs. Women of normal BMI are typically viewed as having a low risk of breast cancer and other health problems and generally are not advised by their healthcare provider to make lifestyle changes. Our data call this assumption into question. Here we propose a study to gather preliminary data testing an intervention to improve body composition (i.e., reduce adiposity while maintaining muscle mass) in postmenopausal normal BMI women with excess adiposity. If the feasibility and preliminary efficacy of the intervention are supported by the data that are generated, we will apply for NIH funds to support a larger trial. This research has strong potential to be practice changing as it will inform diet and exercise recommendations for a population of women who are at higher risk of developing breast cancer but who currently do not routinely receive guidance on primary prevention of breast cancer.

Goal: To conduct a randomized pilot study to evaluate the feasibility and preliminary efficacy of a lifestyle intervention on body composition and metabo-inflammation in postmenopausal women who are of normal weight (BMI 18.5 – 24.9 kg/m^2) but have an elevated risk of breast cancer because of excess adiposity. Our specific aims are as follows:

1. Primary: Evaluate the feasibility of a diet and exercise intervention to decrease body fat in postmenopausal women with normal BMI but high body fat (trunk fat mass ≥ 9.4 kg, the 50th percentile of normal BMI women in the Women's Health Initiative (WHI)). The study will be considered feasible if ≥40% of the women screened are eligible, 75% of the participants are adherent to the health coaching and personal training sessions, and ≥80% of participants complete the 4-month study assessment.

2. Secondary: Assess preliminary efficacy of the intervention by evaluating the post-intervention differences between the intervention and control groups in: (1) circulating markers of inflammation and metabolic dysfunction linked to both excess adiposity and breast cancer (high sensitivity C-reactive protein [hsCRP], fasting insulin, leptin, IL-6, triglycerides, sex hormone binding globulin [SHBG], adiponectin, and HDL cholesterol); (2) body composition (% body fat, trunk fat mass, fat mass, lean mass, fat-free mass).

3. Exploratory: Exploratory outcomes include fitness (6-minute walk test, sit-to-stand test), behavior (physical activity, energy intake, macronutrient consumption), and quality of life (global health-related quality of life, physical functioning, sleep, menopausal symptoms).

   Hypothesis: A lifestyle intervention that emphasizes strength and aerobic training and a high-quality diet with increased protein will reduce adiposity in the absence of muscle loss, lower circulating levels of markers of inflammation, and improve metabolic dysfunction in normal BMI women with excess body fat, as compared with the control group.

   The expected outcome of this project is the demonstration of feasibility of the lifestyle intervention that will be used for a larger trial to assess the impact of changes in body composition on biomarkers of breast cancer risk. The biomarker and body composition data produced by this R21 will be used to estimate effect sizes and variance of outcomes that will be used to plan a larger randomized controlled trial. The proposed project and future trial will advance the field in cancer prevention and control by providing a useful tool for breast cancer risk reduction in women with this risk factor, helping them make informed lifestyle choices to reduce their cancer risk.

2.0 Rationale

Obesity and breast cancer risk: Obesity, measured by BMI, is associated with an increased risk of postmenopausal, ER+ breast cancer when comparing highest to lowest BMI categories.[4] However, the accuracy of BMI alone to identify those at risk for obesity-related disorders has been called into question.[5, 7] BMI is an approximate measure of body size that does not discriminate between fat mass and lean mass. Some individuals considered healthy by virtue of having a normal BMI (18.5 – 24.9 kg/m^2) may in fact have high body fat, which increases the risk for cardiometabolic abnormalities such as hypertension, dyslipidemia, and impaired glucose tolerance.[8] These disorders, when found in individuals with a normal BMI, are termed metabolic obesity and are associated with increased risk of cardiovascular morbidity and mortality.[9]

A growing body of evidence indicates that normal weight women with excess body fat and metabolic obesity also harbor an increased risk of cancer. In a case-cohort study within the WHI, women with elevated fasting insulin levels had a higher risk of breast cancer regardless of whether they were normal weight or overweight.[10] Similarly, in the Sister Study, normal weight women with at least one metabolic abnormality had a nearly equivalent increase in postmenopausal breast cancer risk as overweight and obese women compared to normal-weight women with no metabolic abnormalities.[11] Recently, Dr. Dannenberg and colleagues conducted a secondary analysis of WHI data which included a cohort of over three thousand postmenopausal women with a normal BMI. With a median follow-up of 16.4 years, women in the highest quartiles of total body fat mass, trunk fat mass, and percent body fat as measured by Dual-energy X-ray Absorptiometry (DXA) demonstrated about a doubling in the risk of ER+ breast cancer compared to those in the lowest quartile, despite having a normal BMI.[12] Additionally, higher levels of circulating biomarkers of inflammation and metabolic dysfunction were associated with higher levels of body fat. Collectively, these data highlight the urgent need for clinical trials evaluating the role of fat loss interventions, which have traditionally targeted elevated BMI, for breast cancer risk reduction in normal BMI postmenopausal women with high levels of body fat.

