Title: β-cell Lipotoxicity in Response to Free Fatty Acid Elevation in Youth: African American versus Caucasian Contrast

Running title: Lipid infusion and insulin secretion

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Word Count: 3167
Tables: 2
Figures: 2

Funding—The study was supported by the National Institutes of Health through Grants R01 HD-27503 (SA), K24 HD-01357 (SA), U1L RR024153 and UL1 TR000005 CTSA, the Richard L. Day Endowed chair (SA) from UPMC, research funds from University of Verona and from Italian Ministry of University and Research PRIN 2008CJ7CTW_002 (RCB).
Abstract

Prepubertal African American (AA) youth compared with their Caucasian (C) peers have higher insulin secretion which correlates positively with free fatty acid (FFA) concentration. In our continued efforts to explain the racial disparity in insulinemia, and because FFAs modulate insulin secretion, we hypothesized that AA youth would have a greater response to FFA-induced \(\beta\)-cell insulin secretion than C youth. We compared the short-term effects of FFA elevation on fasting and glucose-stimulated C-peptide-modeled insulin secretion in prepubertal normal-weight AA versus C peers during a 2-hour hyperglycemic clamp (12.5mmol/L) on two occasions: a) infusion of normal saline (NS) and b) infusion of 20% intralipid (IL). During IL infusion, insulin sensitivity (IS) declined comparably in AA and C youth. Glucose sensitivity of 1\textsuperscript{st} and 2\textsuperscript{nd} phase insulin secretion showed a significant condition x race interaction being higher in AA youth. Disposition index, \(\beta\)-cell function relative to IS, declined with IL infusion in both AA and C youth with a significantly greater decrease in Cs compared with AAs. In conclusion, both AA and C prepubertal youth demonstrated a decline in \(\beta\)-cell function relative to IS during IL infusion, indicative of acute lipotoxicity. The greater decline in C youth compared with AAs may suggest that either C youth are more susceptible to \(\beta\)-cell lipotoxicity than AA youth, or alternatively, AA youth are hypersensitive to FFA stimulation of \(\beta\)-cell insulin secretion consistent with our theory.
Short-term exposure to elevated free fatty acid (FFA) concentrations has a stimulatory effect on β-cell function in *in vitro* and *in vivo* animal and human studies (1-8). On the other hand chronic exposure to sustained elevations in FFAs are shown to decrease insulin secretion and result in β-cell lipotoxicity in *in vitro* rat and human islet experiments (4, 9, 10) and *in vivo* human experiments (1, 5, 7, 11, 12).

We previously found that prepubertal AA youth have higher insulin secretion and an upregulated β-cell function relative to insulin sensitivity (IS) compared with their C peers (13). The hyperinsulinemic response in AA youth appears to be over and above the compensatory response to their lower IS, since the disposition index (DI), which is insulin secretion relative to IS, was ~75% higher in AAs than C youth (13). Furthermore, 1*st* and 2*nd* phase insulin concentrations during the hyperglycemic clamp correlated positively and significantly with fasting FFA levels in AAs but not Cs despite similar plasma FFA concentrations (13). Therefore, we hypothesized that the higher insulin secretion in AA prepubertal youth could be driven by a greater sensitivity to the stimulatory effects of FFA on insulin secretion and β-cell function. Thus, the present investigation examined the effects of FFA elevation on glucose-stimulated insulin secretion in AA and C prepubertal normal-weight children, expecting to observe a heightened response in AA children.

**Research Design and Methods**

Twelve AA and thirteen C prepubertal (Tanner I) normal-weight children participated in this study at the Pediatric Clinical and Translational Research Center (PCTRC) of Children’s Hospital of Pittsburgh after Institutional Review Board approval. Parental consent and child assent was obtained prior to study participation. Children were recruited through local newspaper advertisements, fliers posted on public transportation buses, the health campus and the community. All children were in good health as assessed by a medical history, physical
examination and hematological and biochemical tests (Table 1). None were on any medications known to affect glucose metabolism. Family history of type 2 diabetes was obtained in all participants, and a positive history was defined as the presence of known cases of diagnosed diabetes in any of three generations as described before (14).

