**Insulin sensitivity and insulin clearance in adult survivors of malnutrition**

Debbie S. Thompson MBBS1, Patrice M. Francis-Emmanuel DM 1,2, Alan T. Barnett DM 1,3, Tamika Royal-Thomas PhD1, Clive Osmond PhD4, Mark A. Hanson D Phil5, Christopher D. Byrne PhD6, Peter D. Gluckman DSc7, Terrence E. Forrester PhD1, Michael S. Boyne MD1

1Tropical Medicine Research Institute, The University of the West Indies, Mona, Jamaica;

2Department of Medicine, The University of the West Indies, Mona, Jamaica;

3Department of Surgery, Radiology, Anesthesia and Intensive Care, The University of the West Indies, Mona; Jamaica;

4MRC Lifecourse Epidemiology Unit, University of Southampton, UK;

5Institute of Developmental Sciences, University of Southampton, UK;

6Nutrition and Metabolism Unit, School of Medicine, University of Southampton, UK;

7Centre for Human Evolution, Adaptation and Disease, Liggins Institute, University of Auckland, New Zealand.

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*Corresponding author and person to whom reprint requests should be addressed:*

Debbie S. Thompson

Tropical Metabolism Research Unit, Tropical Medicine Research Institute

The University of the West Indies, Mona, Kingston 7, Jamaica.

876-977-6251 (phone), 876-977-0632 (fax)

E-mail: [debbie.thompson@uwimona.edu.jm](mailto:debbie.thompson@uwimona.edu.jm)

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**Abstract**

**Context and Objective:** Among the clinical syndromes of severe acute malnutrition (SAM), adult survivors of marasmus (MS) have worse glucose tolerance than kwashiorkor survivors (KS). We hypothesized that MS have lower insulin sensitivity and insulin clearance (MCR) than KS. We investigated glucose metabolism in a cohort of adult survivors of SAM and a control group that was not exposed to SAM.

**Research Design and Setting:** This was a cross sectional, observational study of Jamaican adults.

**Subjects:** 40 non-diabetic adult survivors of SAM (20 MS and 20 KS) and 13 age, sex and BMI-matched controls. Exclusion criteria: diabetes, pregnancy, lactation, smoking, chronic illnesses and glucocorticoid use.

**Measurements:** We performed 150-minute hyperinsulinaemic, euglycaemic clamps, measured serum adiponectin, anthropometry and body composition on the participants. We estimated insulin sensitivity using the M-value and MCR was calculated.

**Results:** The mean age (± SD) was 27.2 ± 8.1 years, BMI was 23.6 ± 5.0 kg/m2 and 45% were male. MS and KS were similar in age, weight, BMI, fat free mass and fat mass. MS had similar insulin sensitivity to KS (M-weight: 9.1 ± 3.2 vs. 8.7 ± 4.6mg.kg-1.min-1; *P* = 0.51). MCR was also similar among MS and KS (*P* = 0.49). When all SAM survivors were compared to the community controls, both insulin sensitivity and MCR were similar (*P*-values ≥ 0.16).

**Conclusions:** Lean young adult survivors of marasmus and kwashiorkor have similar insulin sensitivity and insulin clearance. It remains to be seen whether this is also true among obese survivors of malnutrition.

**Introduction**

Severe acute malnutrition (SAM) is globally the most important risk factor for illness and death in children, contributing to more than half of deaths worldwide (1, 2). The Wellcome criteria classify marasmus as severe wasting (< 60% weight-for-age) without nutritional oedema, and kwashiorkor as moderate wasting (60 - 80% weight-for-age) with nutritional oedema (3).

Despite the significant differences between kwashiorkor and marasmus, the etio-pathogenesis of these syndromes is not well understood. However, infants admitted with marasmus have higher rates of lipolysis, protein turnover and salvage of urea-nitrogen than infants with kwashiorkor (4, 5); a phenotype similar to that seen in insulin resistant states. These data suggest that marasmus represents an adaptation to starvation whereas kwashiorkor represents a dysadaptation.

Also, infants who developed marasmus had lower birth weights than those who developed kwashiorkor (4) suggesting that the clinical syndromes may have distinct developmental origins. As lower birth weight children may develop insulin resistance in later life (6), we could expect adult survivors of marasmus to be insulin resistant. Previously, we showed that adult survivors of marasmus have more impaired glucose tolerance, fasting hyperinsulinaemia, and worse beta-cell function compared to kwashiorkor survivors as measured during an oral glucose tolerance test (7). Notably, there was a tendency towards more insulin resistance (*P* = 0.06) (7). While the fasting hyperinsulinaemia in marasmus survivors could reflect basal hypersecretion of insulin, it is also possible this could be due to reduced metabolic clearance of insulin. Derangements in insulin sensitivity, insulin secretion, and insulin clearance contribute independently to the development of glucose intolerance (8).