Mechanisms: The preponderance of mechanistic data support the observation that postmenopausal women with normal BMI and excess body fat are at increased risk of breast cancer. Obesity-related effects on adipokines, hormones and inflammation have all been suggested to play a role in breast carcinogenesis.[6, 13-15] Our team was the first to describe breast white adipose tissue inflammation, a process that has subsequently been linked to the development and progression of breast cancer.[16-19] In women with excess body fat, breast adipocyte hypertrophy and death lead to the development of white adipose inflammation. White adipose inflammation is found in the breast tissue of most overweight and obese women[20] and is associated with increased levels of aromatase, the rate-limiting enzyme for estrogen biosynthesis.[20]

Notably, recent research by our team has identified breast white adipose tissue inflammation in nearly 40% of women with normal BMI.[21] In normal BMI women, the presence of breast white adipose tissue inflammation is associated with excess body fat, increased breast aromatase expression and systemic biomarkers of inflammation and altered metabolism (i.e., hsCRP, fasting insulin, glucose, leptin, IL-6, adiponectin, triglycerides, SHBG and HDL-cholesterol).[21] Thus, excess body fat and its associated local (i.e., breast adipose tissue inflammation) and systemic changes can explain why thousands of normal BMI postmenopausal women develop ER+ breast cancer. Although we can use measurements of body composition to identify this new high risk population, this is of limited value until an effective treatment plan is developed.

The scientific premise of this trial rests on data indicating that BMI is an overly simplistic measure of body composition that may provide false reassurance regarding breast cancer risk in postmenopausal women with normal BMI. While women who are overweight or obese are frequently advised to lose weight to reduce the risk of cancer and other health problems, normal BMI women are unlikely to receive such advice. Given that postmenopausal women with normal BMI but excess adiposity face increased health risks, research is needed to determine whether a lifestyle intervention to reduce adiposity can decrease inflammation, improve insulin resistance, and subsequently reduce the risk of breast cancer. We hypothesize that a lifestyle intervention that emphasizes strength and aerobic training and a high-quality diet with increased protein will reduce adiposity in the absence of muscle loss, lower circulating levels of markers of inflammation, and improve metabolic dysfunction in normal BMI postmenopausal women with excess body fat.

3.0 Design and Population

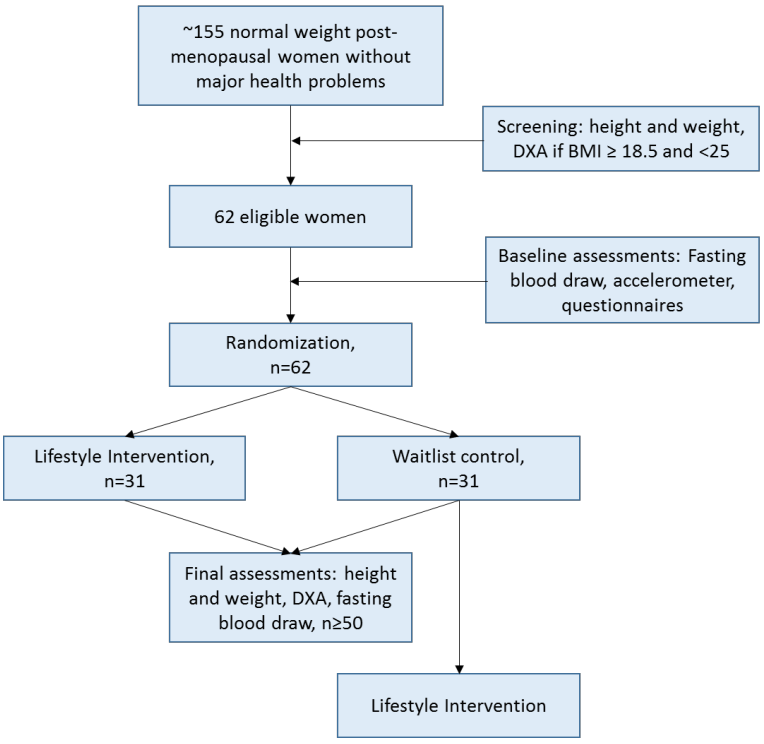
We will recruit normal BMI, postmenopausal women from employees of MD Anderson Cancer Center as well as the general population. MD Anderson has 20,800 employees, and 67% of them are women, which will provide a large starting recruitment pool for the study. In addition, recruiting participants who attend MD Anderson for routine cancer screenings will allow a more varied sample. Women who are interested in the study will first be screened for the following inclusion criteria: 1) age 50-69 years old; 2) postmenopausal woman (absence of menstruation for at least one year, or history of bilateral oophorectomy); 3) self-reported height and weight indicating a BMI ≥18.5 and <25 kg/m^2; 4) no contraindications to exercise (either no positive responses on the Physical Activity Readiness Questionnaire, or clearance from a health care provider certifying that the participant is healthy enough to exercise); 5) no history of invasive cancer, other than non-melanoma skin cancer; 6) no history of renal disease; 7) able to walk without an assistive device; 8) not within 3 months of major surgery; 9) able to speak/read/write in English; and 10) has internet access on a computer or mobile device.

