RELATIVE CONTRIBUTIONS OF ENERGY EXPENDITURE ON PHYSICAL ACTIVITY, BODY COMPOSITION AND WEIGHT GAIN TO THE EVOLUTION OF IMPAIRED GLUCOSE TOLERANCE TO FRANK DIABETES

[1]TERRENCE FORRESTER, [1]RAINFORD WILKS, [2]FAROOK JAHOOR, [3]ADEBOWALE ADEYEMO

[1]Tropical Metabolism Research Unit, University of the West Indies, Mona, Kingston 7, Jamaica
[2]Children's Nutrition Research Center, Baylor College of Medicine, 1100 Bates Street, Houston, Texas, USA
[3]Institute of Child Health, University of Ibadan, Ibadan, Nigeria

Abstract

In modern technological societies the requirement for physical work is diminished and access to food is unrestricted. Under these circumstances a large proportion of the population will gain weight and develop obesity and diabetes. At the individual level, genetic and behavioural factors must combine to lead to an imbalance between energy intake and its expenditure. Weight gain, especially rapid weight gain in a population appears to increase the risk of diabetes sharply. Thus understanding the route to weight gain and obesity, and the modulatory effects of physical activity on development of glucose intolerance is critical to credible intervention strategies to reverse or prevent diabetes in populations especially those in transitional societies. In this proposal we will examine the quantitative importance of non-resting energy expenditure (EE) in populations with rising levels of obesity and high prevalence of diabetes.

1. SCIENTIFIC BACKGROUND AND SCOPE OF THE PROJECT

The migration of an estimated 10 million people out of West Africa some 400 years ago has led to the establishment of stable populations living today in widely contrasting environments. Ancestral populations in Nigeria and Cameroon, living in subsistence agricultural environments, contrast sharply with populations in Jamaica where the nutrition and epidemiological transitions have been accelerating for the past 3 decades. African-origin populations in the United States currently live under the most Westernised conditions. This economic and technological gradient is associated with a steep gradient in the dose of risk factors for hypertension and diabetes, the most powerful determinants being obesity and decreased physical activity [1]. Indeed, the observed prevalence of hypertension and diabetes rise steeply across the diaspora: hypertension prevalence in West Africa is approximately 15%; it is 25% in the Caribbean and 34% in the US. For diabetes, the corresponding prevalence rates are, 1%, 12% and 16%.

To some extent it is intuitively obvious that levels of physical activity have declined with each step of the migration, form rural to urban West Africa, to the Caribbean, and ultimately to the United States. Limited measurements of energy expenditure have been performed among peoples of the African diaspora and none in relation to the risk of diabetes. Our own pilot data indicate that resting metabolic rate (RMR) is constant in ancestral and migrant populations, and that energy expenditure on physical activity declines as adiposity increases. However, the relationship of these changes to glucose intolerance is unknown.

Obesity appears to be a determinant of glucose intolerance in its own right, but it also represents a summary statement of energy balance at any point in time. The effects of obesity on glucose intolerance therefore is a mixture of the impact of body composition on glucose-insulin relationships as well as the modulation of this metabolism by physical activity. Adding to this complexity is the variation introduced by rate of appearance of obesity. The assessment of obesity cross-sectionally conceals the rates at which the changes in body composition occurred, and there is good evidence to show that the risk of diabetes in transitional populations and societies is directly related to the rate of anthropometric change. Rate of change is perhaps more strongly predictive of diabetes risk than is absolute levels of obesity.

In this proposal, we seek to measure the energy expenditure on activity, the rate of weight gain and changes in body composition in a free living population, and to relate these variables to changes in glucose tolerance, and the evolution of impaired glucose tolerance to frank diabetes.

1.1. Specific aims

a) Determine whether risk of incident diabetes and impaired glucose tolerance is related to physical activity in 3 populations of the African Diaspora with widely different levels of obesity.

b) Determine whether risk of incident diabetes and impaired glucose tolerance is related to rate of rise in body weight and change in body composition.

To accomplish these aims we will:

a) Measure the components of the energy budget, RMR, TDEE, and physical activity (PA) at baseline in 300 individuals with impaired glucose tolerance (IGT) and 150 normal glucose tolerant (NGT) controls.

b) Measure body composition using deuterated water dilution and BIA at baseline in these groups and relate this to glucose tolerance.

c) Determine the relationship between RMR, TDEE & PA on glucose tolerance, insulin secretion and insulin sensitivity and define these relationships within the context of rankings of weight, adiposity, and lean body mass in these groups.

d) Measure the change in the energy budget components as well as anthropometry annually for 3 consecutive years, and relate these to evolution to diabetes from IGT.

1.2. Background and significance

1.2.1. Epidemiologic transitions and diabetes

Burden of Disease in Black Populations: USA

It is well recognized that blacks in the US are at increased risk of diabetes. Prevalence rates in 1976 - 1980 among black men (bm) and women (bw) were 8.6 and 11.0% respectively, compared to 5.5 and 7.3% among white men and women [2,3]. Based on data from the Health Interview Survey (HIS), age adjusted rates of known diabetes increased among bm by 105% over the decade 1973 - 1983, while among bw they increased by 33%;during the same interval rates were constant for whites of both sexes [2]. It appears, therefore that the two-fold black:white (B:W) excess described for hypertension will soon be attained for non-insulin dependent diabetes mellitus (NIDDM). Like hypertension, NIDDM is more common among persons of lower socio-economic status and among the obese; both of these factors are more common among blacks, but whether they account for the differential has not been adequately studied. Mediating physiologic factors, such as insulin metabolism and fat distribution, may also contribute to the ethnic patterns, but again the data are limited. B:W differences in health outcomes associated with NIDDM have been well described. In 1980 diabetes was listed as the 7th leading cause of death, and blacks had rates more than twice those of whites (21.3 vs 8.4/100,000) [3]. In Michigan, blacks with NIDDM were 2.6 times more likely to suffer renal failure than whites, after adjustment for higher rates of NIDDM among blacks [4].

