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Validation of pRMR Equations in a Non-Caucasian Sample – Ethnicity as a Variable in Predicting Resting Metabolic Rate

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ABSTRACT

VALIDATION OF pRMR EQUATIONS IN A NON-CAUCASIAN SAMPLE – ETHNICITY AS A VARIABLE IN PREDICTING RESTING METABOLIC RATE

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This study intends to identify a predictive resting metabolic rate (pRMR) equation that is predictive of the measured resting metabolic rate (mRMR) in a sample that includes a group of Black Americans. The handful of commonly used pRMR equations, such as the Mifflin- St. Jeor or Harris-Benedict equations, were created without defining the demographics of their populations, while validation of these equations is typically done with almost exclusively Caucasian subjects. Black and brown Americans require the same evidence of precision in predictions of daily energy needs as Caucasian Americans. When applied to non-Caucasian samples, these equations appear in the literature to be overestimations.

Potentially, differences in body composition and lean body mass (LBM) by ethnicity are the root of this inaccuracy. Thus, ethnicity is an apparent variable worthy of consideration and pRMR equations accurate in a Black sample may need to include LBM to estimate kilocaloric needs. To that end, a sample of 36 Black subjects (Group 1) and 32 Caucasian subjects (Group 2) participated in indirect calorimetry for the determination of which equations are accurate or inaccurate in these respective populations. Thirteen pRMR equations from peer-reviewed articles were compared to subjects' measurements.

The Cunningham equation emerged as the best predictor for the entire sample which included Black and Caucasian participants. The Mifflin – St Jeor equation was surprisingly accurate for the Black group and not the Caucasian group, while the Harris – Benedict equation was accurate

for the Caucasian group alone. Regression analyses revealed significant ΔR^2 when ethnicity was added to the hierarchy. Ethnicity appears to play a role in the validity of pRMR equations. Further research with diverse samples should be done with a larger sample size to fully elucidate the magnitude and direction of the effect ethnicity has on mRMR and pRMR equations' accuracy to identify equations that are generalizable to the entire population.

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Dedication

To all those fighting mediocrity

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CHAPTER 1 INTRODUCTION

To predict an individual's total daily energy expenditure, or metabolic rate, in the course of treatment, a clinician must either predict or measure resting metabolic rate. In the absence of indirect calorimetry, the gold standard of measurement[1], a predictive equation is used to estimate resting metabolic rate in non-critically ill patients. There is no gold standard equation for predicting resting metabolic rate (pRMR) for medical and allied health professionals, but the Mifflin - St. Jeor equation[2] comes close. This formula and many others are often used as an important step in several clinical interventions when indirect calorimetry is unavailable.

Early predictive energy equations relied on body surface area as the largest contributor to basal metabolism.[3] This changed with the Harris-Benedict (HB) equation which used multiple regressions to assign significance to weight, height age, and gender.[3] Inaccuracies in prediction were noted with the HB and the Cunningham equation, developed in 1980, used Harris-Benedict sample data to suggest non – gendered formulas based on lean body mass[4].

Because Cunningham did not create a new sample from which to gather data and discrepancies between pRMR and measured resting metabolic rate (mRMR), discrepancies in using either Cunningham or HB formulas could not be properly explained,[5] a new multiple regression analysis of discrete subjects' indirect calorimetry suggested, that a new variable, weight, had the largest and most consistent correlation to mRMR. Thus, Owen et al. created two new pRMR formulas based on sex, with weight as the only significant variable.[5]

The Mifflin - St. Jeor equation (MSJE), unlike the Owen equation, applies statistical significance to four variables: weight, height, gender, and age. In comparison to the HB and Cunningham, the MSJE is said to be accurate for the "modern-day population." It used a discrete sample of a wide variety of individuals with various body types;[2] data collection began in 1985.[6] After, other researchers recommended the MSJE be considered the gold standard for calculating resting metabolic rate in obese and non-obese adults. Any inaccuracies were explained as misnomers: "the assumption that the [MSJE] is in error, rather than the individual's metabolic rate is nonstandard, is often made because some of the calculation standards were produced from subject groups whose age, body size, body composition, and race do not conform to today's US population."[7] The equation has been verified and validated several times in populations of varying genders and BMIs.[7–9]

While the MSJE has been touted as the most accurate for predicting resting energy expenditure in the general population, this equation, like the others, has a systematic selection bias resulting in limited generalizability. The samples from which these equations are derived do not represent the breadth of the United States population. The participants' racial demographics are largely undefined in the studies, and in the studies offering validation, samples are primarily Caucasian. Subsequent validations in non-white populations suggest the MSJE is a significant overestimation of resting energy expenditure in non-Caucasians[10–14], specifically, Black Americans[12,15–17,14,11]. Even Frankenfield et al. recanted the previous recommendation of "gold standard" 2 years later, positing "noteworthy errors and limitations [of the MSJE] exist when it is applied to individuals and possibly when it is generalized to certain age and ethnic groups."[9]

Furthermore, the Food and Agriculture Organization of the United Nations, which utilizes equations meant for global populations, acknowledges a similar deficit in precision from a lack of representation in its sample populations.[18] The necessity of an accurate, generalizable equation for predicting the resting energy expenditure in a population with all its ethnicities is apparent. To date, a review of the current research suggests there have been few studies designed with a distinct sample of Black Americans to determine an accurate method for predicting resting energy expenditure.

Statement of Problem

The use of popular pRMR equations to estimate energy expenditure for the non-critically ill has questionable accuracy in non-Caucasian subjects. The designation of Caucasian Americans as a measurement standard implies that non-Caucasians are outliers or extenuating circumstances. The American population is diverse; if pRMR equations inaccurately estimate energetic requirements in non-Caucasian populations, then they detract from the competency and quality of care offered to these populations.

Elimination of the very real, and very large population of Black Americans and other people of color that will inevitably use America's healthcare system undermines the validity and generalizability of any population measure or assessment. To provide an idea of just how large the non-Caucasian population, i.e., black, indigenous, or other people of color (BIPOC), is, the U.S. Census Bureau reports the U.S. population consists of roughly 328 million people[19]. They further report that only 60.1% of the population is Caucasian American and non-Hispanic. It follows then that 39.1% or 128,341,654 people in the United States are not Caucasian. Ostensibly, those would appear to be significantly large enough numbers to prompt inclusion in any study claiming to be representative of, or applicable to, the general population.

The correlational relationships between race and health status can become blankets that obscure the quality of evidence applied to non-Caucasian patients. Chronic diseases are often treated as if endemic to certain populations rather than the outcome of applying clinical practices or recommendations that lack evidentiary confirmation. The blanket, in this study, is the suggested inaccuracy in estimation of energy expenditure in Black Americans. Treatment of Black and brown Americans requires the same evidence required to treat Caucasian Americans. A 'one-size-fits-White' approach ignores, not only Black Americans but any indigenous or other people of color receiving interventions designed for Caucasian populations.

Background and Significance

Research suggests that the current most used equations for predicting resting metabolic rate are inaccurate in non-Caucasian populations.[11,12,14,16,17] Studies regularly demonstrate significant differences in mRMR between Caucasian and non-Caucasian samples; mRMR is significantly lower in Black Americans when compared to Caucasians[14]. Resting metabolic rate is approximately 5% higher in Caucasian Americans, regardless of gender or BMI.[15] While the exact number of calories for each individual varies, one study sample found the African American race was associated with a mean 144 $kcal \cdot day^{-1}$ decrease in mRMR.[17]

Commonly used pRMR equations generate similar discussions of ethnicity-specific differences in studies of non-Caucasian demographics, as well.[10,20,21] Differences in energy requirements exist between different ethnic groups and further research is needed to better elucidate them[22] as many formulae were created in consideration of gender differences, but not ethnic. In studies that contain samples consisting of Black Americans, research regularly shows an overestimation of resting energy expenditure by the MSJE and HB formulas, where no such difference is evident in Caucasian populations. One study states directly that commonly used equations systematically overestimated daily caloric requirements in Black Americans by as much as 286 kcal·day^{-1}.[17] The overestimation persists throughout the research; Another investigation found the use of HB, FAO, and MSJE equations overestimated caloric needs in Black females by 138 ± 20, 495 ± 355, 360 ± 288 kilocalories per day, respectively.[23]

For a more simplified context, a person consuming 286 kcal·day^{-1} in excess of their true energy requirements will gain 30 pounds per year. At the end of 2 years, it would be no surprise to find such an individual obese by BMI standards. The rates of obesity seemingly mirror the differences in accuracy of estimation; in 2018, the CDC reported 40% of Black Americans were obese.[19] Other groups typically unrepresented in investigations of metabolic rates show similar rates of obesity: American Indians 39.1%, Hispanic 34.2%, and, while obesity plays a role in all demographics, Caucasians show far less prevalence at 29.9%.[19] Such statistics do not provide any measure of causality; however, they do provide an interesting insight into the potential consequences for the relative overestimations of energy needs in different demographics. Inaccurate RMR predictions that contribute to deviations from eucaloric intake may contribute to unintentional weight gain or loss in any population.[24]

To clarify, overweight and obesity are classified by BMI values of 25 – 29.9 kg·m^{-2} and 30 – 34.9 kg·m^{-2}, respectively. Incidences of major depressive disorder, and rheumatoid arthritis increase with BMI's over 25. Obese patients are at an increased risk for dyslipidemia, heart disease, and stroke. Obesity is a risk factor for esophageal, colon, and liver cancers, among others,[25]and is responsible for an overall mortality increase of 29%, a 41% increase in vascular mortality, and a diabetes-related mortality increase of 210%.[26] Since the etiology of obesity is complex, excessive caloric intake is only one possible cause that could be potentially mitigated with accurate predictions of resting metabolic rate.