Exclusion criteria: 1) MD Anderson employees that report to the Principal Investigator of this study;2) Participants that cannot engage in the exercise program for more than three weeks during the study period and 3) Participants that are currently doing strength exercises that work all major muscle groups (defined as: participants who complete more than 16 repetitions per exercise in their current resistance training regimen, or who don't find their last 1-2 reps to be difficult in their current regimen, or who increase the weight in their current routine). Women will then be screened for the following additional inclusion criteria at a DXA screening visit: 13) A trunk fat mass ≥ 9.4 kg as indicated by a DXA scan and 14) Height and weight indicating a BMI of ≥18.5 and <25 kg/m^2 verified at the screening visit.

We will use the Physical Activity Readiness Questionnaire (PARQ) to screen participants to ensure that they are healthy enough to do unsupervised physical activity. The PARQ is a seven-item questionnaire to screen for health problems that require additional assessment before an individual engages in an exercise program. If participants indicate they have any of the problems on the PARQ, we will ask them to see a physician to obtain a medical release form.

Women who are eligible based on these criteria will be scheduled for an appointment in MD Anderson's Behavioral Research and Treatment Center (BRTC) where height and weight will be measured, and a whole-body DXA will be performed in those who have a normal BMI (Figure 1). Women who have a verified BMI ≥18.5 and <25 kg/m^2 and a trunk fat mass ≥9.4 kg will be consented for the randomized study.

Figure 1: Study design



Participants will be randomized to a 16-week intervention to improve body composition (decrease fat mass and increase lean mass) or to a wait-list control group. Participants will be randomized using a form of covariate-adaptive randomization called minimization, which is similar to stratification in that participants' characteristics are used to assign them to the treatment conditions.[22, 23] Before a participant is assigned to a group, the number of participants in each group with similar covariate characteristics is totaled. These totals are based on the marginal sums of the covariates so that each covariate is considered separately. Participants' treatment assignments are determined based on which group assignment provides the best overall balance with respect to covariates. We will use as covariates age (<60 vs 60+), race/ethnicity (Asian, Black, Non-Hispanic White, Hispanic White, other), and percentage body fat (<= 41.3% vs >41.3%). Participants will be randomized using the Clinical Trial Conduct (CTC) website.

4.0 Research Plan and Methods

Recruitment: Women will be recruited from the MD Anderson Cancer Center employee population as well as the general population. Our initial recruitment strategy will be to recruit employees who have a medical record number that are identified by the OneConnect team. Because these individuals are MD Anderson patients, they are eligible to be seen for visits in the BRTC during COVID restrictions. The research team will contact these employees and provide them with a brief description of the study.

When we can conduct study visits with the general population in the BRTC, we will promote the study on various communication channels, including social media, the Employee Notes weekly online newsletter, the website of the Center for Energy Balance in Cancer Prevention and Survivorship, the Be Well monthly newsletter, and through the MD Anderson Yammer network. Additionally, we will place recruitment posters around MD Anderson and local organizations and distribute recruitment flyers at wellness events. We will also ask participants to share the study information within their personal networks. The study posters, recruitment flyers, and newsletter articles will include general information regarding the study purpose, study rationale, requirements of participants, benefits of participation, and study contact information.

Pre-Screening: Participants who are interested in the study may complete an online screening questionnaire to assess basic eligibility requirements. Those who meet the initial eligibility requirements will be contacted by phone to complete the remaining screening questions. An initial description of the study will be provided, and the research staff will ask questions on the screening questionnaire (Appendix C). Those who are eligible based on their responses to the screening questions will be scheduled for an in-person visit at MD Anderson's Behavioral Research and Treatment Centers for body composition using DXA and measurement of height and weight.

DXA Screening Visit: Two Consent forms are used in this study: One to cover the DXA screening visit and one to cover enrollment and randomization into the study. The screening consent will be signed at the DXA screening visit and covers procedures that take place prior to enrollment in the study. Participants will undergo a whole body DXA (Dual-energy X-ray absorptiometry) to measure body composition as well as measurements for height and weight. All these procedures are required in order to proceed to enrollment and are not optional. Participants who are eligible after assessment of body composition will undergo informed consent for the randomized trial. Staff will explain the study procedures, as well as risks, benefits, and alternatives at each point of consent. Staff will explain the optional procedures and have the participant select yes or no to each optional procedure. The participants will be given the opportunity to ask questions, and those who are eligible and consent to the study will be scheduled for a baseline assessment. Interested patients and study personnel authorized to obtain consent will sign two copies of the informed consent documents. One copy is for the patient and one copy is for the study records.

If a potential participant is not interested in participating in the study at any point, we will collect information regarding reason for refusal. Additionally, if someone is found to be ineligible during the screening process, we will record the reason for ineligibility. This information will allow comparison of study participants with those who refuse and who are ineligible. This will also inform us of potential barriers to conducting a larger trial.

Employee participation will be completely voluntary, and their decision on whether to participate will not be shared outside the research team. Furthermore, all data will be kept confidential, and will not be shared with the employee's supervisor, co-workers, or anyone outside the research team. Since the study is voluntary, employee injuries occurring from participating in this study are not covered by Worker's Compensation as such activities are outside the course and scope of employment. Participation or non-participation will not affect employment status at the institution or employee benefits. Data will be collected on health behaviors including alcohol and tobacco use, but these data will remain confidential. MD Anderson tobacco policy forbids tobacco use in MD Anderson facilities and vehicles and stipulates that employees must be tobacco-free at the time of their hire/appointment. The policy does not forbid current employees from using tobacco outside of MD Anderson facilities/vehicles, so we do not have an obligation to report employees who are using tobacco.