The Caribbean

As recently reviewed by Cruickshank, NIDDM is emerging as a major health problem in the West Indies and among Afro-Caribbeans in the UK [5,6]. Levels of obesity in the West Indies are approaching those in the US, and have increased rapidly [7]. Surveys over the last 2 decades in Jamaica have demonstrated NIDDM rates ranging from 3.9% among all adults to 18.9% among women, ages 54 - 64 [8,9,10]. The most recent surveys report prevalence rates of 10% among men and 16% among women 25 to 74 years, with an age sex adjusted rate of 13.7% [10]. In the St. James Study in Trinidad, diabetes prevalence in men was 8.2% and 14.8% among women.

West Africa

The prevalence of NIDDM in West Africa, however, appears to be very low, although the data are sparse [11]. No urban-rural difference has been recognized in sub-Saharan Africa, nor is there evidence that the rates are rising [12]. The prevalence of hyperglycaemia was reported as 1% in Mali in 1984 -1985 [13]. A study of civil servants in Ghana detected prevalence rates of 0.5% in men and 0.6% in women [14]. In urban Nigeria a prevalence of 1.7% was detected among individuals attending screening clinic [15].

NIDDM is thus an important public health problem for the US and Caribbean black populations and the burden is likely to rise in coming years. Although not currently a big problem in absolute terms for the ancestral populations in West Africa, there is the expectation that a rise in economic fortunes of the region and the accompanying adoption of increasingly technological lifestyles will precipitate an epidemic of diabetes. The full extent of the impending epidemic is as yet unknown, but is estimated at any one time by the sum of the prevalence of NIDDM and impaired glucose tolerance (IGT). This figure for the Jamaican population in 1994-1997 was 27.1% [16].

Secular Trends in Disease Patterns

A broad knowledge base regarding secular trends in risk factors and chronic disease now exists. The transition from infectious to cardiovascular (CV) and neoplastic diseases has been observed in modernizing societies all over the world. In general, increases in CV diseases and smoking related cancers have been correlated and followed the increase in risk factor exposure by approximately 30 years. The US experienced peak CV disease rates around 1965 and is now in a period of decline. Following the introduction of a high fat diet and cigarettes after World War II, Eastern Europe and the USSR are on the up-sloping portion of the epidemic curve [16]. Several Asian and Latin American countries are in the process of adopting lifestyle changes, and similar changes in the disease pattern can be safely predicted. The epidemic of diabetes, on the other hand, does not appear to be emerging in this consistent sequence of 'westernization - exposure/time lag - epidemic'. A number of population groups that have experienced rapid transition from the traditional to modernised lifestyle have already developed rates of NIDDM far in excess of those ever experienced by persons of European descent [17,18,19]. While many of these groups, typically the Melanesians and American Indians [20,21,22,23], have developed extremely high rates of obesity, other populations have also developed high rates of diabetes despite levels of obesity which remain lower than, or similar to, those in Europe and the US. The epidemiologic pattern of NIDDM is thus different in groups of European origin compared with peoples of other origin in developing countries.

Epidemiologic studies have established the strongest associations between NIDDM and obesity, decreased physical activity, and diet [3,17]. Obesity is clearly the major risk factor for NIDDM in all populations. In a recently completed survey in Jamaica, the odds ratio for NIDDM was 5.4 in the highest quartile of BMI (26.6 - 48) for men and 3.3 for women (31.1 - 59.2) compared with the lowest quartile (14.1 - 20.7 for men: 14.3 - 23.5 for women) [10]. A central distribution of fat, measured as the waist hip ratio was an even more powerful predictor; odds ratios for diabetes at the highest quartile of WHR were 17.4 for men(WHR 0.89 - 1.07, highest quartile: 0.68 - 0.79 lowest) and 5.5 for women (0.85 - 1.35, highest quartile) compared with the lowest quartile (0.64 - 0.76).

Given the strength of obesity as a risk factor for NIDDM, it is plausible to hypothesize that the differences across the economic and technological gradient might be simply the result of the variation in the prevalence of obesity. It is certainly true that the most obese populations have experienced the highest rates of NIDDM [17,18,19,20,21,22,23]. Adult prevalence rates of 25% have been observed in Nauru, where mean BMI approaches 34 [18]. Other observations are inconsistent with this hypothesis, however. In Britain, migrants of South Asian origin have high rates of both obesity and NIDDM, whereas those of Afro-Caribbean extraction are less obese but have a similar prevalence of NIDDM. An important recent survey from Mauritius casts doubt on both the simple linear hypothesis for obesity and the primary role of ethnic factors. NIDDM was very common in all the ethnic groups in Mauritius (around 15%), including persons of Indian, Chinese and African origin [25,26]. The level of obesity alone could not explain the high rates, as BMI was not markedly elevated (mean BMI = 24). If reproduced, these findings suggest two extremely important conclusions which may resolve the inconsistencies noted above. First, under similar social conditions the propensity for obesity and NIDDM is equivalent among non-Westernized groups, including those as genetically distant as Africans and Chinese. Second, rates of NIDDM can be very high in populations undergoing rapid westernization, despite modest levels of obesity.

A corollary of this second conclusion would predict that populations with gradual increases in obesity should experience slowly rising rates of NIDDM. The NIDDM trend data in US Blacks support this idea. Attention has been drawn to the paradoxically low rates of NIDDM in US blacks in Chicago in the 1980's, despite high rates of obesity [27,28]. In a community based survey, age adjusted rates of NIDDM and/or hyperglycaemia were higher in whites of both sexes (wm = 12.5%; ww = 9.6%; bm 10.0%; bw 6.3%), despite prevalence rates of severe obesity that were 20% higher in bm compared to wm, and 100% higher in bw [27]. National survey data also demonstrate that blacks did not have higher rates of NIDDM in the 1960's; the HES found race-sex patterns in blood sugar levels post-50 gm load similar to the Chicago survey, with the exception of the lower levels in wm [29]. B;W ratios of reported NIDDM prevalence were 1.0 or 1.1 from 1963 until 1973 [3]. Among military draftees in 1924,

aged 18 - 45, diabetes rates among blacks were 1/3 of those among whites, and had risen to only 2/3 by 1944 [2]. Data on 100,000 members of the Kaiser Health plan collected in the 1960's also demonstrated better glucose tolerance among blacks [30]. B:W rates crossed over in the 1970's, and the trends in NIDDM among blacks are only now reflecting the impact of the long term rise in obesity. This lag in the emergence of NIDDM among US blacks is thus in sharp contrast to the data from Mauritius, where obesity and NIDDM developed rapidly and concurrently. However, rate of weight change in a population is likely to be confounded by opposing change in the expenditure of energy on physical activity and the pattern of this activity.