**Clamp Studies**

Participants were admitted twice within a 1- to 3-week period to the PCTRC for a control experiment of normal saline infusion (NS) plus hyperglycemic clamp and another for a 20% intralipid infusion (IL) plus hyperglycemic clamp, conducted in random order. Clamps were performed the morning after PCTRC admission following a 10- to 12-hour overnight fast as reported (13).

For each condition, one catheter was inserted into a forearm vein for infusion of NS or IL and 20% dextrose and a second catheter was inserted into the contralateral heated hand vein for sampling of arterialized venous blood. At -180 minutes before the start of the hyperglycemic clamp, the infusion of 0.9% NS or 20% IL (20% soybean oil, 1.2% egg yolk phospholipids, and 2.25% glycerol; Kabi Pharmacia, Clayton, NC) was initiated at 0.02 ml/kg/min and continued until the end of the 2-hour clamp. At 0 min, a 2-hour hyperglycemic (12.5 mmol/L) clamp was performed to assess 1\textsuperscript{st} and 2\textsuperscript{nd} phase insulin and C-peptide as reported previously (13), together with measurement of FFA and triglycerides (TG).

**Body composition and abdominal adiposity**

Height and weight were assessed to the nearest 0.1 cm and 0.1 kg, respectively. Dual-energy x-ray absorptiometry (DEXA) was used to determine fat free mass (FFM), fat mass (FM) and percent body fat (%BF). Subcutaneous (SAT) and visceral adiposity (VAT) were assessed using magnetic resonance imaging (MRI), as before (15). SAT and VAT data are missing for one AA subject due to technical difficulties.

**Biochemical measurements**
At each sampling point, blood samples were placed on ice and plasma was immediately separated in a refrigerated centrifuge. Plasma samples were divided into aliquots and stored at -80°C until analysis. Plasma glucose was determined, at the bed-side, by the glucose oxidase method using a glucose analyzer (Yellow Springs Instrument Co., Yellow Springs, Ohio), and plasma insulin and C-peptide by commercially available radioimmunoassay (Linco catalog no. 1011, St. Charles, MO), as reported by us before (16). FFA and TG were determined using enzymatic colorimetric methods [Wako nonesterified fatty acid (NEFA) C test kit; Wako, Osaka, Japan].

**Calculations**

Fasting data were expressed as the means of -30, -15 and 0 minute samples. First-phase glucose, insulin and C-peptide were the means of 2.5, 5, 7.5, 10 and 12.5 minute samples. First-phase FFA was the mean of 5 and 10 minute samples. Second-phase insulin and C-peptide were the means of eight determinations every 15 minutes from 15 to 120 minutes. Second-phase glucose was the mean of 22 determinations every 5 minutes from 15 to 120 minutes. Second-phase FFA was the mean of 30, 60, 90 and 120 minute samples and second-phase TG was the mean of 60 and 120 minute samples. IS was calculated as the rate of glucose disposal during the last 60 minutes of the hyperglycemic clamp minus urinary glucose excretion, divided by the mean insulin concentration during the same period and multiplied by 100 and expressed as μmol/kg/min per pmol/L (13). An index of basal insulin clearance (units: liter·min⁻¹·m⁻²·body surface area) was computed as the ratio of model-derived (see below) basal insulin secretion rate divided by basal insulin concentration (17). An index of insulin clearance during the hyperglycemic clamp was computed as the ratio of the model-derived area under the curve of insulin secretion rate divided by the area under the curve of insulin concentration during the clamp (18).
Insulin secretion was modeled during the hyperglycemic clamp and described as a summation of three components: 1) a basal (postabsorptive) secretion rate, 2) a dynamic secretion component (glucose sensitivity of 1\textsuperscript{st} phase insulin secretion), herein presented as the amount of insulin secreted in response to a rate of increase in glucose concentration of 1 mmol/liter per 1 min and 3) a static or proportional secretion component (glucose sensitivity of 2\textsuperscript{nd} phase secretion) further boosted by a gain factor (17), herein presented as the steady-state insulin secretion rate in response to a step increase in glucose concentration of 1 mmol/liter (19-22). Parameters were estimated by implementing this minimal model of C-peptide secretion in SAAM-II 1.2 software (SAAM Institute, Seattle, WA). Numerical values of the unknown parameters were estimated by using nonlinear least squares. Weights were chosen optimally, i.e. equal to the inverse of the variance of the measurement errors, which were assumed to be additive, uncorrelated, with zero mean, and a constant coefficient of variation (CV) of 13%. Data are expressed per kg body weight. Results of the analyses did not change when data were expressed relative to body surface area (m\textsuperscript{2}) or fat free mass (FFM). \(\beta\)-cell function relative to IS, i.e. DI, was calculated as the product of IS x glucose sensitivity of 1\textsuperscript{st} or 2\textsuperscript{nd} phase insulin secretion (23).