RESEARCH PURPOSE

The primary focus of this analysis is to determine which currently existing pRMR equation, if any, provides a reliable prediction of RMR in a sample including Black Americans. Derivatively, this study aims to increase the prescience of the need for accurate assessments, recommendations, and evidence-based clinical practices in every population. Tangentially, the overall contribution to the literature will hopefully highlight the potentially overlooked obligation for representative sampling in population-specific applications and will emphasize the systematic nature of inadequacies in health care by further elucidating the relationship between RMR and ethnicity.

RESEARCH QUESTIONS AND OBJECTIVES

HYPOTHESES

- mRMR in the Black Group is less than the Caucasian Group
- MSJE and HB equations overestimate resting energy expenditure in the Black Group
- Lean Mass based equations will predict RMR for Black Americans with more accuracy than weight-based equations

OBJECTIVES

- Find a clinically applicable method that accurately predicts RMR for Black Americans.
- Determine whether estimating energy expenditure should consider ethnicity.
- Highlight the necessity of evidence of precision in predictions of daily energy needs for BIPOC populations

Chapter 2 Literature Review

Resting Metabolic Rate

Resting energy expenditure (REE), resting metabolic rate (RMR), and basal metabolic rate (BMR) all refer to the energy requirements for maintenance of basic metabolic function and accounts for the largest portion of total daily energy requirements, approximately 70%.[2,4,20] Total daily energy expenditure (TDEE) includes resting metabolic rate as well as energy required for physical activity; for example, a bedridden or immobile individual would have a TDEE equal to RMR.[4]

Highly metabolically reactive organs (HMRO) account for a majority of basal metabolic requirements.[27] In descending order of caloric demand, the heart, kidneys, brain, and liver, considered HMRO, are followed by skeletal muscles, other organs, and finally adipose.[27] Specifically in units of $kcal \cdot kg^{-1} \cdot day^{-1}$, 440 are required by the heart and kidneys, 240 by the brain; 200 are required by the liver, 13 by skeletal muscle, 12 for residual organs and tissues, and 4.5 for adipose tissue in adults under 50 years;[27] for those over 50, those requirements are not significantly less.

A sample of 64 women (n = 34 Black Americans, n = 30 Caucasian Americans) and 35 men (n = 8 Black Americans, n = 27 Caucasian Americans) performed forty contiguous axial magnetic resonance images, dual-energy X-ray absorptiometry scans, and indirect calorimetry measurements on all subjects. This data yielded 2 studies[28,29] that demonstrated individual variations in the masses of the organs account for 70% of mRMR; the inclusion of the trunk accounted for 75% of the variability.[28] Additionally, there is a significant difference in the average masses of organs between

Black ethnicity and Caucasian ethnicity.[29] Black HMROs appear to have a lower average mass than Caucasian Americans.[17,28,29] Similarly, women typically have a lower resting metabolic rate than men[2,3,13,23], which is likely also due in part to overall differences in HRMO mass, which may also account for the reduction in metabolic needs as age increases. While age and sex as variables are generally acknowledged as significant in nearly every predictive metabolic equation, ethnicity is not.

Relatively small idiosyncratic variations in HMRO mass significantly affect resting energy expenditure across individuals,[28] but resting metabolic rates are relatively constant within individuals, save a few exceptions. External factors like sleep deprivation[14], changes in lean body mass[16], illness or injury[1], nicotine[30], caffeine[31], and physical activity[14] can be affective. Food intake is thermogenic and requires energy; the use of stimulants and illness, especially critical illness, can increase resting metabolic rate as well[1]. These attributes must be controlled for when measuring RMR.

mRMR

Indirect calorimetry has long been the reference standard for measuring resting metabolic rate as all energy-producing reactions in the body depend on oxygen use or carbon dioxide production. The measurement of gas exchange during respiration provides the ratio of O_2 consumed to CO_2. The ratio of these provides a respiratory quotient (RQ).

$$RQ = \frac{CO_2\ produced}{O_2\ consumed}$$

However, since RQ is a measure of cellular respiration, the respiratory exchange ratio (RER), which is easier to assess, approximates cellular metabolism by measuring oxygen inspired, VO_2, and carbon dioxide expired, VCO_2.[32] RER is typically measured with respirometry. The rate and ratio of O_2 to CO_2 allow metabolic rate calculations using the Weir equation[33] in units of $kcal \cdot min^{-1}$ which can be extrapolated into $kcal\cdot day^{-1}$. Other devices measure RER and create outputs in $Kcal\cdot min^{-1}$ or $Kcal\cdot day^{-1}$: the *Deltatrac II*, as a tool of indirect calorimetry, has been validated as a reference standard. The *Vmax series* was found to demonstrate acceptable validity in comparison to *Deltatrac II* with 5-10%.[1]

Indirect calorimetry provides measurements of energy expenditure during the moments measured. A person in an excited or nervous state may breathe at a different rate than they might if they were truly at rest. Thus, to facilitate accurate measurements, energy needs at rest should be performed in quiet rooms with mild lighting.[1] The temperature of the room can affect metabolic rate, so repeated measurements should be done in consistent conditions. The nature of the equipment or measurement itself can cause nervousness or excitement which will present a high coefficient of variation in readings. This can be mitigated by facilitating quiet time in subjects before testing[34]. Typically, ten minutes of resting should provide a coefficient of variation of less than 10%, achieving a steady state, with the first five minutes of data discarded.[34,35]

Further suggestions for indirect calorimetry require 12 hours of fasting from food and nicotine and 24 hours of abstinence from physical activity. The test itself will show variations in expenditure depending on the position of the subject, so measurements should be taken while reclining[1].

PRMR

In the absence of indirect calorimetry, resting metabolic rate can be predicted rather than measured. Several equations are available; this review will cover the most commonly used equations and others relevant to this study's demographic.

The Harris-Benedict equation is often viewed as the first equation created for predicting resting metabolic rate and a discussion of metabolic equations would be remiss to not include it. Indeed, the work was published in 1918.[3] A sample of 136 men, 103 women, and 94 newborn infants without fevers in self-reported good health revealed a significant difference in the kilocaloric requirements, referred to as "heat production" of adults that was not present in infancy. The researchers further noted that variability in the weights of internal organs reflected variations in individual heat production and that the formulation itself could only be applied to men or women of "the same race as the anthropologist knows them."[36] Subjects were to lie still and quietly while kilocaloric demand was measured indirectly, via oxygen consumption and carbon dioxide output through a Douglas bag. Fifteen-minute measurements were taken. Multiple regression models dismissed the previously held notion of body surface area as a significant predictor of metabolic need and instead suggested that weight, stature, and age were better predictors of resting energy expenditure.[3,36] Recent attempts at validating the equation's efficacy in predicting resting metabolic rates have nearly unanimously found the Harris-Benedict to be a significant overestimation in the general population, but not so in obese or geriatric populations.[2,4,5,7,17]

The Cunningham equation[3] is traditionally presented as the most accurate equation for athletes due to the increased amount of lean body mass in comparison to the general population.[37] Haaf and Weijs found the Cunningham equation to hold 84.9% accuracy in male athletes (n=53) and 78.4% in female athletes (n=37).[37] However, Cunningham did not design it as such; Cunningham reanalyzed the subject data used to create the Harris-Benedict equation with the *exclusion* of 16 athlete subjects.[4] The ethnicity of study participants is not disclosed in the initial study[36], or Haaf and Weijs' application.[37]

Mifflin et al. analyzed data from 498 individuals participating in the RENO Diet-Heart Study. Demographic information is not provided beyond female (n = 247) and male (n = 251). BMIs ranged from 17$\frac{kg}{m^2}$ - 42$\frac{kg}{m^2}$; participants' percentage of ideal body weight ranged from 78% to 193%. Using previously gathered anthropometric data, i.e., height in cm, weight to the nearest 0.55kg, body fat via skin calipers, and indirect calorimetry, the researchers derived an equation that has, ostensibly, stood up to validation over the years. Resting energy expenditure measured with indirect calorimetry, weight, height, body fat percentage, waist to hips ratio, and body mass index along with measured resting energy expenditure were analyzed with stepwise multiple regression. In this sample, sex was highly correlated with fat free mass ($r = 0.83$). Although Pearson coefficients indicated fat free mass had the highest single correlation ($r = 0.80$), the researchers surmised measuring fat free mass in regular clinical practice, would become cumbersome. The correlation increased when the percentage of ideal body weight was included but the researchers did not include them in the final equation because of fears physicians may not perform the additional steps required to calculate those variables. The final presented formula, the Mifflin St. Jeor equation, accounted for 71% of the observed variability in mRMR ($R^2 = 0.71$) and is currently used today in clinical practice.[2]