The discussion above serves to underscore the complex nature of the epidemiologic transition as it relates to the emergence of obesity and NIDDM. Thus a primary research challenge is to understand the role of physical activity and obesity in the evolution of high rates of diabetes. Cross sectional relationships are the complex result of prior exposure [31]. For example, abrupt changes in obesity increases the cross sectional relationship with NIDDM while slower changes dilutes such a relationship. A population undergoing accelerated social change experiences a precipitous increase in fat and refined carbohydrate intake, a decline in physical activity, and a simultaneous increase in obesity and NIDDM. In contrast, populations with more gradually evolving lifestyles may include a large number of overweight individuals, who are relatively accommodated to both the dietary patterns and consequent obesity.

Energy expenditure

Although large scale studies using accurate measures of EE have not been conducted, some data are available which suggest that EE is an important predictor of obesity, and glucose intolerance [37,43,53]. One of the key questions addressed in this application is therefore the quantitative impact of the components of EE on weight gain and glucose intolerance in a representative sample of free-living individuals. Specifically, under normal conditions of a sedentary lifestyle, does inter-individual variation in either RMR or non-resting EE contribute to the risk of obesity and diabetes? RMR is a function of lean body mass, and non-resting EE explains more of the variance in total daily energy expenditure (TDEE) between individuals [53]. It is obviously the component under behavioural control and we, therefore, focus our primary attention on that component of the energy budget.

Survey questionnaires are inaccurate methods for capturing this information: correlations with doubly labelled water estimates of energy expenditure are generally in the range of 0.3 [54]. Similarly, heart rate monitors cannot distinguish anxiety related heart rate increases from exertion-induced changes when pulse remains in the range of 80 - 100 beats per minute, where the bulk of calorie expenditure takes place. The technical framework for assessing energy balance has two paradoxical aspects. On the one hand, the relationships described by the energy budget are obvious. Intake and expenditure must balance to maintain weight. Expenditure has only three components, viz RMR (usually 75% of total EE), non-RMR (about 20%) and a small, fixed contribution from the thermic effect of food, which can be ignored. An individual can gain weight in one of three ways: eat to excess, have a low RMR, or a low non-RMR, assuming the existence of euthyroid status. Among normal individuals, RMR is tightly regulated. Its contribution to obesity is not well defined and may be limited. On the other hand, excess intake and low levels of activity are likely to be important determinants of obesity for many individuals. These relationships are not straightforward, however. As an individual gains weight, muscle mass also increases and greater energy is required for movement. As a result, the correlation between BMI and TDEE is paradoxically positive, around 0.7. In absolute terms, therefore, obese individuals expend more calories than those who are lean and they eat more. Thus, the state of obesity is a result of long term failure to balance intake and expenditure. The cause of this imbalance which must either be excess intake or reduced expenditure can be defined only in relative terms. That is, over the period of weight gain, the individual consumed excess calories relative to what was burned. The testable question is, why do some individuals fail to balance intake with expenditure over long periods? One hypothesis must be that the proportion of calories expended in physical activity, normalised to lean mass or weight, is the determining factor. The alternative hypothesis is that energy intake is excessive.

Finally, these relationships linking energy intake and energy expenditure to changes in body composition do affect glucose insulin relationships. The body composition glucose intolerance relationships are themselves modified by physical activity, such that increasing exposure to physical activity increases insulin sensitivity and reduces tissue resistance to the action of the hormone. A hypothesis is therefore that at any level of adiposity, energy expenditure on physical activity will ameliorate the predicted decline in glucose tolerance. There will be differences in the extent of this amelioration which relate to the pattern of energy expenditure on physical activity, whether daily, sustainable expenditure, or intermittent, intense physical activity.

The data available do suggest that accurate measurement of the energy budget in population based samples could provide important insights into the problem of obesity and the closely associated disease, diabetes. Importantly, it lays the basis for potential interventions to reverse adverse trends in population weight gain and the associated diabetes epidemic.

3. PRELIMINARY STUDIES

There is an ongoing NIH funded study of the prevalence and determinants of hypertension and diabetes in Jamaica: in collaboration with colleagues in Nigeria and the USA, we produced information on populations living in the three contrasting environments. Within this context we have examined 1600 individuals of both sexes between 25 and 74 years with a 75 gm oral glucose tolerance test, and related glucose tolerance to anthropometry.

We have recently received a renewal of this NIH grant to explore among other things, the genesis of obesity and its contribution to the evolution of hypertension. This project calls for measurement of the following:

a) energy budget in 30 individuals at all three sites, Nigeria, Jamaica and the US. Energy budgets will be measured using DLW to measure TDEE, indirect calorimetry to measure RMR, fat free mass with Deuterium dilution, BIA, and skinfolds.

b) In addition, in a larger sample of 1000 individuals which will be drawn from the population already surveyed (2400), we plan to measure anthropometry and body composition using BIA, as well as their RMR. We will first relate these measurements to blood pressure cross sectionally. These individuals will be followed up over a 1 year period and weight change assessed; weight gain and body composition changes will be related to the evolution of blood pressure.

For the CRP, it is proposed to take advantage of this ongoing work, and to add on those components relating directly to the questions surrounding deterioration of IGT to NIDDM (see work proposed).