MD Anderson participants that are employees that meet all eligibility requirements and are enrolled in the main study and are trained by a fitness center trainer will also sign the fitness center indemnification form (attached as appendix). This form is required for all employees that join the fitness center and signatures will be maintained by fitness center staff.

The research staff will also request a mailing address and an email address from the potential participant so that their materials/information on the internet survey can be sent as soon as the consent form is received by the research staff. All surveys will be administered online to the participant's preferred email address.

Assessments: Participants will attend in-person assessments at MD Anderson's BRTC at baseline and 4 months (See Appendix for questionnaires). Participants randomized to the waitlist-control group may also complete an 8-month assessment. To increase compliance with assessments, we will call and send email reminders, and provide compensation. Participants will be contacted by a combination of phone and email up to six times per assessment time point before excluding a participant from the study. Even if participants have not been adherent with the intervention, we will contact them to complete the 16-week assessments and 8-month assessments (if applicable). Before assessment visits, the participants will be given an accelerometer to wear for 7 days (Actigraph wGT3X-BT, Pensacola, FL).The participant will complete 7 days of accelerometer wear within 1 week (+/-) of their assessment visit. The participant will return the accelerometer either in person at the BRTC assessment visit, or by mail using a pre-paid mailing envelope. At each time point, participants will complete 3 online 24-hour dietary recalls using NCI's Automated Self-Administered (ASA-24) online tool (https://asa24.nci.nih.gov/).[24] Three dietary recalls ($4 \geq n \geq 2$) provide a valid estimate of macronutrient intake (e.g., total energy, % energy from fat, carbohydrate and protein), key food group and nutrient data.[25-27] We will email participants a link to online questionnaires to measure demographics, co-morbidities, health behaviors, physical activity (Godin Leisure Time Exercise Questionnaire),[28] sleep (Pittsburgh Sleep Quality Index),[29-31] menopausal symptoms (Breast Cancer Prevention Trial symptom checklist),[32] physical functioning (PROMIS short form),[33] and global quality of life (PROMIS short form).[34,35] The online questionnaires will also assess medications and supplements.. At the assessment visits, we will assess body composition using whole-body DXA (screening DXA will be used for baseline if it was done in the past 30 days); measure height, weight, and waist and hip circumference; and administer a 6 minute walk test and the 30-second sit-to-stand test.[36-38] Skin carotenoids (which provides an indicator of intake of fruits and vegetables) will be measured using the Veggie Meter, a reflection spectroscopy device that non-invasively measures skin

carotenoid levels. The study coordinator will have the participants place their index finger into a 'finger cradle' for 10 seconds three times to get an average measure of F/V. At each time point, we will measure participant's one-repetition maximum (1RM) of the strength exercises in the resistance training program to calibrate intensity of the strength training protocol. 1RM tests will be performed in the following sequence, ordered from largest to smallest muscle groups and alternating between upper and lower body movements to allow for sufficient rest: leg press, chest press, leg extension, seated row. We will collect a fasting (≥8 hours) blood sample to measure plasma biomarkers of inflammation and metabolism. Fasting glucose and lipids will be assessed by LabCorp. Plasma will be processed in the BRTC or the Population Science Laboratory at MD Anderson, aliquoted, and kept frozen in a -80°C freezer. At the end of the study, samples will be shipped on dry ice to the Pollak lab at McGill University for analysis of hsCRP, IL-6, leptin, adiponectin, insulin and SHBG. The blood samples will not be identifiable, and no PHI will be shared with McGill University.

Assessments – Optional Procedures: Participants will be given the option to complete additional procedures at the in-person assessments.

Optional Continuous Glucose Monitor (CGM): CGM measures blood glucose concentrations in the interstitial fluid in real-time through a tiny sensor inserted under the skin. CGM are increasingly being used in research that demonstrates the effect that physical activity has on improving insulin sensitivity and glucose metabolism in both diabetic and non-diabetic populations. Participants interested in participating in the CGM optional procedure will have the CGM inserted into the back of the upper arm at the baseline and 4-month follow-up visits and wear it for 14 days each time. Participants can choose either to self-insert the sensor during the session or to have the CGM inserted by trained study staff. If the participant prefers self-insertion, instructional videos available from the Freestyle Libre website will be used to guide the sensor insertion along with staff assistance. Study staff will activate the sensor through a CGM reader. Upon activation, the inserted sensor will start recording blood glucose data continuously for 14 days. At the end of 14 days, participants will remove the CGM sensor independently or with help from the study staff and will return the sensors to study staff via a prepaid mailing envelope or drop-off at the BRTC.

Optional Urine Sample: Participants will be offered to participate in the optional urine sample collection to measure F2- isoprostanes and the urine metabolite. Those who participate will be instructed to collect a urine sample in a urine collection cup during the baseline and 4-month follow-up visit. Urine samples will be received and stored by the by the Population Science Laboratory at MD Anderson and then shipped to the Eicosanoid Core Lab at Vanderbilt University for analysis at the end of the study. The urine samples will not be identifiable, and no PHI will be shared with Vanderbilt University.