\textbf{Statistical Analyses}

Data are presented as mean ± SEM. Statistical significance was set at \(p<0.05\). PASW Statistics (version 18, SPSS Inc., Chicago, IL) was used for the statistical analyses. As there are no prior studies examining the effects of IL infusion on insulin secretion in AA vs. C, adult or youth, our sample size was estimated using our previous data that demonstrated a 150\% higher 1\textsuperscript{st} phase insulin in AA compared with C children (13). Power calculations indicated that to detect a 100\% difference between AA and C in the response to IL infusion, 11 subjects would need to complete the study assuming 80\% power and 5\% \(\alpha\) error. To detect a 120\% or a 150\% difference in the response to IL infusion, 11 or 8 subjects, respectively, would need to complete the study. This
assumed a 90% power and 1% α error. Student t-tests were used to determine differences in subject characteristics between the two groups. Non-parametric statistics were used when appropriate. Categorical variables were compared using the $\chi^2$ analysis. A 2X2 repeated measures ANOVA (within subjects factor of condition, i.e., NS vs. IL; between subjects factor of race, i.e., AA vs. C; condition x race interaction) was used to analyze hormone and substrate concentrations. Adjustments for VAT were made in the primary outcome variables of basal insulin secretion rate, β-cell glucose sensitivity of 1\textsuperscript{st} and 2\textsuperscript{nd} phase insulin secretion, IS and DI.

Results

**Participant Characteristics (Table 1)**

Prepubertal AA and C children were similar in age, BMI, FFM, FM, and %BF. AA youth had lower VAT as reported before (13, 24), but similar VAT/SAT ratio.

**Baseline postabsorptive and hyperglycemic clamp hormone and substrate profiles (Table 2)**

Consistent with prior findings AA youth had higher fasting insulin concentration (13) and lower fasting TG concentrations compared with C youth (15). In response to IL infusion, fasting glucose, insulin, C-peptide, FFA and TG increased in both groups. First phase glucose concentrations were slightly higher in Cs compared with AA youth during both NS and IL conditions. IL infusion increased 1\textsuperscript{st} and 2\textsuperscript{nd} phase FFA and TG concentrations similarly in AA and C children. First and 2\textsuperscript{nd} phase insulin and C-peptide concentrations increased with IL infusion but the increase in all was greater in AA than C youth.

**Model-derived parameters of insulin secretion (Figure 1A-C)**

Basal insulin secretion rate increased significantly and similarly with IL infusion in both AAs and Cs (Figure 1A). During NS infusion 1\textsuperscript{st} phase insulin secretion (Figure 1B) was significantly
higher in AAs vs. Cs, consistent with prior findings (13). During IL infusion there was a significant condition x race interaction with the increase in glucose sensitivity of 1\textsuperscript{st} and 2\textsuperscript{nd} phase insulin secretion being higher in AAs (Figure 1B and 1C). After adjusting for VAT, the condition x race interactions remained significant.

Consistent with prior observations of lower insulin clearance in AA youth (13), both basal and clamp insulin clearance were significantly lower in AA than in C children (Table 2). During IL infusion, no significant changes in insulin clearance were detected either in the basal state or during the clamp (Table 2).