4. METHODS

4.1. Measurement of energy expenditure

The measurement of EE in free-living populations is problematic. The most accurate method of determining EE in the laboratory is a form of indirect calorimetry, respiratory gas exchange. With respiratory gas exchange either RMR or TDEE can be calculated knowing the volume of oxygen consumed and carbon dioxide produced, as measured in a respiration chamber or with a metabolic cart [55,56,57,58]. However, this method is physically restrictive and it cannot be assumed that expenditure measured while enclosed in a chamber or wearing a face mask connected by hose to a metabolic cart is representative of TDEE in free-living individuals.

While mechanical methods, i.e., heart rate monitors, pedometers, and octameters can be worn in the free-living state, they are significantly less accurate than respiratory gas exchange (+/- 15 - 40%) [59,60,61]. In addition, heart rate monitors require that multiple oxygen consumption-heart rate calibration curves be constructed for every participant [62,63], and data from electronic motion sensors are not easily translated into EE values [54]. Although some investigators have been able to predict oxygen consumption using an octameter and heart rate monitor in tandem with moderate accuracy [64], this is not reproducible [65]. Food intake and physical activity surveys are subjective, indirect measures of EE are prone to considerable error (+/- 25%), yet they are very commonly used [56,66,67,68,69,70,71]. The correlation between EE by DLW and activity questionnaire is at best 0.3 - 0.4 928) while for diet records a negative correlation is often observed [66]. Clearly therefore, the relationship between EE, weight change and glucose tolerance will be obscured by the imprecision of these self reported methods

Within the last 14 years the doubly labelled water (DLW) technique to measure TDEE in free living individuals has been developed. DLW refers to water that has been labelled with the isotopes deuterium and ^{18}O and the method is based on the differential elimination of the two isotopes [72]. Deuterium is lost only as water and ^{18}O is eliminated both as water and carbon dioxide and their respective elimination rates can be calculated by measuring the decrease in isotope content of body fluid, urine, saliva or plasma over time. The difference between the two rates is, therefore, an indirect measure of carbon dioxide production, from which TDEE can be calculated according to the methods of indirect calorimetry [73,74]. The most important feature of the DLW method is that it provides an integral of carbon dioxide production over the period studied, usually 7 to 14 days, yet requires only

periodic sampling of urine. This method has been extensively validated against respiratory gas exchange and is accurate to within 1% with a SD of 3 to 7%, depending upon the dose of stable isotopes and the length of the metabolic period [73,77].

An additional benefit of the DLW method is the accurate measurement of body composition provided using the dilution principle. One necessary step in the determination of TDEE is the calculation of the volume, i.e., total body water (TBW), in which the stable isotopes are diluted. Since water is assumed to comprise a consistent proportion of body mass, it is possible to calculate with great accuracy an individual's fat free mass and therefore, fat mass.

Daily EE has three components: RMR (or basal MR), thermic effect of food, and physical activity. RMR typically comprises 50 to 60% of TDEE, thermic effect of food about 10%, and physical activity the remaining proportion. When TDEE is measured using DLW and RMR is measured using respiratory gas exchange, non-resting expenditure can be calculated. The thermic effect of food is a relatively constant proportion of energy expenditure [78], therefore, non-resting EE can be considered a measure of physical activity.

To date the DLW method has been primarily employed in controlled experimental projects, and until recently the high costs of isotopes and their mass spectrometric analyses have prevented its use in large scale studies. In November 1993, the cost per participant was quoted at $1200 by a laboratory providing such analyses. However, recent reduction in the price of isotopes and improvements in analytical technology has forced prices down to the range of $300 to $350 per participant. This reduction in costs make DLW competitive with other less valid ways of measuring TDEE.

4. RESEARCH DESIGN AND METHODS

a) Restating the specific aims as tasks to be accomplished, the primary goals are:

b) Screen 2000 individuals with repeat fasting blood glucose to identify 300 individuals with impaired glucose tolerance (IGT), and randomly select 150 individuals from the population with normal glucose tolerance. In these groups, we will measure the components of the energy budget; RMR by indirect calorimetry, TDEE with doubly labelled water, and physical activity by difference

c) Use deuterated water and BIA to measure body composition at baseline also in these groups.

d) Assess glucose insulin relationships in these groups by use of a 75g OGTT to measure glucose tolerance, and by calculations using the HOMA model to calculate insulin sensitivity, and the 30 minute increment in insulin to glucose to derive and index of insulin secretion.

e) Measure the change in the energy budgets, anthropometry and glucose insulin relationships annually for 3 years, and

f) Relate these changes in glucose insulin relationships and the evolution of IGT to NIDDM to baseline physical activity and anthropometry as well as the change in these characteristics.

5. PROPOSED WORK

To achieve these goals we will accomplish the following:

5.1. Sampling procedure

From among the population recruited by cluster sampling (probability proportionate to size) for the initial survey (n = 3,000), 300 individuals with IGT will be identified by:

- subjecting a random sample of 2000 individuals 25 - 65 years to repeat fasting glucose estimations on two occasions a week apart.

- selecting 150 NGT individuals randomly drawn from the subgroup above who have normal repeat fasting glucose estimations.

5.2. Measurements

5.2.1. Anthropometry and blood pressure

Weight, height, and waist and hip circumferences will be measured with an established protocol [79,80]. Weight is measured to the nearest 0.1kg utilizing a calibrated electronic digital scale; standing height is measured without shoes to the nearest 0.1 cm. For both waist and hip circumferences, the measurement is repeated. If two measurements differ by more than 0.5 cm, a third measurement is taken. Mid upper arm circumference will also be measured. The circumferences will be used as measurements of regional fat distribution. All measurements will be made blinded to group assignment.

5.2.2. Body composition

Total body water (TBW) will be determined in the course of the TDEE calculations. The basis of TBW determination is the dilution principle (C1V1 = C2V2), i.e., if a known amount and concentration of a substance is diluted in a larger volume and you can measure the final concentration of the substance, the final volume can be calculated. Therefore, TBW can be calculated using measurement of the abundance of either isotope, deuterium or 18O after complete equilibration.

Except in cases of malnutrition, sever dehydration, and in some cases such as AIDS or nephropathies, TBW is assumed to make up a constant proportion (73%) of lean body mass (also known an fat free mass) [81]. Fat free mass, therefore, can be calculated from TBW. Fat mass can then be calculated as the difference between total body weight and fat free mass. Calculation of fat mass provides a more valid measure of adiposity tan BMI [82].