There is currently no gold standard for pRMR, but, in 2003, Frankenfield et al., recommended the MSJE be considered the gold standard for calculating resting metabolic rate in obese and non-obese adults[7]. 130 volunteers submitted to indirect calorimetry which was compared to the MSJE, HB, and Owen predictive equations. MSJE predicted the highest percent of subjects' RMR ± 10%. The Mifflin – St. Jeor equation was applicable across BMIs: 70% of obese participants and 80% of non-obese subjects' pRMR were within range of mRMR.[7] However, Frankenfield's sample (n=130), contained 127 Caucasians and 3 non-Caucasians. Of the 3 non-Caucasian subjects, researchers saw no observable differences but acknowledged there were too few subjects to make a statistical determination. However, in 2005, the same researchers concluded in a subsequent systematic review that a limitation of the MSJE exists when it is applied to individuals rather than populations or certain age and ethnic groups.[9]

The Owen equation is presented in separate studies for men and women. The men's study included 60 men of Caucasian, African or Asian descent aged 18 to 82 years.[5] Forty-four women of undisclosed demographics, aged 18 – 65 years, were included in the women's study.[38] Study protocols were similar: height, weight, age, and measurements of fat free mass were collected for each participant; indirect calorimetry was performed after a 12 hour overnight fast and 30 minutes of bed rest in a quiet room before measurement.[5,38] The results of the study of women show the variable with the single highest correlation with RMR is lean body mass, in the overall sample ($r = 0.77$), even stronger in athletes ($r = 0.96$) but still striking in non – athletes in the sample (0.75). In female non – athletes, only fat free mass measured with skin calipers was more strongly correlated ($r = 0.77$).[38] The men's study yielded similarly strong correlations between both lean body mass ($r = 0.74$) and fat free mass measured with skin calipers ($r = 0.78$) and RMR.[5] In both cases, the final formula capitulated to the significance of weight in consideration of the ease of implementation in clinical practice[5] and lack of statistical difference between predictions.[38]

Owen's stated inclusion of non-Caucasian subjects in his study was unique, however, the study did not describe any differences in kilocaloric need in relation to ethnicity. A study of 114 subjects, 30% Black and 64% women, found being of African descent was associated with a 158 kcal/ day decrease in mRMR when compared with Caucasians.[17] Anthropometric data, fat mass, DEXA body scans, and mRMR were compared to the Mifflin- St. Jeor, Cunningham, and Harris-

Benedict pRMR equations. The MSJE and Harris-Benedict were significant overestimations across the board in Black subjects. The Cunningham equation, however, showed no significant deviation from mRMR. For Caucasian subjects, MSJE was once again validated, but the Cunningham and Harris-Benedict equations significantly underestimated and overestimated mRMR, respectively.

The researchers were able to quantify the role of body composition in their study sample: for every 1 kg increase in lean body mass, there was an increase of 28.2 $kcal \cdot day^{-1}$ in mRMR; for every 1 kg increase in total body fat, mRMR increased 5.9 $kcal \cdot day^{-1}$. For every centimeter increase in hip circumference, mRMR was estimated to decrease by 6.7 $kcal \cdot day^{-1}$. Differences in regional body composition, which appear to parallel differences in ethnicity, when ignored may cause significant overestimations in energy expenditure in Black Americans. The sum of this study suggests that ethnicity may need to be included as an independent variable.[17]

There have been several attempts at rectifying this apparent difference between pRMR and mRMR. The Sabounchi equations were created from the logistic regression of secondary data and include several ethnic populations.[11] The Anjos equation[10] was created for and validated in Brazilian populations, citing the inaccuracies of MSJE and FAO equations when applied to subtropical groups.[13] The University of Memphis equation was created from a discrete sample and demonstrated a high level of accuracy for predicting RMR in both African-American and European-American women.[39] However these equations are not commonly used and have not received publicity nor gained the traction of the equations uniquely accurate in Caucasian populations.

It has been suggested that the Cunningham equation, which is based on lean body mass, has the most accurate estimation of energy needs in Black American populations;[17] however, it is primarily used for predictive resting energy expenditure in trained athletes.[37] With Caucasian populations, this equation is far less accurate than the commonly used MSJE.[17] Similarly inaccurate in Caucasian populations is the Anjos equation. A study comparing measured metabolic rate in 148 (women=89, men= 59) Brazilian subjects to existing commonly used predictive resting metabolic equations and the Anjos equation demonstrated that the Anjos equation predicted within 10% of mRMR in 77.5% of women tested, and 59.3% of men; the Schofield (FAO), and Mifflin- St. Jeor equations are significant overestimations of mRMR in Brazilians.[13]

Lean body mass as a single predictor holds significance in several studies.[2,4,5,38] Calculating lean body mass requires, for its most basic approximation, a skinfold caliper, but this is often dismissed as a cumbersome, time-wasting technique in clinical practice.[2] Skinfold calipers require minimally more effort than any other anthropometric procedure and may offer increased accuracy and universality for predictive equations

Relation to Current Research

The body of research supports the correlation between certain anthropometric variables and RMR. Weight, age, height, and gender provide a context for predicting resting metabolic rates; however, other anthropometric variables like waist to hips ratio, and lean body mass (LBM), which are uncommonly used, may need to be considered in non- Caucasian populations. pRMR is useful in settings where indirect calorimetry is unavailable, but further elucidation of the caloric needs in populations other than Caucasian is necessary and any research done thusly must include subjects that are not Caucasian.

CHAPTER 3 METHODS

SAMPLE SIZE

An a priori power analysis provides the sample size, given $\alpha = 0.05$, $1 - \beta = 0.80$ and medium-large effect size, $d = 0.65$. This study's sample size should be 39 participants per group, n = 78, for between group comparison.[40] This study was able to recruit 68 eligible participants, 36 in the Black group and 32 in the Caucasian.

SUBJECTS AND DESIGN

This quasi- experimental study assigns 68 men and women aged 18 – 65 to groups based on ethnicity (Black group and Caucasian group). Ethnicity was defined by self-reports of grandparents' ethnicity: participants were assigned to the Black group if at least 3 grandparents are Black and the Caucasian group if at least 3 grandparents are Caucasian. Subjects exceeding 150 kilograms, or 1.9 meters were excluded due to limitations in DEXA capabilities. Additional exclusion criteria will include subjects reporting acute illnesses, fever >100.0 °F, or answering yes to any COVID-19 safety questions. Subjects who are pregnant or lactating, have a BMI < 18, missing limbs, those with hip replacements or external fixators. trained athletes and subjects who are unable or unwilling to fast for 12 hours, abstain from stimulant use, or vigorous activity before testing will also be excluded.

All participants were informed of the risks and benefits associated with this investigation and provided written consent to participate in accordance with Northern Illinois University's Institutional Review Board. Subjects were recruited via online social media posts and posted flyers

(Appendix B) around campus and the community. Prospective subjects were instructed to contact the researcher by phone. All testing took place on Northern Illinois University campus in Anderson Hall, Human Performance and Neuro – Muscular Laboratories.

PROCEDURE OVERVIEW

1. Complete consent form
2. Conduct intake interview
3. Weight, Height Measurements
4. Indirect calorimetry
5. Waist and Hip Measurements
6. DEXA scan
7. Breakfast

PROCEDURE

Upon arrival at the metabolic laboratory, subjects will fill out duplicate informed consent forms: one for the researcher's records and one for the participants. Temperatures for each participant were taken and recorded. Interviews were conducted to confirm abstinence from physical activity for 24 hours, fasting from stimulant consumption (i.e., caffeine or nicotine) for 24 hours, and food for 12 hours. The interview was administered by a lab assistant and will include self-reports of illness, COVID-19 safety, subject's ethnicity, subject's grandparents' ethnicities, sex, and date of last menstrual cycle (Appendix A). After the interview, participants will move on to the measurement phase of the study. All measurements were done in the absence of jewelry and shoes.

WEIGHT AND HEIGHT

Weight and height were measured with scales and stadiometers calibrated to the manufacturer suggestions. The Inbody 570 has a weight capacity of 250 kg with weight graduations of 0.1 kilograms, this weight was measured to the nearest 0.1 kg. The Health-O-Meter stadiometer, model 402EXP, measured height in centimeters, ranging from 60 cm to 213 cm in 1 mm increments. Tools were disinfected and recalibrated after each use. Subjects were asked to stand straight with weight evenly distributed on both feet, heels together with arms at the sides, and to look straight ahead. The headpiece was lowered to touch the crown of the head and lightly compress the hair.

Waist circumferences were measured by placing non-stretchable tape around the waist at the region of the navel for males and between the bottom of the rib and top of the hip bone for females. Subjects were asked to stand erect with abdomen relaxed, arms at the side, and feet together. The participants were asked to breathe normally then the measurements were taken at the end of the normal expiration. Hip circumferences were measured around the largest portion of the buttocks. All anthropometric measures were taken twice, and an average of both measures recorded. All iterations were taken by the same assistant.

DEXA

Full body scans will then be performed using Horizon DEXA System (Hologic Canada ULC, Mississauga, ON, Canada). DEXA scans were performed according to manufacturer instructions after manufacturer suggested calibration. Participants were instructed to wear comfortable clothing without metal fasteners, or a gown was provided. A negative pregnancy test, provided by the research team, was required for female participants. Once safety questions included in the DEXA software were answered, the technician administered a full body scan. The lights were dimmed and the room quieted; the only sounds were ambient and from the movement of the Horizon DEXA System. Total lean mass, fat mass, and regional lean and adipose mass were recorded. Surfaces were sanitized and dried after the administration of each test.