We therefore hypothesized that adult survivors of marasmus could have lower insulin sensitivity and lower insulin clearance compared to adult survivors of kwashiorkor. We measured insulin sensitivity and insulin clearance during a hyperinsulinaemic euglycaemic clamp (HEC) in adult survivors of SAM with no self-reported history of diabetes mellitus as well as in community controls who never experienced SAM.

**Materials and Methods**

*Study design*

As previously described (4), we assembled a retrospective cohort of 1336 patients who were admitted to the metabolic ward of the Tropical Metabolism Research Unit, Jamaica at age 6-18 months from 1963 to 1992. The mortality rate was low (i.e. 4.1%) during this period. Community health aides and nurses were able to trace 221 of these marasmus and kwashiorkor survivors (MS and KS, respectively). Of these, 20 MS and 20 KS agreed to have a 150-minute HEC. We also recruited 13 community controls that never had SAM and who consented to the clamp study. These controls were matched for age ± 5 years, sex, and body mass index (BMI) ± 2 kg/m2 and were selected from the same street address as the cases. We excluded participants with a history of diabetes and those who were pregnant, lactating, using tobacco, had chronic illnesses or who used glucocorticoids. The Faculty of Medical Sciences/ University Hospital of the West Indies Ethics Committee approved the study protocol. Each participant gave written informed consent.

*Procedures*

Participants were admitted to the metabolic ward and after a 10 hour overnight fast, their studies were started at 0800 hr. Urine β-HCG was performed on all female participants to rule out pregnancy. We measured anthropometry, body composition by DEXA, and drew blood for serum adiponectin.

*HEC*

Insulin was infused through a left antecubital fossa catheter at a rate of 40 µIU/m2/min. 20% dextrose water was infused at a variable rate to maintain blood glucose at or near 5 mmol/l. Blood was sampled every 5 minutes for glucose concentration (YSI Instruments, Yellow Springs, OH) through a retrograde cannula in the right hand that was kept in a warm box set at 50°C. Blood was collected every 10 minutes in a fluorinated tube to measure plasma glucose and a heparinized tube to measure insulin concentration.

*Assays*

Glucose concentration was determined by the glucose oxidase method. Plasma insulin was measured using an immunoassay technique (ALPCO Diagnostics, NH, USA) which had an analytical sensitivity of 0.399 µIU/mL. The intra-assay coefficient of variation (CV) was 3.1% in our laboratory and the inter-assay CV was < 8%. Serum adiponectin was measured using a commercial ELISA kit (Linco Research, MO, USA) which had a sensitivity of 7.8ng/ml. The intra-assay and inter-assay CVs were ≤ 8%. Adiponectin was not measured in the community controls due to inadequate amounts of stored serum.

*Calculations and Data Analysis*

Steady state during the clamp was defined as a thirty minute period 1-2 hours after the start of the insulin infusion, where the coefficients of variability for plasma glucose, plasma insulin and glucose infusion rate were ≤ 5%. Mean parameter values during the steady state were used to calculate whole body insulin-mediated glucose uptake (M; mg/kg/min):

M = GIR - SC, where GIR is the glucose infusion rate and SC is the space correction.

GIR = *∑(rate of infusion ) x 17/weight (kg) x time (min)* (9)

SC (mg/kg/min) = *(G2-G1) x 0.063* (9)

G2 and G1 are the plasma glucose concentrations (mmol/L) at the end and beginning of the 30-min time period, respectively. M was also normalized for lean mass (M-lean). The insulin sensitivity index *(*M/Iratio) was calculated by dividing M by the mean insulin concentration during the same period of the clamp. MCR was calculated during the steady state as:

MCR = *insulin infusion rate/(mean insulin - basal insulin)* (9).

This computation is based on the assumption that basal insulin secretion is unchanged by the insulin infusion (9).

HOMA-IR was calculated as follows:

HOMA-IR = fasting glucose (mmol/L) × fasting insulin (μU/ml)/22.5 (10)

Fat mass, fasting insulin, adiponectin, M, M-lean, M/I and MCR data were skewed and were log transformed to normality. Independent T-tests were used to compare the mean differences in adult anthropometry, body composition, adiponectin and indices of glucose metabolism between MS and KS and malnutrition survivors and controls. Multivariate regression analyses were used to assess the correlations between adult anthropometry, body composition, adiponectin and indices of glucose metabolism. All measurements were adjusted for age and sex in the analyses. Analyses were performed using Stata 12.1 (StataCorp, Texas) and *P*-values ≤ 0.05 were statistically significant.