5.2.3. BIA

TBW will also be determined using BIA. BIA measures the impedance to the flow of an applied low alternating current by body tissues that is undetectable by the subject. Impedance is a function of two components, the resistance of the tissues and the additional opposition, called reactance, due to the capacitance of cells. The measured impedance of body tissues provides an estimate of total body water from which FFM and FM can be calculated [83,84].

Subjects will be placed in the supine position with limbs abducted. Current-supplying electrodes will be placed on the dorsal surfaces of the right hand and foot at the metacarpals and metatarsals respectively. Electrodes for the detection of the current will be placed at the pisiform prominence of the right wrist and between the medial and lateral malleoli of the right ankle. The BIA analyses (Model BIA 101Q, RJL Systems, Clinton Twp., MI) will be attached to the electrodes and generate an excitation current of 800µA at a signal frequency of 50kHz. Resistance to current flow is measured by the BIA analyser. The instruments are calibrated every day before use and serviced regularly.

Multiple equations have been developed for the prediction of total body water from BIA measurements. Published equations calculate body water to within 0.2+/- 1.5 kg of that by deuterium dilution in these populations [83]. For participants in the EE sub-study, total body water will also be measured using isotopes, a method based on the dilution principle.

5.2.4. Energy expenditure

EE will be measured in 300 unrelated individuals with IGT and 150 with NGT in our survey sample using the doubly labelled method over a 2-week measurement period [85]. Based on available lists we will identify appropriate participants between the ages of 25 and 65, equal numbers of men and women, free of chronic illness limiting activity.

5.2.5. Resting metabolic rate

RMR will be measured using respiratory gas exchange in the clinics with a metabolic cart (Datex Deltatrac, Sensor Medics, Anaheim, Ca). In order to control the thermic effect of food the participants will be asked to fast from 10pm the previous evening. Participants will be positioned comfortably in a supine position on the examination couch. The clear ventilated hood of the metabolic cart will be placed over the participant's head and respiratory gases will be measured for 45 to 60

minutes. O_2 and CO_2 are continuously sampled during the procedure and using the modified Weir equation [86], EE is calculated.

The metabolic cart will be calibrated with standard gases prior to each RMR measurement. Once every two weeks an alcohol burn will be done to calibrate the flow rate of gases through the instrument and provide standardized measurements for comparisons across sites. Every 20th participant will be recalled for a second measurement to monitor intra person variability. The within-person CV for this method is 1.5% in our laboratory.

5.2.6. Total energy expenditure

The TDEE measurement will begin at the clinic visit. The protocol for TDEE measurement requires a baseline spot urine sample, oral administration of the DLW, collection of the first 3 urine voids post dose (same day), collection of a spot urine sample on day 8 (midpoint) and collection of the final spot urine on day 14. On the morning of the examination, prior to the measurement of RMR, a spot urine collection will be made and the participant will be given a dose of water determined from data gathered at the screening examination. Spot urine collections will also be collected at 2, 4 and 6 hours after isotope administration. Exact 2-hour spacing is not crucial. What is important is that the first three urine after dosing with DLW are sampled; it requires 3 to 4 hours for the isotope to fully equilibrate with body water.

Before leaving the clinic, the participants will be instructed on the collection of urine on days 8 and 14. The participants will be visited on day 7 by the field staff and reminded to collect the spot sample on the following day (day 8). The participant will then return to clinic on day 14 for the final spot urine collection to end the study period. While at the clinic on day 14, each participant will have their weight measured and body fat determined by BIA.

5.2.7. Physical activity

Physical activity will be calculated from the DLW and respiratory gas exchange data as non-resting EE. While the difference between TDEE and RMR includes both physical activity and the thermic effect of food, thermic effect of food will be considered to be a consistent proportion of TDEE and thus not separated from physical activity [87]. The actual measurement of the thermic effect of food will not be performed because it places unreasonable burden on the participant, requiring 4 to 6 hours of respiratory gas exchange i.e., 4 to 6 hours remaining awake but motionless under a hood.

REFERENCES

[1] COOPER, R., ROTIMI, C., ATAMAN, S., MCGEE, D., OSOTIMEHIN, B., KADIRI, S., MUNA, W., KINGUE, S., FRASER, H., FORRESTER, T., BENNETT, F., WILKS, R., Hypertension in seven populations of African origin, Am J Publ Health, **87** (1997) 160 - 168.

[2] HARRIS, M.I., Epidemiologic correlates of NIDDM in Hispanics, whites, and blacks in the US population, Diab Care, **14 (Suppl 3)** (1991) 639 - 648.

[3] Diabetes in America, HARRIS, M.I., HAMMAN, R.F., Eds., US DHHS, NIH Publication No. 85-1468, Bethesda, MD (1985).

[4] COWIE, C.C., PORT, F.K., WOLFE, R.A., SAVAGE, P.J., *et al.*, Disparities in incidence of end stage renal disease according to race and type of diabetes, N Engl J Med, **321** (1989) 1074 - 1079.

[5] CRUICKSHANK, J.K., Cardiovascular disease in black and Indian origin populations outside the USA. In CRUICKSHANK, J.K., BEEVERS, D.G., (Eds.) Ethnic Factors in Health and Disease. Butterworth & Co Ltd. (1989) 255 -279.

[6] CRUICKSHANK, J.K., ALLEYNE, S.A., Black West Indian and matched white diabetics in Britain compared with diabetics in Jamaica: body mass, blood pressure, and vascular disease. Diab care, **10** (1987) 170 - 179.

[7] FORRESTER, T., WILKS, R., BENNETT, F., MCFARLANE-ANDERSON, N., MCGEE, D., COOPER, R., FRASER, H., Obesity in the Caribbean, In: The Origins and Consequences of Obesity. Ciba Foundation Symposium 201, John Wiley & Sons, Chichester, UK (1996) 17 - 31.