Participants were to lie down in the center of the table with the palms of the hands lying flat on the table or facing the hips. Body position and regions were assigned according to the following image and rubric.[41]

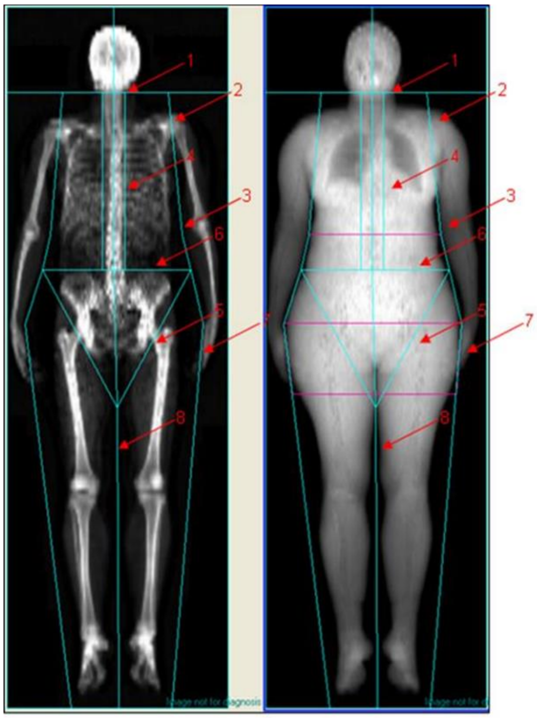


FIGURE 1: (LEFT) DEXA REGIONS AND BODY POSITIONING

1. Head region (not assessed in this study): the Head partition is located immediately below the chin at the disc between C5 and C6 on the spine.
2. Left and Right Arm region: Both arm cuts pass through the acromion process and are as close to the body as possible.
3. Left and Right Forearm position: Both forearm positions are as close to the body as possible and separate the elbows and forearms from the body.
4. Left and right spine: Spinal partitions are aligned vertically as close to the spine as possible without including the rib cage.

5. Left and right pelvis region and position: Both pelvis partitions pass through the femoral necks and do not touch the pelvis or femur.
6. Pelvis top: The pelvis top is immediately above the top of the pelvis placed between L2 and L3 on the spine.
7. Left and right leg: Both leg partitions separate the hands and forearms from the legs.
8. Centre leg: The Centre leg partition separates the right and left leg and is centered between the left and right first toes

   A partition inside the Truncal region is just below the sternum, roughly between T7 and T8. This Truncal region includes the area below the head and above the pelvis.

Indirect Calorimetry

The True One 2400 (Parvo Medics, Salt Lake City, UT, USA) was calibrated according to manufacturer specifications and the filter was replaced for each participant. Subjects were instructed to lie prone in a dimly lit, quiet, temperature-controlled room. Participants were instructed to lie for 10 minutes before indirect calorimetry began. Measurements were taken using a canopy hood for 20 minutes with the first 5 minutes of data discarded. A coefficient of variation ≤10% was required for the data set. Measurements were discarded and testing readministered[34] if a respiratory quotient of 0.93- 1.3 was recorded. An RQ in this range may indicate there is a problem with the machinery i.e., gas leak or miscalibration, or possibly that the participant is not in a fasted state. Once testing was complete all equipment was cleaned and sanitized and breakfast was provided to participants.

Data Analysis

Statistical analyses were performed using SPSS version 26.0 (SPSS Inc., Chicago, IL, USA). Data analyses included descriptive statistics, means, standard deviations, t-tests, and correlations.

Linear regression analyses and correlational relationships were also assessed at significance $\alpha \leq 0.05$. Differences between groups were identified. Fourteen pRMR equations (Table 1) were compared to mRMR for each group and the entire sample. Tables 2 and 3 outline the variables and the analysis to be performed.

Data Safety and Monitoring

Group data were analyzed without names. All identifying subject data was confidential. Each subject was assigned a random personal identification number (PIN) and all data is stored in password protected files on a private computer. Any paper documentation is kept in a locked office with locked storage and limited access.

TABLE 1: pRMR EQUATIONS

Name	pRMR equation	pRMR with LBM equation
Harris – Benedict Men[36] (kcal·day^{-1})	66.47 + 13.75(w) + 5.0 (h) – 6.75(a)	
Harris – Benedict Women[36] (kcal·day^{-1})	655.09 + 9.56(w) + 1.84(h) – 4.67(a)	
Cunningham Equation[4] (kcal·day^{-1})	500 + 22 (LBM)	
Owen Men[5] (kcal·day^{-1})	Non- Athletes	290 + 22.3(FFM)
	879 + 10.2(w)	
Owen Women[38] (kcal·day^{-1})	Non- Athletes	334 + 19.7(FFM)
	795 + 7.18(w)	
Mifflin- St. Jeor Men[2] (kcal·day^{-1})	9.99(w) + 6.25(h) – 4.92(a) + 5	19.7(FFM) +413
Mifflin- St. Jeor Women[2] (kcal·day^{-1})	9.99(w) + 6.25(h) – 4.92(a) – 161	19.7(FFM) +413
Anjos Equation Men[10] (kJ·day^{-1})	41.79(w) + 29.86 (h) +11.69 (a) -1884.93	
Anjos Equation Women[13] (kJ·day^{-1})	37.46(w) + 37.13(h) – 2.92(a) – 3407.09	
Weir Equation[33] (kcal·day^{-1})	1440 (3.94 VO_2 + 1.11 VCO_2)	
University of Memphis equation[39] (kJ·day^{-1})	616.93 - 14.9(a) + 35.12(w)+19.83 (h) - 271.88(e)	
University of Memphis[39] (kcal·day^{-1})	147.45 - 3.56(a) + 8.39(w) +4.74(h) - 64.98(e)	
Sabounchi Structure 4 Men[11] (kcal·day^{-1})	421+ 20.6(FFM) + 1.9(FM)	
Sabounchi Structure 4 Women[11] (kcal·day^{-1})	397 + 20.6(FFM) + 1.9(FM)	
Sabounchi Structure 11 Men[11] (kcal·day^{-1})	787 - 3.28(a) + 13.6(FFM) + 5.95(FM)	
Sabounchi Structure 11 Women[11] (kcal·day^{-1})	678 - 3.28(a) + 13.6(FFM) + 5.95(FM)	

TABLE 2: VARIABLE OVERVIEW

Variable	Measurement
pRMR (continuous)	Height, Weight, Age, Sex, LBM, Fat Mass Equations from Table 1
%Fat Mass	DEXA scan
Lean Body Mass	Regional DEXA scan
Fat Mass	Regional DEXA scan
mRMR	Metabolic Cart
Ethnicity	self-reported ethnicity

TABLE 3: SUMMARY OF VARIABLES AND STATISTICAL TESTS

Hypothesis	Variables	Statistical Tests
mRMR in the Black Group is less than the Caucasian Group	mRMR	Independent samples t-test
MSJE and HB equations overestimate resting energy expenditure in the Black Group	Independent Variables: mRMR, height, weight, age, sex, ethnicity, LBM, FM, WHR Dependent Variables: pRMR (equations) Dependent Variables: pRMR (equations)	Paired samples t-test
Lean Mass based equations predict RMR for Black Americans with more accuracy than weight-based equations	Independent Variables: mRMR, height, weight, age, sex, ethnicity, LBM, FM, WHR Dependent Variables: pRMR (equations)	Hierarchical linear regression, stepwise regression

CHAPTER 4 RESULTS

A total of 68 participants were included in the following analyses, including 38 females. Participants were assigned to a group by ethnicity (Caucasian, n=32; Black, n=36). The Black group (female, n = 20; male, n = 16) was comparable to the Caucasian group (female, n = 18; male, n = 14)) in all measures except Hours Slept Night Prior. The Black group slept roughly an hour less than the Caucasian group the night before testing ($p < 0.05$).

The overall sample was 25.94 ± 1.08 years of age (mean ± SE), 169.4 ± 9.07cm in stature, 83.86 ± 19.82kg with 30.3 ± 11.3% adiposity. The Black group was slightly younger, at 25.64 (8.24) years of age (mean(SD)) while the Caucasian group's age was 26.25 (9.76). The Black and Caucasian groups were of similar heights: 167.62 (8.14) cm and 171.43 (9.76) cm, respectively. Weight (kg), lean body mass (kg) fat mass (%) were also comparable (Table 2). Later results offer a glance at differences between measurements by gender.

While the groups had similar percentages of obese participants (BMI >29.9), obese Black female participants' weight was notably higher than the obese Caucasian female participants ($p > 0.05$). This statistical difference was not present in males.

TABLE 4: SAMPLE CHARACTERISTICS

Characteristics	All Subjects *Mean (SD)* n = 68	Black *Mean (SD)* n = 36	Caucasian *Mean (SD)* n = 32
Male (*n*)	30	16	14
Female (*n*)	38	20	18
Days Since Menstruation	*10.22 (8.72)*	*12.69 (10.51)*	*7.73 (1.16)*
Age (years)	*25.94 (8.93)*	*25.65 (8.24)*	*26.25 (9.76)*
Height (cm)	*169.4 (9.07)*	*167.62 (8.14)*	*171.43 (9.74)*
Weight (kg)	*83.86 (19.82)*	*86.34 (21.56)*	*81.07 (17.58)*
Fat Mass (%)	*30.03 (11.3)*	*31.22 (12.98)*	*28.68 (9.06)*
Lean Body Mass	*63.93 (11.41)*	*61.84 (14.01)*	*66.29 (7.0)*
Hours Slept Prior Night	*7.14 (1.7)*	*6.64 (1.93)***	*7.73 (1.17)***

** $p < 0.05$

REGIONAL LEAN MASS DISTRIBUTION BY ETHNICITY

The sample overall had 63.94% ± 11.41 lean mass. The two groups were comparable in this regard, although the Caucasian group was slightly higher. However, total lean mass measurements were nearly equal; the Black group had 53.37kg (11.24) while the Caucasian group had 52.20kg (13.73) lean mass (Table 3).