**Insulin sensitivity and β-cell function relative to insulin sensitivity, DI (Figure 2A-C)**

During IL infusion IS declined comparably in AA and C children (Figure 2A). During NS infusion 1\textsuperscript{st} phase DI was significantly higher in AAs compared with Cs, consistent with prior observations (13). During IL infusion, DI, β-cell function relative to IS calculated from either modeled 1\textsuperscript{st} or 2\textsuperscript{nd} phase insulin, decreased (Figure 2B-C), with a significantly greater decline in 2\textsuperscript{nd} phase DI in Cs compared with AAs (Figure 2C). After adjusting for VAT, the IL induced decline in DI and the condition x race interaction remained significant.

**Visceral adiposity and intralipid-induced β-cell function and insulin sensitivity**

To assess whether the amount of visceral fat per se, irrespective of race, could predict lipid-induced changes in β-cell function and IS, we analyzed the whole cohort with AAs and Cs combined. Bivariate relationships revealed that VAT correlated with IL-induced change in glucose sensitivity of 2\textsuperscript{nd} phase insulin secretion ($r=-0.43$, $p=0.04$) with no significant correlations observed with any of the other outcome measures. Additionally, we performed multiple regression analyses with VAT, age, race and sex as independent variables and the
change in each of IS, glucose sensitivity of 1\(^{st}\) and 2\(^{nd}\) phase insulin secretion and DI as dependent variables. VAT, age and sex did not contribute to the variance in any of the dependent variables; however, race contributed 20.2% (p=0.03) to the variance in IL-induced change in glucose sensitivity of 1\(^{st}\) phase and 21.5% (p=0.02) of 2\(^{nd}\) phase insulin secretion.

**Discussion**

In our efforts to explain the hyperinsulinemia and the upregulated β-cell function relative to IS in AA youth, we hypothesized that the pancreatic β-cell in AA children is more sensitive to the stimulatory effect of FFA on insulin secretion compared with Cs. Our hypothesis was based on our previous observations that for the same degree of IS, glucose-stimulated insulin secretion is significantly higher in AA vs. C children, and correlated with plasma FFA concentrations (13, 25). In the present study, using C-peptide modeling during a hyperglycemic clamp, a 5-hr IL infusion resulted in a significant decline in DI, β-cell function relative to IS, in both AA and C youth, indicative of acute lipotoxicity. However, there was a significant racial contrast in absolute β-cell response to IL infusion with a greater 1\(^{st}\) and 2\(^{nd}\) phase insulin secretion in AA youth compared with their C peers, and a lesser decline in DI in AA youth. This may suggest that either C youth are more susceptible to β-cell lipotoxicity than AA youth, or alternatively, AA youth are hypersensitive to FFA stimulation of β-cell insulin secretion consistent with our theory.

Most *in vitro* (4) and *in vivo* (2, 3, 6-8) studies, but not all (26), demonstrate that acute FFA elevations increase glucose-stimulated insulin secretion, and is an important component for basal insulin secretion (27, 28). In the present study, we demonstrate a 32% increase in basal insulin secretion in response to FFA elevation. This is in agreement with our previous observation of FFA elevation significantly increasing basal C-peptide by ~28% with no differences between AA and C adolescents (16). Among lean men and women, 6-hrs of intralipid + heparin infusion increased glucose-stimulated insulin secretion by 37%, as assessed via an intravenous glucose
tolerance test (6). In lean women, FFA elevation increased glucose-stimulated (7 mmol/L) plasma insulin concentrations by 30% with further increases observed at higher plasma glucose concentrations (11 mmol/L), although the latter increase was driven by a reduction in endogenous insulin clearance (8). Similarly, in healthy men, during a 2-step hyperglycemic clamp, acute IL infusion increased glucose-stimulated insulin secretion ~3 ½ fold during 10 mmol/L plasma glucose concentration with further increases observed during 20 mmol/L plasma glucose concentration (7). We are not aware of literature reports on race-related differences in response to IL infusion and β-cell function. Our investigation demonstrates that while AAs responded to IL infusion with a 15% and 32% increase in 1st and 2nd phase insulin secretion, Cs demonstrated a decrease of 7% and 15% in 1st and 2nd phase insulin secretion. The difference in the response to intralipid infusion between AA and Cs occurred despite adjusting for their level of VAT. In further support of a role of FFA in β-cell insulin secretion is the observation that treatment of fasted adults with nicotinic acid, which suppresses lipolysis and FFA, resulted in a decline in stimulated-insulin area under the curve (28). The greater decline in obese (70%) vs. normal-weight individuals (40-50%) was suggestive of a greater dependency on circulating FFAs for glucose-stimulated insulin secretion in obese than normal-weight adults. Our present findings would suggest that this could be the case in AA youth who might have a greater dependency on circulating FFA for glucose-stimulated insulin secretion compared with Cs. Furthermore, our prior findings of a significant positive relationship between fasting FFA concentrations and 1st phase insulin secretion in AA, with no such relationship in C (13), lends further support to such a theory. It remains to be determined if suppression of FFA levels may demonstrate a racial contrast in FFA dependency of β-cell insulin secretion.