[8] FLOREY, C DUV., MCDONALD, H., MIALL, W.E., The prevalence of diabetes in a rural population of Jamaican adults, Int J Epidemiol, **1** (1972) 157 - 166.

[9] MILLER, G.J., KIRKWOOD, B.P., BECKLES, G.L.A., Incidence of hypertension and non-insulin diabetes mellitus and associated risk factors in a rapidly developing country: The St. James survey, Trinidad. J Epidemiol Community Health, **50** (1996) 497 - 504.

[10] WILKS, R., ROTIMI, C., BENNETT, F., MCFARLANE-ANDERSON, N., KAUFMAN, J., COOPER, R.S., CRUICKSHANK, J.K., FORRESTER, T., Diabetes in the Caribbean: Results of a population survey from Spanish Town Jamaica (in Press).

[11] MCLARTY, D.G., POLLITT, C., SWAI, A.B.M., Diabetes in Africa, Diabetic Medicine, **7** (1990) 670 - 684.

[12] MCLARTY, D.G., SWAI, A.B.M., KITANGE, H.M., *et al.,* Prevalence of diabetes and impaired glucose tolerance in rural Tanzania, Lancet, **1** (1989) 871 - 874.

[13] FISCH, A., PICHARD, E., PRAZUCK, R., LEBLANC, H., SIDIBIE, Y., BRUCKER, A., Prevalence and risk factors of diabetes mellitus in the rural region of Mali (West Africa): A practical approach. Diabetologia, **30** (1987) 859 - 862.

[14] POBEE, J.O.N.M., Risk factors for coronary heart disease in an African population: The civil servants project in Accra, Cardiovasc Epidemiol Newsletter, **29** (1980) 8.

[15] OHWOVORIOLE, A.E., KUTI, J.A., KABIAWU, S.I.O., Casual blood glucose levels and prevalence of undiscovered diabetes mellitus in Lagos metropolis Nigerians, Diabetes Res Clin Pract, **4** (1988 152 - 158.

[16] COOPER, R., SCHATZKIN, A., The pattern of mass disease in the USSR, Int J Health Serv, **12** (1982) 459 - 480.

[17] JARRETT, R.J., Epidemiology and public health aspects of non-insulin dependent diabetes mellitus, Epidemiologic Reviews, **11** (1989) 151 - 171.

[18] DOWSE, G.K., ZIMMET, P.Z., FINCH, C.F., COLLINS, V.R., Decline in the incidence of epidemic glucose intolerance in Nauruans implications for the "Thrifty Genotype", Am J Epidemiol, **133** (1991) 1093 - 1104.

[19] KING, H., ZIMMET, P., Trends in the prevalence and incidence of diabetes: Non-insulin dependent diabetes mellitus, World Health Statistics Quarterly, **44** (1988) 190 - 196.

[20] SAVAGE, P.J., HARLAN, W.R., Racial and ethnic diversity in obesity and other risk factors, Ethnicity Dis, **1** (1991) 201.

[21] ZIMMET, P., Epidemioloy of diabetes and its macrovascular manifestations in Pacific populations: The medical effects of social progress, Diabetes Care, **2** (1979) 144 - 153.

[22] O'DEA, K., Westernization and non-insulin dependent diabetes in Australian aborigines, Ethnicity Dis, **1** (1991) 172.

[23] KNOWLER, W.E., PETTIT, D.J., SAVAGE, P.J., BENNETT, P.H., Diabetes incidence in Pima Indians: Contributions of obesity and parental diabetes, Am J Epidemiol, **113** (1981) 144 -156.

[24] MCKEIGUE, P.M., SHAH, B., MARMOT, M.G., Relation of central obesity and insulin resistance with high diabetes prevalence and cardiovascular risk in South Asians, Lancet, **337** (1991) 382 - 386.

[25] DOWSE, G.K., GAREEBO, H., ZIMMET, P.Z., *et al.*, High prevalence of NIDDM and impaired glucose tolerance in Indian, Creole and Chinese Mauritians, Diabetes, **39** (1990) 390 - 396.

[26] DOWSE, G.K., ZIMMET, P.Z., GAREEBO, H., *et al.*, Abdominal obesity and physical activity as risk factors for NIDDM and impaired glucose tolerance in Indian, Creole and Chinese Mauritians, Diabetes, **14** (1991) 271 - 282.

[27] COOPER, R., LIU, K., STAMLER, J., *et al.*, Prevalence of diabetes/hyperglycemia and associated cardiovascular risk factors in blacks and whites: Chicago Heart Association Medical Detection Project in Industry, Am Heart J, **108** (1984) 827 - 832.

[28] PERSKY, V., DYER, A., STAMLER, J., *et al.*, The relationship between postload plasma glucose and blood pressure at different resting heart rates, J Chron Dis, **32** (1979) 263 - 268.

[29] Blood glucose levels in adults: United States (1960 - 1962); NCHS, Vital and Health Statistics Series 11, No. 18. Washington DC (1966).

[30] DALES, L.G., SIEGELAUB, A.B., FELDMAN, R., *et al.*, Racial differences in serum and urine glucose after glucose challenge, Diabetes, **23** (1974) 327 - 332.

[31] KUMANYIKA, S., GOLDEN, P., Cross-sectional differences in health status in US racial/ethnic minority groups: Potential influence of temporal changes, disease, and lifestyle transitions, Ethnicity Dis, **1** (1991) 50 - 59.

[32] NICHAMAN, M.Z., GARCIA, G., Obesity in Hispanic Americans, Diabetes Care, **14** (1991) 191 - 194.

[33] WING, R.R., KULLER, L.H., BUNKER, C., *et al.*, Obesity, related behaviours and coronary heart disease risk factors in black and white pre-menopausal women, Int J Obesity, **13** (1989) 511 - 519.

[34] KUSHNER, R.F., RACETTE, S., NEIL, K., SCHOELLER, D.A., Measurement of physical activity among black and white obese women, Obesity Res, **3** (1995) 261s - 265s.

[35] LOUARD, R., HENDLER, R., WELLE, S., AMATRUDA, J., Differences in basal metabolic rate and total energy expenditure in black and white overweight women, Int J Obesity, **18** (1994) 6.