Optional Stool Sample Collection: Participants interested in participating in the optional stool collection procedure will be provided with the OMNIgene GUT kit before their baseline and follow-up visits. This kit includes all the supplies and instructions needed to collect a stool sample at no cost to the participant. Storage media in the collection tube stabilizes the sample at room temperature for up to 2 months from the date of collection. Study participants may feel uncomfortable collecting stool samples, but the collection procedure poses no physical risk to participants. Participants that agree to the optional procedure will answer 3 questions related to the stool sample collection regarding bowel procedures, colonoscopy, and medications with a focus on antibiotics, antifungals, antiparasitics, antivirals and/or probiotics within one month of stool sample collection. This information may be

verified by phone, email, or in-person communication and recorded in the secure study database.

Upon receipt, stool samples will be received by the Population Science Laboratory (PSL) at MD Anderson. Participants will either mail the sample directly to PSL or bring the sample to the BRTC at their baseline and follow-up visits for the study staff to deliver to PSL. Samples labeled with a study ID (no PII) will be aliquoted and long-term banked at -80C and/or transferred in batches to the microbiome service cores for sequencing. All biological specimens will be labeled with a unique study identification number with no other personally identifiable information when provided to collaborating laboratories or service cores for further processing and analysis.

8-month assessment: Participants that are in the waitlist-control group will be offered the intervention after the 4-month assessment. Participants that opt into the delayed intervention will be asked to complete an 8-month assessment that encompasses all procedures as the 4-month assessment with the exception of the optional procedures.

Measure	Baseline	16-Week Follow-Up	32-Week Follow-Up (Control group only)
Accelerometer (physical activity)	X	X	X
ASA-24 dietary recalls	X	X	X
Demographics	X		
Co-morbidity	X	X	X
Health behaviors (alcohol and smoking)	X	X	X
Godin Leisure Time Exercise Questionnaire (self-report physical activity)	X	X	X
Pittsburgh Sleep Quality Index	X	X	X
Breast Cancer Prevention Trial symptom checklist (menopausal symptoms)	X	X	X
PROMIS short form – physical functioning	X	X	X
PROMIS short form – global (quality of life)	X	X	X
Body composition (whole-body DXA, height, weight, and waist and hip circumference)	X	X	X
Cardiorespiratory fitness (6-minute walk test, 30 second sit-to-stand)	X	X	X
Blood sample (plasma biomarkers of inflammation and metabolism)	X	X	X
Veggie Meter (F/V intake)	X	X	X
Optional procedures: CGM, urine sample, stool sample	X	X	
Satisfaction		X	X

Participants randomized to the intervention arm will receive a Fitbit, a wearable physical activity tracking device. They will set up a Fitbit account, which will give them access to an online portal and a mobile app that allows them to view their tracked physical activity information. Participants in the intervention arm will receive the Fitbit, Aria weight scale, food scale, and resistance bands at the baseline assessment, including instructions on how to set up the Fitbit and scale.

Compensation: To compensate participants for their time spent on assessments, they will receive up to $50 in electronic gift cards for each assessment completed, which will be delivered to their preferred email address. At the baseline, 4-month and 8-month assessments, gift cards will be distributed as follows: $30 for completing the in-person assessment and completing the online questionnaire (these two items are required at baseline in order to continue on study), $5 for returning the accelerometer after 7 days of wear, $5 for completing each of the three assigned ASA24 dietary recalls, and a $10 bonus for completing all assessments.

Participants that are not MD Anderson employees working on the main campus can be provided a parking voucher at each in-person assessment to cover parking fees in the Mays Clinic garage.

Participants will be allowed to keep their Fitbit at the completion of the study. If the Fitbit is lost, stolen, or damaged, participants will not have to pay for a replacement. Once it has been reported as such to study staff, we will loan a replacement Fitbit to the participant for the remainder of their participation in the study. If the loaner Fitbit is lost, stolen, or damaged, another loaner will be provided. Loaner Fitbits will be returned to study staff after the completion of the study either in person or via a prepaid envelope mailed to the participant.

Participants will not keep the accelerometer. If it is lost, stolen or damaged, participants will not have to pay for a replacement. Once it has been reported as such to study staff, we may send a replacement accelerometer, depending on the timeframe when it is reported. This decision will be up to the discretion of the research team and the Principal Investigator.

Within six weeks of the end of the follow-up assessment, we will provide participants with the results of their cardiorespiratory fitness tests and changes in body composition as measured by the DXA. Participants that are deemed ineligible at the DXA screening visit will get their DXA results after the screening visit.