Groups differed in 3 regions of measured Lean Mass; Legs, Trunk-to-Total Body Mass ratio, and Trunk-to-Total-Lean Mass ratio. The Black group had a slightly greater lean mass in the legs compared to the Caucasian group. Both truncal ratios were greater in Caucasians ($p < 0.05$). Caucasian's truncal lean mass was greater overall but did not rise to the level of significance.

TABLE 5: REGIONAL LEAN MASS DISTRIBUTION

Region	All Subjects *Mean (SD)* n = 68	Black *Mean (SD)* n = 36	Caucasian *Mean (SD)* n = 32
Total Lean Mass (kg)	52.82 (12.40)	53.37 (11.24)	52.20 (13.73)
Percent LBM (%)	63.94 (11.41)	61.84 (14.00)	66.30 (6.99)
Trunk (kg)	25.39 (5.72)	24.82 (5.07)	26.04 (6.39)
Arms (kg)	5.79 (2.01)	5.99 (1.86)	5.57 (2.16)
Legs (kg)	18.19 (4.76)	19.09 (4.39)*	17.17 (5.01)*
Trunk: Lean Mass Ratio	0.30 (0.041)	0.46 (0.016)**	0.50 (0.176)**
Trunk: Total Mass Ratio	0.48 (0.245)	0.29 (0.042)**	0.32 (0.034)**

REGIONAL ADIPOSITY BY ETHNICITY

Groups had similar distributions of adiposity. The sample's mean percent body fat was 31.73 ± 9.24. The Black group had slightly higher percent body fat overall. The group's Android Gynoid ratio was slightly lower than the Caucasian group indicating a trend towards larger hips. The ratio of truncal fat mass to limb fat mass and legs fat mass were both larger in the Black group as well, indicating that more fat mass is stored in the limbs of the Black group than the Caucasian. Yet, overall truncal fat mass is also greater in the Black group. None of these differences rose to the level of being statistically significantly different.

TABLE 6: REGIONAL ADIPOSITY

Region	All Subjects *Mean (SD)* n = 68	Black *Mean (SD)* n = 36	Caucasian *Mean (SD)* n = 32
Total Fat Mass (kg)	27.31 (13.25)	28.36 (14.15)	26.13 (12.27)
Percent Fat Mass (%)	31.73 (9.24)	32.04 (10.43)	31.38 (7.83)
Trunk (kg)	12.34 (6.51)	12.95 (7.71)	11.65 (4.86)
Arms (kg)	3.16 (2.02)	3.30 (2.27)	2.99 (1.71)
Legs (kg)	10.29 (4.55)	11.12 (5.32)	9.35 (3.31)
Estimated VAT Mass (kg)	0.44 (0.218)	0.43 (0.231)	0.45 (0.207)
Android Gynoid Ratio	0.94 (0.167)	0.94 (0.180)	0.95 (0.153)
Truncal FM Limb FM Ratio	0.93 (0.216)	0.90 (0.24)	0.97 (0.19)
Trunk/Legs Fat Mass Ratio	0.93 (0.216)	0.91 (0.168)	0.89 (0.125)

CORRELATIONS BETWEEN MRMR AND ANTHROPOMETRIC MEASURES

The Caucasian group demonstrated a modest positive correlation between age and mRMR (0.543, $p < 0.05$) in contrast to the Black group (- 0.351, $p < 0.05$). Weight and sex (male dummy coded as "1") had a slightly higher positive correlation with mRMR (0.754 and 0.557, $p < 0.01$) in the Caucasian group than in the Black group (0.744 and 0.455, $p < 0.01$). The Caucasian group also had a stronger correlation between mRMR, leg lean mass and arm lean mass(0.841 vs 0.814, $p < 0.01$ and 0.817 vs. 0.779, $p < 0.01$); however, Black participants had a stronger correlation with truncal fat mass (0.438, $p < 0.05$ vs 0.185), truncal lean mass (0.826 vs 0.801, $p < 0.01$), and android gynoid ratio (0.503, $p < 0.01$ vs. 0.367, $p < 0.05$). Table 3 offers side by side comparisons for each group and the overall sample.

TABLE 7: CORRELATIONS BETWEEN MRMR AND ANTHROPOMETRIC MEASURES

Measure	All Subjects	Black	Caucasian
Age (years)	*-0.213*	- 0.351**	*-0.114*
Height (cm)	*0.633****	0.516**	*0.723****
Sex	*0.502****	0.455***	*0.557****
Weight	*0.714****	0.744***	*0.754****
Total Fat Mass	*0.256***	*0.421***	*0.109*
Fat Mass (%)	*-0.162*	*0.058*	*- 0.427***
Arms Fat Mass	*0.283***	*0.299*	*0.304*
Legs Fat Mass	*0.119*	*0.22*	*0.029*
Trunk Fat Mass	*0.308***	*0.438***	*0.185*
Android Gynoid Ratio	*0.436****	*0.503****	*0.367***
Total Lean Mass	*0.829****	*0.844****	*0.831****
Arms Lean Mass	*0.786****	*0.779****	*0.817****
Legs Lean Mass	*0.794****	*0.814****	0.841***
Trunk Lean Mass	*0.813****	*0.826****	*0.801****
Trunk: Lean Mass Ratio	*0.109*	*-0.08*	*0.275*
Trunk: Total Mass Ratio	-0.23	*-0.204*	*- 0.606****
Hours Slept Prior Night	*0.164*	*0.103*	*0.223*
Days Since Menstruation	*0.112*	*-0.112*	*0.446*

*** p <0.01
**p <0.05

MRMR AND PRMR BY ETHNICITY

The sample had a mean mRMR of 1727.78 (328.09) kcal/day (Table 6). The Black group had a lower mean mRMR than the Caucasian group but the difference was minimal and not statistically significant (p >0.05). When comparing Black males to Caucasian males or Black females to Caucasian females the differences between them did not rise to the level of statistical significance.

Overall, males had a greater measured resting metabolic than females both between and within groups ($p < 0.01$).

TABLE 8: MRMR BY ETHNICITY AND DISTINGUISHED BY SEX

Group	mRMR
Total Sample (n = 68)	1727.78 (328.09)
Black (n = 36)	1699.78 (310.54)
Caucasian (n = 32)	1756.65 (347.83)
Black Male (n = 16)	1858.42 (291.37)
Caucasian Male (n = 14)	1972.87 (347.56)
Black Female (n = 20)	1582.90 (275.93)
Caucasian Female (n = 18)	1588.04 (244.55)

VALIDATION OF EQUATIONS BY ETHNICITY

All equations (Table 1) were correlated ($p <0.01$) with mRMR for each group and the overall sample (Table 7). Pearson correlations for the Black group were stronger than the Caucasian group for nearly all of the equations. The strongest correlations in the sample were seen between Black females, the MSJE, and Harris – Benedict Equations. Caucasian males overall had weaker correlations with each of the equations.

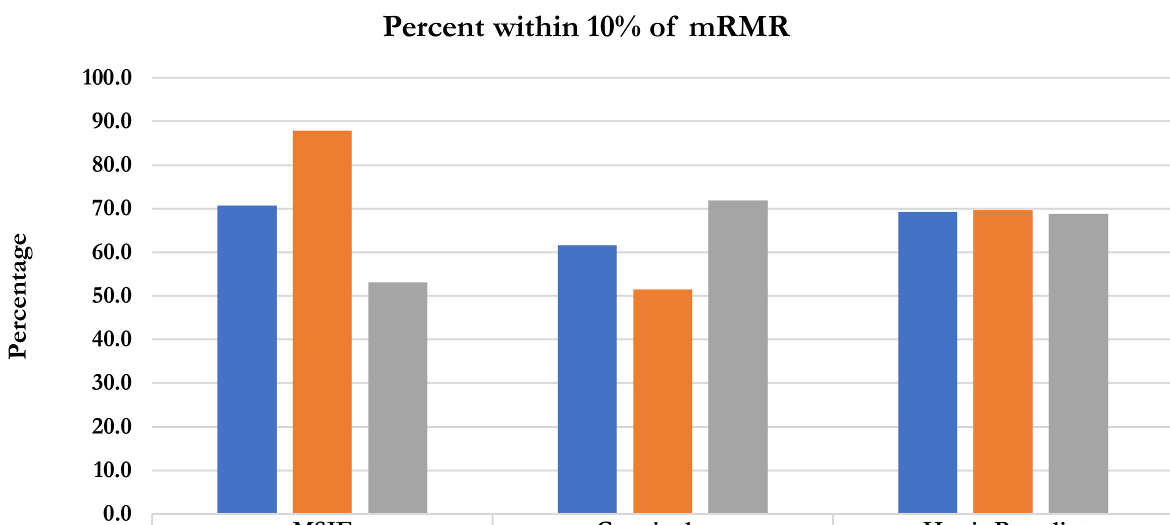
Paired samples t-tests were run with mRMR and each of the equations from table 1. The table below (Table 8) shows which pRMR equations were significantly different from the mean of the group mRMR. An asterisk denotes a statistically significant difference between the mean values. The Mifflin – St. Jeor Equation was statistically significantly different for the entire sample but, surprisingly demonstrated similarity in the Black group overall ($p > 0.05$); the means were nearly the same. In the Caucasian group, the MSJE's predicted metabolic rate was statistically significantly different. When analyzed by ethnicity and gender, the MSJE was different from the mean mRMR of females, not males, of the Caucasian group. The correlations between MSJE and the Caucasian group were weaker than the Black group. The Harris – Benedict equation was similarly exclusive to the mRMR of Caucasian groups, both males and females.