**Results**

Data were analysed for 40 survivors of SAM and 10 controls; 2 controls had no detectable basal insulin in the assays and 1 had fasting hyperglycemia. The 10 controls that were analyzed were matched to 7KS and 3MS. The mean age (± SD) was 27.2 ± 8.1 years, BMI was 23.6 ± 5.0 kg/m2 and 45% were male. There was no difference between MS and KS with respect to age, weight, height, waist, fat free mass, fat mass and insulin (Table). MS had a higher fasting glucose than KS after adjusting for age and sex (*P* ≤ 0.001). There was no difference in insulin sensitivity between MS and KS using HOMA-IR, M, M-lean or M/I. Adjustment for age, sex and BMI did not change these relationships (*P*-values > 0.2; data not shown).

MCR was similar among MS and KS even after adjusting for age and sex. There was a positive correlation between M/I and MCR (r = 0.91, *P* = 0.01) which remained after adjusting for age and sex (*P* = 0.02) (Figure). However, there was no correlation between M or M-lean and MCR. Similarly, using pairwise comparisons, there were no correlations between MCR and age, BMI, and total fat mass.

Serum adiponectin was similar between groups even after age, sex and BMI adjustment. There was no significant association between M-weight and adiponectin adjusting for age and sex (*P* = 0.26). When we compared all survivors of SAM (i.e. MS and KS combined) with community controls, there was no difference in anthropometry, body composition, insulin sensitivity and MCR. However the controls had higher fasting plasma glucose compared to SAM survivors (*P* =0.001) even after adjusting for age and sex (*P ≤* 0.001).

**Discussion**

Contrary to our hypothesis, our study demonstrated that young, lean, non-diabetic survivors of marasmus and kwashiorkor have similar clamp-derived insulin sensitivity. They were also similar using HOMA-IR, which is considered by many to be a marker of hepatic insulin resistance (11). This is consistent with our prior data that showed no differences in insulin sensitivity using the Matsuda index (7). It is possible that a larger sample may show differences, but if they exist, they are probably small. Since survivors of marasmus are at higher risk of glucose intolerance, these data suggest that beta-cell dysfunction would be the pivotal mechanism (7). Children who develop marasmus have lower birth weight (about 333 g) than those who develop kwashiorkor (4), so developmental factors such as intra-uterine growth restriction may indeed influence beta cell function and/or beta cell mass.

Low birth weight individuals are prone to gain more weight in later life possibly due to altered appetite with higher protein targets (12). Thus, we might expect persons who had marasmus to gain weight more rapidly in an obesogenic environment, develop visceral adiposity and then become insulin resistant in later life. Teleologically, individuals who had marasmus are poorly adapted to environments which expose them to surfeit food. It would be interesting to re-measure insulin sensitivity in these participants several years from now when they may have gained weight, or in obese survivors of malnutrition. Accordingly, Afro-Caribbean children do not show an association of birth weight with insulin sensitivity, but reduced insulin sensitivity is seen in those with faster postnatal weight gain (13).

Peripheral C-peptide levels more accurately reflect pancreatic insulin secretion rates than do peripheral insulin levels (14) but we had no data on C-peptide concentrations. Despite this, our data showed a positive correlation between M/I and insulin clearance. Insulin resistance has been associated with reduced insulin clearance (15) and the latter appears to be a compensatory mechanism to preserve β-cell function and to maintain peripheral insulin levels (16). Reduction in insulin clearance, in addition to augmentation of insulin production, is an important contributor to the compensatory hyperinsulinemia that develops in response to insulin resistance (17). We posit that the similar insulin clearance in MS and KS had might be a reflection of their similar insulin sensitivity.

Notably, in Afro-Caribbean populations, low levels of serum adiponectin may play a causal role in the development of glucose intolerance independent of insulin sensitivity (18). However, there were no differences between the marasmus and kwashiorkor groups and their absolute concentrations are similar to prior normative data in Caribbean populations (18). This suggests the degree of adipocyte inflammation is similar in these lean survivors. We are unaware of any prior data on serum adiponectin in adult survivors of infant malnutrition.

In conclusion, our data showed no differences in insulin sensitivity, insulin clearance or serum adiponectin levels between adult survivors of marasmus and survivors of kwashiorkor. It would be instructive to investigate these relationships in obese survivors of malnutrition.

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**Author contributions:** DST participated in the study design, patient enrollment, data collection and interpretation, and wrote the first draft. PMFE and ATB participated in data collection and interpretation. CO and TRT did statistical analyses and participated in the data interpretation. CDB and MAH participated in the study concept and study design and interpretation of data. PDG participated in the study concept, data interpretation and obtained funding. TEF conceptualized the study, participated in the design, data analysis, and data interpretation, and obtaining funding. MSB participated in the study concept and design, data collection, data analysis and interpretation and acts as guarantor. DST acts as corresponding author and guarantor. All authors revised the report for important intellectual content and approved the final version.

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