Intervention: The intervention will involve two high-resistance circuit training sessions per week,[40] 150 minutes of moderate intensity aerobic training, and diet recommendations that include higher protein (approximately 25% of calories from protein) and modest caloric restriction (estimated energy expenditure less 5-10%). This circuit-style resistance training approach has previously demonstrated to safely improve body composition in normal-weight women.[41] Further, this resistance training approach alone, energy restricted higher protein dietary approach alone, and the combination of these approaches have demonstrated improvements in body composition among overweight and obese adults.[40, 42-44]

Exercise: Participants randomized to the intervention group will complete their resistance training virtually via videoconference. Participants may have the option to complete some resistance training in person as the facility permits. Participants will complete two sessions per week under the supervision of

a personal trainer. The method of resistance training utilized will be high-resistance circuit training, which is a circuit-style resistance training approach. Consistent with previous research,[51] an undulating periodization program will be followed (see Table 1), which produces superior results compared to non-periodized resistance training regimens.[45] Participants will rest for 2 minutes between each series (sequence of three exercises) within each circuit and for 2 minutes between the circuits. After a general, full body warm-up, participants will complete the exercises in the circuit (one or two) using the following sequence: 12 repetitions at 50% 1RM, 10 repetitions at 60% 1RM, 8 repetitions at 75% 1RM. Percentages of 1RM will be estimated based on participants' ratings of perceived exertion during exercise sets. Exercise reps may be adjusted at the discretion of the trainer and based on the participants' fitness level. Participants will also weigh themselves once a week at home using the connected scale provided. The health coach will monitor the weight logging for compliance and will provide reminders as needed.

Participants will be encouraged to engage in safe practices before and after exercise, such as maintaining adequate hydration before, during and after exercise, engaging in an adequate warm-up and cool-down, stretching after each work out, and ceasing exercise if feeling lightheaded or dizzy. Personal trainers will be certified by national organizations such as ACSM (American College of Sports Medicine) or ACE (American Council on Exercise) and trained to teach the circuit training safely. Personal trainers will also be CPR (cardiopulmonary resuscitation) and first aid certified. If a participant becomes injured while on study and is unable to perform the prescribed exercises, we will recommend they consult with their physician. The study team will collect the participant's location for each virtual visit. In the case that the study team witnesses a medical emergency that requires immediate medical attention during the virtual visit, the study team will call 911 and provide the participant's address. In the case of a medical emergency that requires immediate medical attention while in the fitness center, the fitness center staff will call CODE BLUE to comply with MD Anderson policy. Participants that cannot engage in the exercise program for more than three weeks of the program may need to withdraw from the program, however, this will be up to the participant and Principal Investigator's discretion.

Aerobic exercise will be done in whatever setting the participant chooses. We will prescribe 150 min of aerobic exercise/week, after an initial 4-week ramp up period (see Table 1). In most cases, the participants will do brisk walking, but they may substitute other activities that enable them to meet the cardio heart rate target as calculated by the Fitbit Charge. This is between 60-84% of their heart rate reserve. Participants will receive a free Fitbit® activity monitor with heart rate monitoring (which they can keep) to help them adhere to exercise and heart rate targets.

Participants who miss or are not able to complete a supervised training session will be encouraged to do the exercises on their own. Participants will be provided with a video and/or written instructions to describe the exercises. If a participant completes a resistance training session on their own, it will be documented as an unsupervised session in the study database.

Employee participants who leave MD Anderson before completion of the study will be allowed to continue in the study. In these instances, the supervised exercise sessions can be held virtually or in the exercise room of the Behavioral Research and Treatment Center as the facility permits. These sessions would be supervised by the PI's study team and not fitness center employees.

Table 1: Week by week intervention outline.

Week	Health coaching with Registered Dietitian or Health Educator	Resistance exercise	Aerobic exercise	
			Min/week	Sessions/week
1	Health coaching lesson 1	1 set/circuit	50	2-3
2	Health coaching lesson 2	2 sets/circuit	75	3-4
3	Health coaching lesson 3	3 sets/circuit	100	3-4
4	Health coaching lesson 4	1 sets/circuit	125	3-5
5	Health coaching lesson 5	2 sets/circuit	150	3-5
6	Health coaching lesson 6	3 sets/circuit	150	3-5
7	Health coaching lesson 7	3 sets/circuit	150	3-5
8	Health coaching lesson 8	1 sets/circuit	150	3-5
9	Health coaching lesson 9	2 sets/circuit	150	3-5
10	Health coaching lesson 10	3 sets/circuit	150	3-5
11	Health coaching lesson 11	3 sets/circuit	150	3-5
12	Health coaching lesson 12	1 sets/circuit	150	3-5
13	Health coaching lesson 13	1 sets/circuit	150	3-5
14	Health coaching lesson 14	2 sets/circuit	150	3-5
15	Health coaching lesson 15	3 sets/circuit	150	3-5
16	Health coaching lesson 16	1 sets/circuit	150	3-5