TABLE 9: CORRELATION OF MRMR AND PRMR BY ETHNICITY AND SEX

Measurement	Sample (n = 68)	Black (n = 36)	Black Male (n = 16)	Black Female (n = 20)	Caucasian (n = 32)	Caucasian Male (n = 14)	Caucasian Female (n = 18)
MSJE	0.828	0.874	0.736	0.921	0.809	0.643	0.806
MSJE - FFM	0.829	0.844	0.746	0.860	0.831	0.701	0.847
Cunningham	0.829	0.844	0.746	0.860	0.831	0.701	0.847
Owen	0.656	0.583	0.751	0.045	0.719	0.637	0.847
Owen - FFM	0.821	0.836	0.736	0.860	0.819	0.701	0.481
Anjos	0.824	0.856	0.712	0.899	0.808	0.712	0.741
U Memphis	0.800	0.810	0.734	0.923	0.797	0.646	0.805
Sabounchi Formula 11	0.739	0.870	0.732	0.873	0.682	0.447	0.850
Sabounchi Formula 4	0.828	0.851	0.725	0.873	0.822	0.673	0.873
Harris - Benedict	0.821	0.867	0.733	0.929	0.799	0.624	0.818

Analysis of the sample overall revealed only one equation was not statistically significantly different from the mRMR, the Cunningham Equation (Table 8). The means of the Harris – Benedict equation performed better than expected ($0.05 > p < 0.10$) when compared to the measured resting metabolic rate for the overall sample but means were statistically significantly different for the Black group. The remainder of the equations were different from mRMR ($p < 0.05$).

Figure 2 denotes the percent of pRMR calculations that fell within 10% of mRMR. The MSJE demonstrates 70.8% of values within 10% of mRMR for the total sample. Notably, the MSJE predicted 87.9% of the Black sample's mRMR, performing much better than in the Caucasian group. Even so, these values replicate those predicted in the original study: Mifflin et. al reported in their study that the equation accounted for 71% of mRMR variability in the overall sample.[2] Individual variations in mRMR for the Caucasian group, i.e. pRMR within 10% of mRMR, were alternatively identified 71.9% of the time by the Cunningham equation.



	MSJE	Cunningham	Harris Benedict
Sample	70.8	61.5	69.2
Black	87.9	51.5	69.7
Caucasian	53.1	71.9	68.8

Figure 2: Percentage of pRMR estimations within 10% of mRMR

Table 10:mRMR compared to pRMR estimations (kcals/day) Mean(SD)

Equation	Sample (n = 68)	Black (n = 36)	Caucasian (n = 32)	Black Male (n = 16)	Black Female (n = 20)	Caucasian Male (n = 14)	Caucasian Female (n = 18)
mRMR	1727.78 (328.09)	1699.78 (310.54)	1756.65 (347.83)	1858.42 (291.37)	1582.90 (275.93)	1972.87 (347.56)	1588.48 (244.55)
MSJE	1681.41 (280.03)**	**1698.47** **(280.91)**	1663.81 (282.50)**	**1838.36** **(235.40)**	**1595.40** **(271.71)**	**1896.10** **(193.73)**	1483.15 (194.52)**
MSJE - FFM	1546.04 (250.84)**	1540.48 (206.29)**	1551.78 (293.11)**	1705.11 (147.60)*	1419.17 (152.68)**	1793.96 (259.89)**	1363.42 (138.33)**
Cunningham	**1743.92** **(275.04)**	**1649.07** **(246.36)**	**1750.21** **(321.37)**	**1918.32** **(161.84)**	**1604.81** **(167.40)**	**2015.76** **(284.96)**	**1542.67** **(151.87)**
Owen	1540.88 (263.12)**	1534.73 (256.52)**	1547.03 (273.53)**	1770.51 (213.40)*	1373.40 (123.85)**	1793.95 (160.08)**	1198.16 (138.51)**
Owen - FFM	1418.49 (297.75)**	1534.73 (256.52)**	1414.68 (327.47)**	1643.21 (206.23)**	1271.14 (194.53)**	1693.06 (286.81)**	1284.41 (138.33)**
Anjos	1524.05 (295.39)**	1531.84 (284.90)**	1516.00 (310.19)**	1786.70 (206.66)*	1391.80 (241.71)**	1786.69 (206.66)**	1305.47 (188.47)**
U Memphis	1530.28 (198.46)**	1514.30 (211.18)**	1546.77 (186.35)**	1658.82 (159.46)**	1489.54 (224.75)**	1658.81 (159.46)**	1459.63 (159.68)**
Sabounchi Formula 11	1543.16 (303.09)**	1536.65 (231.16)**	1549.87 (367.89)**	1788.50 (425.33)*	1445.82 (220.69)**	**1788.50** **(425.33)**	1364.27 (158.39)**
Sabounchi Formula 4	1551.37 (285.11)**	1556.83 (252.81)**	1545.74 (319.88)**	1789.21 (310.74)*	1443.13 (227.63)**	1789.21 (310.74)**	1356.38 (158.06)**
Harris - Benedict	1772.78 (328.09)*	1790.79 (312.17)**	**1754.65** **(347.84)**	2027.23 (241.68)*	1660.93 (249.90)**	**2027.23** **(241.68)**	**1542.03** **(168.00)**

*** p <0.01, **p<0.05

Bolded similarities to mRMR

REGRESSION MODELING

A hierarchical multiple regression analysis was run to determine if prediction of mRMR improved with the addition of Ethnicity to variables typically included in equations, i.e., Weight, Height, Age, and Sex. Ethnicity was added to the hierarchy to assess the change in variability that may be attributed to ethnicity. The following tables detail each regression model. Models in Table 9 and Table 11 are presented in pairs. Model 1A in Table 9 includes only weight in the regression analysis; the addition of ethnicity in Model 1B causes a statistically significant $\Delta R^2 = 0.037$, and $\Delta F = 5.045$ ($p < 0.05$). Including ethnicity as an additional predictor to weight and sex in Model 3 also resulted in a significant change in R^2: $\Delta R^2 = 0.031$ ($p < 0.05$). Adding ethnicity to weight and height in Model 2B did not result in a ΔR^2 that rose to the level of statistical significance.

TABLE 11: PREDICTING MRMR FROM WEIGHT, ETHNICITY - HIERARCHICAL REGRESSION

Model	R	R^2	ΔR^2	F Change	p - value
1A	.714[a]	0.510	0.510	65.482	0.000
1B	**.739[b]**	**0.547**	**0.037**	**5.045**	**0.028**
2A	.803[c]	0.644	0.135	23.438	0.000
2B	**.807[d]**	**0.651**	**0.007**	**1.163**	**0.285**
3A	.805[e]	0.649	0.139	24.506	0.000
3B	**.824[f]**	**0.680**	**0.031**	**5.939**	**0.018**

a. Predictors: (Constant),Weight (kg)
b. Predictors: (Constant), Weight (kg), Ethnicity
c. Predictors: (Constant),Weight (kg), Height (cm)
d. Predictors: (Constant), Weight (kg), Height (cm), Ethnicity
e. Predictors: (Constant),Weight (kg), Sex
f. Predictors: (Constant), Weight (kg), Sex, Ethnicity

Table 12 outlines the ΔR^2 int the regression model when adding Sex, Age, and ethnicity to Weight and Height. In Model 4, the addition of Age and Ethnicity to Weight and Height did not result in any meaningful ΔR^2. However, the addition of ethnicity to Weight, Height, Age in Model 5, and Weight, Height, Age, Sex in Model 6 resulted in $\Delta R^2 = 0.014$ and $\Delta R^2 = 0.016$, respectively ($p < 0.10$). Sorting the data set by sex (Table 12) revealed that the addition of Ethnicity to the Weight, Height, Age model was significant for females (n=38) but not for males (n=30). This model shows that for males in this sample, adding variables beyond weight and height were statistically inconsequential.

When using Lean Mass as the base variable in the hierarchy (Table 13), neither Height, Age, Sex, nor Ethnicity contributed meaningfully to ΔR^2; the effects of Ethnicity were nearly non-existent when added to Lean Mass in Model 2. In fact, no other variable led to a significant ΔR^2 when added to Lean Mass.