[36] MORRISON, J.A., ALFARO, M.P, KHOURY, P., THORNTON, B.B., DANIELS, S.R., Determinants of resting energy expenditure in young black girls and young white girls, J Pediatr, **129** (1996) 637 - 642.

[37] KNOWLER, W.C., PETTITT, D.J., SAAD, M.F., *et al.*, Obesity in Pima Indians: Its magnitude and relationship with diabetes, Am J Clin Nutr, **53** (1991) 1543s - 1551s.

[38] National Heart Lung & Blood Institute Conference on Obesity and Cardiovascular Diseases in Minority Populations. Executive summary. Conference highlights, conclusions and recommendations, Am J Clin Nutr, **53** (1991) 1507s - 1511s.

[39] RAVUSSIN, E., FONTVIELLE, A.M., SWINBURN, B.A., BOGARDUS, C., Risk factors for the development of obesity, Ann NY Acad Sci, **683** (1993) 141 - 150.

[40] BOUCHARD, C.A., TREMBLAY, J.P., DSEPRES, A., *et al.*, The response to long-term overfeeding in identical twins, New Engl J Med, **322** (1990) 1477 - 1482.

[41] STUNKARD, A.J., HARRIS, J.R., PEDERSON, N.L., MCLEAN, G.E., The body mass index of twins who have been reared apart, New Engl J Med, **322** (1990) 1483 - 1487.

[42] RICE, T., TREMBLAY, A., DEROIAZ, O., PERUSSE, L., RAO, D.C., BOUCHARD, C., A major gene for high resting metabolic rate unassociated with body composition: Results from the Quebec Family Study, Obesity Res, **4** (1996) 441 - 449.

[43] RAVUSSIN, E., LILLIOJA, S., KNOWLER, W.C., CHRISTIN, L., *et al.*, Reduced rate of energy expenditure as a risk factor for body-weight gain, New Engl J Med, **318** (1988) 467 - 472.

[44] PERUSSE, L., DESPRES, J.P., LEMIEUX, S., RICE, T., RAO, D.C., BOUCHARD, C., Familial aggregation of abdominal visceral fat level: results from the Quebec Family Study, Metabolism, **45** (1996) 378 - 382.

[45] FORBES, G.B., Do obese individuals gain weight more easily than non-obese individuals? Am J Clin Nutr, **52** (1990) 222 - 227.

[46] CONSIDINE, R.V., SINHA, M.K., HEIMAN, *et al.*, Serum immunoreactive-leptin concentrations in normal weight and obese humans, N Engl J Med, **334** (1996) 292 - 295.

[47] DAGOGO-JACK, S., FANELLI, C., PARAMORE, D., BROTHERS, J., LANDT, M., Plasma leptin and insulin relationships in obese and non-obese humans, Diabetes, **45** (1996) 695 - 698.

[48] LEVIN, N., NELSON, C., GURNEY, A., VANDIEN, R., DESAUVAGE, F., Decreased food intake does not completely account for adiposity reduction after ob protein infusion, Proc Natl Acad Sci USA, **93** (1996) 1726 - 1730.

[49] CLEMENT, K., VAISSE, C., MANNING, B.S., *et al.*, Genetic variation in beta 3-adrenergic receptor and an increased capacity to gain weight in patients with morbid obesity, New Engl J Med, **333** (1995) 352 - 354.

[50] URHAMMER, S.A., CALUSEN, J.O., HANSEN, T., PEDERSEN, O., Insulin sensitivity and body weight changes in young white carriers of the codon 64 amino acid polymorphism of the beta 3-adrenergic receptor gene, Diabetes, **45** (1996) 1115 - 1120.

[51] BOUCHARD C. Human obesities: chaos or determinism? In Obesity in Europe 91, AILHAUD, G., GUY-GRAND, B., LAFONTAN, M., RICQUIER, D., Eds. London: JOHN LIBBEY (1992) 7 - 14.

[52] ASTRUP, A., BUEMANN, B., WESTERN, P., *et al.*, Obesity as an adaptation to a high-fat diet: Evidence from a cross sectional study, Am J Clin Nutr, **59** (1994) 350 - 355.

[53] RISING, R., HARPER, I.T., FONTVIELLE, M.A., *et al.*, Determinants of total daily energy expenditure: Variability in physical activity, Am J Clin Nutr, **59** (1994) 800 - 804.

[54] MONTOYE, H.J., KEMPER, H.C.G., SARIS, W.H.M., WASHBURN, R.A., Measuring physical activity and energy expenditure, Champaign IL, Human Kinetics (1996).

[55] RAVUSSIN, E., LILLIOJA, S., KNOWLER, W.C., *et al.*, Reduced rate of energy expenditure as a risk factor for body weight gain, New Engl J Med, **18** (1988) 467 - 472.

[56] SCHOELLER, D.A., RACETTE, S.B., A review of field techniques for the assessment of energy expenditure, J Nutr, **120** (1990) 1492 - 1495.

[57] JEQUIER, E., SCHUTZ, Y., Long term measurements of energy expenditure I humans using a respiration chamber, AmJ Clin Nutr, **38** (1983) 989 -998.

[58] RAVUSSIN, E., LILLIOJA, S., ANDERSON, T.E., CHRISTIN, L., BOGARDUS, C., Determinants of 24-hour energy expenditure in man: Methods and results using a respiratory chamber. J Clin Invest 1986; 78, 1568 - 1578.

[59] LIVINGSTONE, M.B., PRENTICE, A.M., COWARD, W.A., *et al.*, Simultaneous measurement of free-living energy expenditure by doubly labelled water method and heart rate monitoring, Am J Clin Nutr, **52** (1990) 59 - 65.

[60] RACETTE, S.B., KUSHNER, R.F., SCHOELLER, D.A., Comparison of heart rate and physical activity recall questionnaire with doubly labelled water in obese women, Med Sci Sports Exerc, **27** (1995) 126 - 133.

[61] HOYT, R.W., JONES, T.E., BAKER-FULCO, C.J., *et al.*, Doubly labelled water measurement of human energy expenditure during exercise at high altitudes, Am J Physiol, **266** (1994) R966 - R971.