Diet: Reduced calorie, higher protein diets have been shown to reduce fat mass while preventing the loss of lean mass.[44] We will recommend that participants eat a modestly hypocaloric diet (5-10% less than their individualized total daily caloric needs). Individualized total daily caloric needs will be calculated using the revised Harris Benedict equation[46] to calculate basal metabolic rate (667 + [9.740 * weight in kg] + [1.729 * height in cm] – [4.737 * age in years] and multiplied by an activity factor of 1.55 to account for the recommended exercise regimen. We will then subtract 5-10% of the participant's calculated total daily caloric needs in order to determine her caloric prescription for the intervention. The macronutrient distributions within the dietary prescription are as follows: approximately 25% of calories from protein, 45-50% of calories from CHO, and 25-30% calories from fat. Participants will be counseled by either a registered dietitian or master's level health educator regarding strategies to meet the dietary goals of the intervention. For example, to meet the higher protein macronutrient goal (~25% kcal protein), participants will be instructed to increase their consumption of lean protein food sources (e.g., skinless, boneless chicken breast; broiled fish; legumes; tofu; Greek yogurt; etc.) and/or incorporate protein supplementation into their typical dietary routine (i.e., why protein shakes). Additionally, sample eating plans will be provided and dietary advice will be consistent with dietary guidelines for cancer prevention recommended by the American Cancer Society[49] and the American Institute for Cancer Research. Participants will be instructed to use either My Fitness Pal® or Fitbit for dietary tracking, both of which are popular commercially available free websites/mobile apps that allow for dietary and activity tracking (the Fitbit can be paired with My Fitness Pal to automatically import activity data) and provide feedback on energy balance and macronutrient intake. Participants will also have weekly sessions with either a health coach or a registered dietitian to support adherence to the exercise and diet recommendations. Coaching and behavioral change methods will be based on the evidence-based Diabetes Prevention Program, which we have successfully applied in other intervention studies.[31, 47] Body weight of participants randomized to the intervention group will be monitored weekly; if weight loss exceeds 2% in any 3-week period, we will increase the participant's caloric prescription to her calculated total daily caloric needs.

Control group: To help retain the control group participants and keep them from beginning a lifestyle change program, we will use a waitlist control design. They will be offered the intervention after the 4-month assessment. Participants who opt in to the delayed intervention will complete an 8-month assessment.

Because this is a low-risk study, we will not file deviations if participants miss a single exercise or coaching session. Instead, we will log deviations for participants that meet the following criteria at the end of their study participation: missed more than 50% of total supervised resistance training sessions and/or missed more than 70% of coaching sessions with a health coach. We will maintain study records to document these missing data and will account for these in our analyses.

Biomarkers:
Plasma levels of insulin (Mercodia), as well as leptin, adiponectin, SHBG, hsCRP, and IL6 (R&D Systems) will be measured by ELISA as in our previous studies.[18, 21, 50] Serum levels of total, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) cholesterol and triglycerides will be determined in MD Anderson's clinical chemistry lab.

5.0 Statistical Considerations and Sample Size Justification

Aim 1: Evaluate the feasibility of a diet and exercise intervention to decrease body fat in postmenopausal women with normal BMI but high body fat. Feasibility will be assessed based on: i) eligibility rates; ii) intervention adherence; and iii) retention. Eligibility rate will be calculated as the number of eligible women identified divided by the total number completing a DXA scan. Intervention adherence for an individual is defined as attending at least 11 of the sessions with the personal trainer and at least 11 of the health coaching sessions. The intervention adherence at the study level is then defined by the percentage of participants adhering to the intervention. Retention will be assessed by the percentage of participants completing the 4-month study assessments. We will calculate rates, frequencies, and 95% confidence intervals (CIs) for these measures. Criteria for feasibility will be defined as: i) an eligibility rate of ≥ 40%; ii) a study intervention adherence rate of ≥ 75%; and iii) a retention rate of ≥ 80% in each arm. All three criteria need to be met to declare the study to be feasible.

Aim 2: Assess preliminary efficacy of the intervention on (1) circulating markers of inflammation and metabolic dysfunction linked to both excess adiposity and breast cancer; (2) body composition. Exploratory outcomes include fitness, behavior, and quality of life. Hypothesis: A lifestyle intervention that emphasizes strength and aerobic training and a high-quality diet with increased protein will reduce adiposity in the absence of muscle loss, lower circulating levels of markers of inflammation, and improve metabolic dysfunction in normal BMI women with excess body fat.

We will assess the effects of the intervention on each outcome variable evaluated at the 4-month assessment by: 1) calculating its mean and SD by arm; 2) performing a two-sample t-test; and 3) testing the between-group difference using linear regression analysis, controlling for the baseline measurement as well as the factors used in the minimization: age (<60 vs 60+), race/ethnicity (Asian, Black, Non-Hispanic White, Hispanic White, other), and % body fat (<= 41.3% vs >41.3%).[48] Additional exploratory analyses will treat both of the covariates age and % body fat as continuous variables. Tests will use a two-sided 5% significance level without adjustment for multiple testing. Given the exploratory nature of the study, results of the tests will be used for hypothesis generation. Finally, descriptive statistics including, e.g., mean, SD and CI for mean, will be calculated to assess within-group change of each outcome variable between the 4- and 8-month assessments for participants who opt into the intervention in the control group.

Sample Size Justification: With the remaining funding for the study, we will not be able to recruit the originally planned 62 participants. Thus, we propose to reduce the sample size of the study to 40 participants (20 per group), based on the currently observed data in the trial. The new sample size is still justified mainly based on evaluation of the primary endpoints of feasibility (Aim 1), by incorporating current estimates of the relevant parameters.