TABLE 12: PREDICTING MRMR FROM WEIGHT, HEIGHT, SEX, AGE, ETHNICITY - HIERARCHICAL REGRESSION

Model	R	R^2	ΔR^2	F Change	p - value
4A	.810[a]	0.656	0.012	2.183	0.145
4B	**.815[b]**	**0.665**	**0.008**	**1.464**	**0.231**
5A	.826[c]	0.682	0.038	7.321	0.009
5B	**.835[d]**	**0.696**	**0.014**	**2.806**	**0.099**
6A	.831[e]	0.691	0.009	1.700	0.197
6B	**.841[f]**	**0.707**	**0.016**	**3.154**	**0.081**

a. Predictors: (Constant),Weight (kg), Height (cm), Age
b. Predictors: (Constant),Weight (kg), Height (cm), Age, Ethnicity
c. Predictors: (Constant),Weight (kg), Height (cm), Sex
d. Predictors: (Constant), Weight (kg), Height (cm), Sex, Ethnicity
e. Predictors: (Constant),Weight (kg), Height (cm), Sex, Age
f. Predictors: (Constant),Weight (kg), Height (cm), Sex, Age, Ethnicity

TABLE 13: WEIGHT, HEIGHT, AGE, ETHNICITY BY SEX - HIERARCHICAL REGRESSION

Sex	Model	R	R^2	ΔR^2	F Change	p - value
Female	1	.807[a]	0.652	0.652	65.479	0.000
(n = 38)	2	.817[b]	0.667	0.015	1.554	0.221
	3	.848[c]	0.719	0.052	6.085	0.019
	4	.882[d]	0.778	0.059	8.488	0.006
Male	1	.660[a]	0.436	0.436	20.081	0.000
(n = 30)	2	.728[b]	0.530	0.095	5.036	0.034
	3	.740[c]	0.547	0.017	0.884	0.356
	4	.740[d]	0.547	0.000	0.000	0.997

a. Predictors: (Constant),Weight (kg)
b. Predictors: (Constant),Weight (kg), Height (cm)
c. Predictors: (Constant),Weight (kg), Height (cm), Age
d. Predictors: (Constant), Weight (kg), Height (cm), Age, Ethnicity

Stepwise regression revealed 3 predictive variables for this sample: Weight, Age, and Hrs slept the night before (Table 13). A hierarchical regression analysis was run using the same variables (Table 14) with the addition of ethnicity. Similar to other models, the addition of Ethnicity resulted in a significant ΔR^2, 0.033 ($p < 0.05$). Notably, the reported R in the final model of the stepwise regression is lower than R in the hierarchical regression using the same variables.

TABLE 14: PREDICTING MRMR FROM LEAN MASS, HEIGHT, AGE, SEX AND ETHNICITY - HIERARCHICAL REGRESSION

Model	R	R^2	ΔR^2	F Change	p - value
1	.804[a]	0.646	.647[a]	115.285	0.000
2	**.816[b]**	**0.666**	**0.006**	**1.049**	**0.310**
3	.804[c]	0.646	.000[b]	0.010	0.922
4	.808[d]	0.652	.006[c]	1.091	0.300
5	.815[e]	0.663	.011[d]	1.894	0.174
6	**.818[f]**	**0.669**	**.006[e]**	**1.073**	**0.304**

a. Predictors: (Constant), Lean Mass
b Predictors: (Constant), Lean Mass, Ethnicity
c. Predictors: (Constant), Lean Mass, Height (cm)
d. Predictors: (Constant), Lean Mass, Height (cm), Age
e. Predictors: (Constant), Lean Mass, Height (cm), Age, Sex
f. Predictors: (Constant), Lean Mass, Height (cm), Age, Sex, Ethnicity

TABLE 15: STEPWISE REGRESSION OF DATA SET

Model Summary

Model	R	R^2	ΔR^2	F Change	p - value
1	.834[a]	0.695	0.695	66.125	0.000
2	.861[b]	0.741	0.046	4.975	0.034
3	.885[c]	0.782	0.041	5.134	0.032

a. Predictors: (Constant), Weight (kg)
b. Predictors: (Constant), Weight (kg), Age
c. Predictors: (Constant), Weight (kg), Age, Hrs Slept Night Before

TABLE 16: STEPWISE VARIABLES, ETHNICITY ADDED

Model Summary

Model	R	R^2	ΔR^2	F Change	p - value
1	.734[a]	0.538	0.538	23.318	0.000
2	.756[b]	0.571	0.033	4.485	0.038

a. Predictors: (Constant), Hrs Slept Night Before, Age, Weight (kg)
b. Predictors: (Constant), Hrs Slept Night Before, Age, Weight (kg), Ethnicity

Two separate, new equations were created using this data set by regression analysis. The variables used are Weight and Lean Mass, which may have a better application in the general population. The variable WT is weight, in kilograms; LBM is lean mass, given in kilograms. Equations 1 and 2 follow:

$$(5 * WT) + (15.3 * LBM) + 436 = RMR$$

EQUATION 1: BANNISTER EQUATION FORM 1

$$(5 * WT) + (17 * LBM) + 279 = RMR$$

EQUATION 2: BANNISTER EQUATION FORM 2

These variables were statistically significant in the entire sample, both groups and the informal subgroups ($p < 0.01$). Both Forms were statistically similar to each group, subgroup, and the entire sample ($p > 0.05$). Table 15 outlines the similarities between the Bannister Form pRMR equations and mRMR.

TABLE 17: *BANNISTER FORMS* VS MRMR

Measurement	Sample (n = 68)	Black (n = 36)	Black Male (n = 16)	Black Female (n = 20)	Caucasian (n = 32)	Caucasian Male (n = 14)	Caucasian Female (n = 18)
mRMR	1727.78 (328.09)	1699.78 (310.54)	1858.42 (291.37)	1582.90 (275.93)	1756.65 (347.83)	1972.87 (347.56)	1588.48 (244.55)
Bannister Form 1	1736.03 (274.91)	1745.94 (247.66)	1875.13 (209.76)	1650.75 (233.92)	1725.80 (304.13)	1957.03 (277.52)	1545.94 (177.47)
Bannister Form 2	1676.80 (295.84)	1686.23 (264.43)	1829.64 (222.22)	1580.57 (246.70)	1667.07 (329.11)	1919.20 (299.87)	1470.96 (188.89)

In comparison to the individual performance of the commonly used equations, the Bannister Forms perform better consistently across all groups and subgroups. Bannister form one matched the MSJE in predicting the entire sample without the previously demonstrated statistical difference. Bannister Form 2 performed better in the overall sample than any of the other equations. When stratified by ethnicity, Bannister Form 1 predicted, within 10%, 71.8% of the Caucasian sample's mRMR. Bannister Form 2 predicted 84.8% of the Black sample's mRMR (Figure 3).

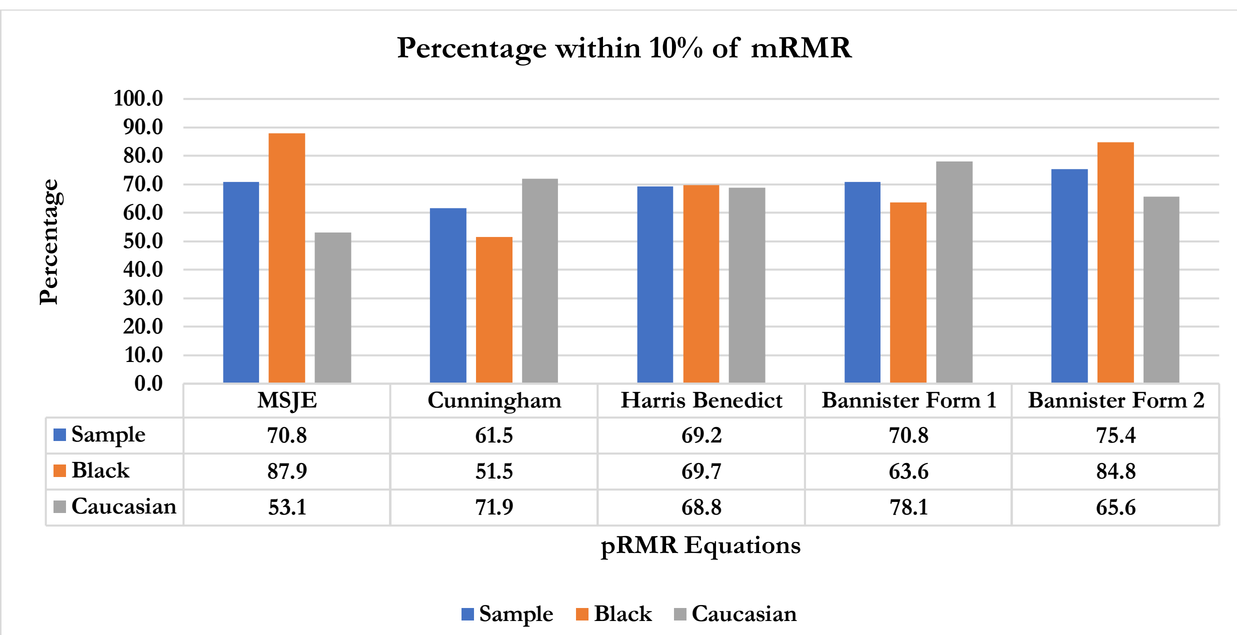


FIGURE 3: COMPARISON OF BANNISTER FORMS TO COMMONLY USED PRMR EQUATIONS

CHAPTER 5 DISCUSSION

Body composition between the two groups is largely the same, but the value of each region may need to be considered differently. Mean Total fat mass in each group was not statistically different but may be practically different. The Black group had a slightly higher total fat mass and while fat is not typically a large contributor mRMR, only about 5 kcals/kg,[27] a larger proportion of it may skew estimations by pRMR. Mean truncal lean mass in the Black group was slightly less than the Caucasian group. Similar to total fat mass, the difference was not statistically significant. However, the trunk is where the highly metabolically reactive organs are located. These organs contribute the most to mRMR and small differences in their mass can lead to a large difference in energy expenditure.[29] The ratio of truncal lean to total lean mass, and even total mass overall, is notably, statistically significant between groups ($p < 0.005$). The ratio is smaller in the Black group, meaning that truncal lean mass is a smaller portion of total mass, and total lean mass, in the Black group (table 3).