[62] LI, R., DURNBERG, P., HAUTVAST, J.G.A.J., A critical evaluation of heart rate monitoring to assess energy expenditure in individuals, Am J Clin Nutr, **58** (1993) 602 - 607.

[63] MONTOYE, H.J., TAYLOR, H.L., Measurement of physical activity in population studies: A review, Human Biol, **56** (1984) 196 - 216.

[64] HASKEIL, W.L., YEE, M.C., EVANS, A., IRBY, P.J., Simultaneous measurement of heart rate and body motion to quantitate physical activity, Med Sci Sports Exerc, **25** (1993) 109 - 115.

[65] LUKE, A., MAKI, K., BARKEY, N., MCGEE, D., COOPER, R.S., Simultaneous monitoring of heart rate and motion to assess energy expenditure, Med Sci Sports Exer, **29** (1997) 144 - 148.

[66] SCHOELLER, D.A., BANDINI, L.G., DIETZ, W.H., Inaccuracies in self-reported intake identified by comparison with the doubly labelled water method, Can J Physiol Pharmacol, **68** (1990) 941 - 949.

[67] ACHESON, K.J., CAMPBELL, I.T., EDHOLM, O.G., MILLER, D.S., STOCK, M.J., The measurement of daily energy expenditure- an evaluation of some techniques, Am J Clin Nutr, **33** (1980) 1155 - 1164.

[68] EDWARDS, J.E., LINDEMAN, A.K., MIKESKY, A.E., STAGER, J.M., Energy balance in highly trained female endurance runners, Med Sci Sports Exerc, **25** (1993) 1398 - 1404.

[69] BLACK, A.E., PRENTICE, A.M., GOLDBERG, G.R., *et al.*, Measurements of total energy expenditure provide insights into the validity of dietary measurements of energy intake, J AM Diet Assoc, **93** (1993) 672 - 679.

[70] RISING, R., HARPER, I.T., FONTVIELLE, A.M., FERRARO, R., SPRAUL, M., RAVUSSIN, E., Determinants of total daily energy expenditure: Effect of age and body fat on physical activity in Pima Indian men, Am J Clin Nutr, **59** (1994) 800 - 804.

[71] HOWAT, P.M., MOHAN, R., CHAMPAIGNE, C., *et al.*, Validity and reliability of reported dietary intake, J Am Diet Assoc, **94** (1994) 169 - 173.

[72] SCHOELLER, D.A., VAN SANTEN, E., Measurement of energy expenditure in humans by doubly labelled water method, J Appl Physiol, **53** (1982) 955 - 959.

[73] RAVUSSIN, E., HARPER, I., RISING, R., BOGARDUS, C., Energy expenditure by doubly labelled water: Validation in lean and obese subjects, Am J Physiol, **261** (1991) E402 - E409.

[74] SCHOELLER, D.A., RAVUSSIN, E., SCHUTZ, Y., *et al.*, Energy expenditure by doubly labelled water: Validation in humans and proposed calculation, Am J Physiol, **250** (1986) R823 - R830.

[75] SEALE, J.L., CONWAY, J.M., CANARY, J.J., Seven day validation of doubly labelled water method using indirect room calorimetry, J Appl Physiol, **74** (1993) 402 - 409.

[76] COLE, T.J., COWARD, W.A., Precision and accuracy of doubly labelled water energy expenditure by multipoint and two-point methods, Am J Physiol, **263** (1992) E965 - E973.

[77] JEQUIER, E., SCHUTZ, Y., Classical respirometry and the doubly labelled water energy expenditure method: Appropriate applications of the individual or combined techniques, Proc Nutr Soc, **47** (1988) 219 - 225.

[78] THORNE, A., Diet induced thermogenesis: An experimental study in lean and obese individuals, Act Chir Scand, **558** (1992) 6 - 59.

[79] ROTIMI, C.N., COOPER, R.S., ATAMAN, S.L., MCGEE, D., OSOTIMEHIN, B., KADIRI, S., MUNA, W., KINGUE, S., FRASER, H., Distribution of anthropometric variables and the prevalence of obesity in populations of West African origin: The international collaborative study on hypertension in blacks, Obesity Res (1995) 95S - 106S.

[80] ATAMAN, S.L., ROTIMI, C., COOPER, R., MCGEE, D., OSOTIMEHIN, B., KADIRI, S., KINGUE, S., MUNA, W., FRASER, H., FORRESTER, T., WILKS, R., Standardization of blood pressure measurement in an international comparative study, J Clin Epidemiol, **49** (1996) 869 - 877.

[81] MOORE, F.D., OLESEN, K.H., MCMURREY, J.D., *et al.*, The body cell mass and its surrounding environment, Philadelphia, PA, Saunders (1963) 13 - 42.

[82] SMALLEY, K.J., KNERR, A.N., KENDRICK, Z.V., COLLIVER, J.A., OWEN, O.E., Re-assessment of body mass indices, Am J Clin Nutr, **52** (1990) 405 - 408.

[83] KUSHNER, R.F., SCHOELLER, D.A., Estimation of total body water by bio-electrical impedance analysis, Am J Clin Nutr, **4** (1986) 417 - 424.

[84] LUKASKI, H.C., BOLONCHUK, W.W., HALL, C.B., SIDERS, W.A., Validation of tetrapolar bio-electrical impedance methods to assess human body composition, J Appl Physiol, **40** (1986) 59 - 66.

[85] SCHOELLER, D.A., RAVUSSIN, E., SCHUTZ, Y., *et al.*, Energy expenditure by doubly labelled water: Validation in humans and proposed calculations, Am J Physiol, **250** (1986) R823 - R830.

[86] WEIR, J.B., New methods for calculating metabolic rate with special references to protein metabolism, J Physiol, **109** (1949) 1 - 9.

[87] PANDOFF, K.B., GIVONI, B., GOLDMAN, R.F., Predicting energy expenditure with loads while standing or walking very slowly, J Appl Physiol, **43**, 577 - 581.