If the true eligibility rate is 45%, then there will be a 87% probability for us to screen 100 participants or less to arrive at 40 participants who are eligible and randomized; if the true eligibility rate is 35%, then there will be only an 17% probability for us to screen 100 participants or less to arrive at 40 participants eligible and randomized. Alternatively, if the true eligibility rate is 50% (as is close to the currently observed eligibility rate of 52% in the trial), then there will be a 98% probability for us to screen 100 participants or less to arrive at 40 participants who are eligible and randomized.

If the true intervention adherence rate is 80% (or 70%), then there will be an 80% (or 42%) probability for us to observe a >= 75% intervention adherence rate (i.e., at least 15 out of 20 patients adhering to the intervention). However, if the true intervention adherence rate is 85% (a more conservative estimate than the currently estimated intervention adherence rate of 89% [=8/9]), then there will be an 93% probability for us to observe a >= 75% (15 out of 20 patients) intervention adherence rate.

If the true retention rate in each arm is 85% (or 75%), then there will be an 83% (or 41%) probability for us to observe a >= 80% retention rate (i.e., at least 16 out of 20 patients are retained in each arm). If the true retention rate across arms is 85% (or 75%), then there will be an 86% (or 30%) probability for us to observe a >= 80% retention rate (i.e., at least 32 out of the 40 participants completing the 4-month assessment). Note that a retention rate of 85% is a more conservative estimate than the currently estimated retention rates of 89% (= 8/9), 100% (= 10/10) and 95% (= 18/19) in the intervention, control and across groups, respectively.

To justify the sample size of approximately 16 participants per arm (after 20% attrition) for assessing the effect sizes (Aim 2), a two-sided 95% CI for the difference of two means between arms will extend 0.693 SD unit from the observed difference in means, assuming that the CI is based on the large sample z statistic (nQuery 7.0).

In sum, while the precision of the effect size estimates for the secondary aims (specifically, Aim 2) will be reduced due to the reduced sample size, the “power” of declaring study feasibility (Aim 1) is generally increased once incorporating the current estimates of the feasibility parameters without considerably increasing the probability of falsely declaring study feasibility when the study is infeasible.

6.0 Data Monitoring Plan

Because this is a pilot study involving minimal risk interventions, we request a waiver of Data and Safety Monitoring Board (DSMB) review.

Study data will be collected and managed using REDCap (Research Electronic Data Capture) electronic data capture tools hosted at MD Anderson. REDCap (www.project-redcap.org) is a secure, web-based application with controlled access designed to support data capture for research studies, providing: 1) an intuitive interface for validated data entry; 2) audit trails for tracking data manipulation and export

procedures; 3) automated export procedures for seamless downloads to common statistical packages; and 4) procedures for importing data from external sources. In the case of multi-center studies REDCap uses Data Access Groups (DAGs) to ensure that personnel at each institution are blinded to the data from other institutions.

REDCap (https://redcap.mdanderson.org) is hosted on a secure server by MD Anderson Cancer Center's Department of Research Information Systems & Technology Services (RISTS). REDCap is a two-tier pHp-based web application that can be hosted on a variety of hardware and operation systems. The application also relies on web server software and a database server to function properly. RISTS works with the Data Center Operations Team (DCOT) to deploy and configure the servers which host REDCap. The application is deployed on a RedHat Linux server that is maintained and monitored by DCOT. The open-source Apache web server is used in the REDCap deployment. The servers are backed up nightly and have well-defined processes for disaster recovery. The database used to store project related information is maintained in a MySQL database cluster which is supported by a RISTS Database Administrator (DBA). The database server, like the application server, also has a well-defined process for disaster recovery.

REDCap has undergone a Governance Risk & Compliance Assessment (05/14/14) by MD Anderson's Information Security Office and found to be compliant with HIPAA, Texas Administrative Codes 202-203, University of Texas Policy 165, federal regulations outlined in 21CFR Part 11, and UTMDACC Institutional Policy #ADM0335. Those having access to the data file include the study PI and research team personnel. Users are authenticated against MDACC's Active Directory system. The application is accessed through Secure Socket Layer (SSL). All protected health information (PHI) will be removed from the data when it is exported from REDCap for analysis. All dates for a given patient will be shifted by a randomly generated number between 0 and 364, thus preserving the distance between dates. Dates for each patient will be shifted by a different randomly generated number. Following publication study data will be archived in REDCap.

Furthermore, all records that will not be collected electronically will be contained in a participant specific folder in a locked office or file room. The principal investigator and designated study personnel will have access to these data files.

The Fitbit privacy policy is provided to all participants. Fitbit.com will have access to participant email addresses. Fitbit data will be collected by the research staff through a third-party company, Fitabase. Fitabase may have access to Fitbit activity data (health behaviors) and participant email addresses. Participant names will not be disclosed to Fitabase.

All participants will be registered in the CORE (Clinical Oncology Research) or OnCore system at MD Anderson. Adverse events will be reported to the IRB (Institutional Review Board) by the research team, in compliance with MD Anderson procedures. Because this is a low-risk study, we will not file deviations if participants are unable to complete a survey or use study devices. Instead, we will maintain study records to document these missing data and will account for these in our analyses.

7.0 References

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