The correlations between anthropometric measures reveal some differences between groups. Age was negatively correlated with mRMR ($p<0.05$) in the Black Group while the Caucasian Group neared almost no relationship between age and mRMR. It is well established that mRMR decreases with age,[2,36] but this data suggests that the strength of this relationship may vary by ethnicity. In the Caucasian group, %Fat Mass was strongly negatively correlated with mRMR ($p<0.05$) whereas the Black group showed no relationship whatsoever. In the Black Group, Total Fat Mass (kg) showed a significant positive correlation ($p<0.05$) with mRMR where the relationship was nearly non-existent in the Caucasian Group. It may be due to the differences in fat mass. Although the mean Total Fat Mass and mean %Fat Mass were not significantly different between groups, the Black group had a larger mean and larger standard deviation for both variables. Fat contributes a relatively small amount of kcal/kg/day. This interplay possibly indicates that fat mass may need to be considered after some certain percent Fat Mass.

Leg lean mass is also notably different between groups. The mean Leg lean mass for the Black group is 19.09kg (4.39) and less than the Caucasian group's leg lean mass of 17.17 ± 5.01kg ($p < 0.05$). Presumably, lean mass in the leg includes predominantly skeletal muscle. DEXA scans differentiate between bone and tissue mass for each region; thus, bone density is not a consideration. Skeletal muscle contributes about 13 kcals/kg which is much lower than the HMRO found in the trunk. The Black group has more lean mass in the legs, but it is not contributing as much as the truncal lean mass would. It follows that the Black group would have a lower mRMR and might perhaps require an estimation that accounts for these differences in body composition.

The Black group's mean mRMR is less than the Caucasian group's but these differences did not rise to statistical significance (Table 6). In this study, gender revealed a different story. Black females and Caucasian females of each sample had very similar mean mRMRs, 1582.90 ± 275.93 kcals/day and 1588.04 ± 244.55 kcals/day, respectively. Males displayed a larger difference; Black males' mean mRMR was 1873.17 ± 297.78 kcals/day while Caucasian males' was 1972.87 ± 347.56 kcals/day. A larger sample size might provide more clarity in the difference and its magnitude, as the relevant literature suggests the differences between these two groups is significant and consistent across genders.[15] Again, the small group size may be the cause for such a deviation from the current literature.

Surprisingly, and in opposition to aforementioned body of research, the Mifflin – St. Jeor equation was not statistically different from the Black group but was from the Caucasian group. The MSJE performed shockingly well in the Black group, i.e. 87.9% of the predictions were within 10% of mRMR. Predicting 87.9% of Black participant's mRMR ± 10% exceeds even the original author's findings for the general population. For the Caucasian group, only ~53% of predictions accounted for individual variability in mRMR. However, the individual mRMR for the entire sample was predicted within 10%, 70.8% of the time. Not only does this replicate nearly the exact reported results of the MSJE, it begs the question of the demographic of the original authors sample, which was not reported.[2] In other validation studies, pRMR in Caucasian participants displayed much greater accuracy.[7,14,29]

The Harris-Benedict equation appears to have performed well in the Black group when considering percentage of mRMR, better than the Cunningham equation. However, when taken as a population measure, the statistical significance of the difference between the group average and the pRMR estimation cannot be ignored. Thus, the Harris – Benedict equation was predictive for the Caucasian group but not so for the Black group. This demonstrates that ethnicity may still play a role but concluding the directionality and strength of that relationship is unclear in this study due to the small sample size. The Cunningham equation came forward as a front runner for the entire sample and this was further supported by the hierarchical regression in which the weight of ethnicity all but disappeared when lean body mass was the predictive variable. It appears that for this sample, Lean Mass is a better predictor overall, rather than preferable for a specific ethnicity.

The Cunningham equation includes only Lean Mass as a predictive variable. Hierarchical regression revealed $R=0.804$ ($p< 0.05$) for Lean Mass, the highest R for any variable tested with the sample. It should be noted that R in the stepwise regression in table 13 is much higher than the regression run with the same variables in table 14. This is often the case with stepwise regression and a typical drawback of using this method of modeling.[42] Additionally, the small sample size may not fully represent variability that should be attributed to ethnicity in this sample. Increasing the number of variables decreases the degrees of freedom and reduces the power of the analysis. The hierarchical regressions with several variables, like Model 4 in Males on Table 11, would provide a clearer picture in a larger sample.

This study produced 2 new equations based on weight and lean body mass. The Bannister Forms (Equation 1 and Equation 2) performed better in the sample and groups than did any of the other equations. The other equations showed significant differences between either the sample or the groups indicating that the magnitude of the percent difference between pRMR and mRMR were large for certain individuals or groups. The Bannister Forms mitigated that difference in a large regard. Although the Bannister Forms had variability in the accuracy of prediction when stratified by ethnicity, this variability upon stratification is less than the variability found in the commonly used equations (Figure 3). The Bannister Form 1 equation had high accuracy when predicting individual mRMR in the Caucasian as did Bannister Form 2 in the Black group. However, in each equation, the accuracy of the other groups out performed the other equations in many cases. These equations may have a place in predicting resting metabolic for the general population but require further validation.

The sample in the study was nearly 53% Black. Black people are largely unrepresented in research on campus and the larger body of literature. Although the results of the study do not support all the hypotheses, the value of the sample remains. The very real population of Black, Indigenous, and other people of color should be included in any study that strives to be generalizable. A one-size-fits-White approach ignores, not only Black Americans but any indigenous or other people of color receiving interventions designed for Caucasian populations.

Strengths

The overall sample provides a data set for comparison and assessment of each pRMR equation. The inclusion of a sizeable sample of non-Caucasians lends credibility and a step towards generalizability and further provides a needed step towards inclusivity. The protocol is reproducible and can be built upon with additional groups and participants. This study provides a solid foundation for additional comparisons between genders and additional ethnicities. The current data set provides a foundation that can be built upon with additional participants and additional group members.

This study also allowed the development of 2 pRMR equations. The newly developed Bannister Forms were created with a sample of defined Caucasian and Black participants. Future validation of these equations may yield results that are generalizable to the population.

Limitations

This study's results differ from the current literature regarding pRMR equations accuracy in black subjects. The aforementioned differences in gender/ethnicity-specific measurements in mRMR or the surprising differences in pRMR and mRMR for Caucasians may be directly related to small group size. The differences in mRMR between Black females and Caucasian females are especially of notice. Several Black females had higher BMIs, % body fat, and body weight than the general population. In a larger sample, these participants would likely have become outliers. In such a small sample of Black females, the BMI becomes a more substantial indicator of possible skew to the data. The other subgroups did not display such a deviation from the literature.

Another limitation is the difference in hours slept the night prior. While the hours of sleep required are highly idiosyncratic, sleep deprivation can lead to a reduction in mRMR. Although a previous study demonstrated that regardless of the being sleep deprived or well rested, the difference between groups persisted,[14] future research would need a large enough sample size to allow for control of this variable.

CONCLUSION

Neither the sample's mRMR nor any group's mean mRMR was statistically significantly different from the Cunningham equation. Estimating energy needs using Lean Mass could benefit the population overall. The MSJE demonstrated great accuracy with the black group but limited predictive ability with the Caucasian group suggesting ethnicity appears to play a role in the predictive power of regression models using commonly accepted predictive variables. The ethnicity effect is mitigated by lean mass, but equations using weight, height, and sex may benefit from the inclusion of ethnicity. In lieu of including factors for ethnicity, the Bannister Forms may offer a genderless alternative.

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APPENDIX A: INTAKE SCREENING QUESTIONNAIRE

Intake Questionnaire

Masks were worn at all times until instructed to remove.

Date:

Name:

Age:

Sex:

Ethnicity:

Maternal grandparents' ethnicity:

Paternal grandparents' ethnicity:

Have you fasted for 12 hours? Y N

Have you used nicotine, caffeine, or any other stimulant in the past 12 hours? Y N

Have engaged in any physical activity more strenuous than walking in the past 24 hours? Y N

Are you pregnant or nursing? Y N

Have you undergone a hip replacement or any other plates, pins, or external fixators? Y N

Have you undergone an amputation of any limb? Y N

Have you undergone surgery in the past 6 months? Y

Days since last menstruation?

Current medications and/or supplements?

Current medical diagnoses?

How many hours of sleep last night? How many hours of sleep the night before?

` **1Q**

Masks were worn at all times until instructed to remove.

Have you received a diagnosis of COVID-19 in the past 14 days? Y N

Have you had close contact with someone with a lab-confirmed COVID-19 diagnosis in the past 14 days? Y N

Have you experienced any of the following COVID-19 symptoms: Y N

Shortness of breath or difficulty breathing?
Repeated shaking with chills?
Muscle pain?
Sore throat?

Loss of taste or smell?
Fever (temperature of 100 or greater)?

Appendix B: Sample Recruitment Flyer