We and others have shown that infusion of IL and/or elevation of plasma FFA concentration is associated with a decline in in vivo insulin sensitivity (2, 3, 7, 16). In a prior study using a hyperinsulinemic-euglycemic clamp together with IL infusion, we found that the
decline in IS was similar between AA and C adolescents (16). The findings from the current study in prepubertal children are consistent with that in adolescents. Since FFAs induce insulin resistance, expressing 1\textsuperscript{st} and 2\textsuperscript{nd} phase insulin secretion in absolute terms without consideration of IS may not provide a complete picture of the dynamic relationship between the decline in IS and the change in β-cell function consequent to IL infusion. Therefore, in addition, we elected to express insulin secretion relative to IS, or the DI. We originally anticipated that DI would remain comparable to the NS condition as insulin secretion would compensate for the decline in IS. Contrary to our expectation, the present data demonstrate that β-cell function relative to IS declines during IL vs. NS infusion in both AA and C youth, suggestive of a lipotoxic phenomenon. This seems to be more pronounced in C (39\% and 44\% decline in 1\textsuperscript{st} and 2\textsuperscript{nd} phase DI) than in AA youth (22\% and 12\% decline). This is the first demonstration that a β-cell lipotoxic phenomenon might be operational in young prepubertal youth. The potential ramification of such an observation is the adverse impact that high fat foods might have on β-cell function starting early in life and progressing with aging, and racial predisposition to β-cell dysfunction driven by dietary factors. We previously demonstrated that increased fat/carbohydrate ratio in the diet of prepubertal youth is associated positively with 1\textsuperscript{st} phase insulin secretion and negatively with IS (13).

The strengths of this investigation include a) the paired design of IL vs. NS conditions in the same youth participant, b) the use of the “gold standard” hyperglycemic clamp to determine \textit{in vivo} insulin secretion, and c) modeling of insulin secretion from C-peptide data which overcomes race–related differences in insulin clearance (13, 29). A potential limitation could be that both IS and insulin secretion and DI were derived from a single hyperglycemic clamp instead of two separate clamps, a hyperinsulinemic-euglycemic clamp for IS and a hyperglycemic clamp for insulin secretion. However, we recently demonstrated, in 330 youth, that IS from the hyperglycemic clamp correlates with IS from the euglycemic clamp (r=0.90, p<
0.001), and DI derived from a single hyperglycemic clamp correlates with DI from two clamps, a euglycemic clamp and a hyperglycemic clamp \((r=0.85, p<0.001)\) (23); thus supporting the use of a single hyperglycemic clamp (23). Further, a single clamp approach is similar to the IVGTT approach from which both IS and acute insulin response are measured, and DI is calculated (30). Another potential limitation of our study is that we could not assess the impact of family history of type 2 diabetes on β-cell response to FFA elevations because our study was not powered to address this. However, there was no difference between AAs and Cs with respect to family history of type 2 diabetes (Table 1). Others have demonstrated that family history of diabetes is associated with an enhanced susceptibility to lipotoxicity in adults (12). Additional studies in youth are needed to assess if this familial risk is present early in life.

In conclusion, in the present study both AA and C prepubertal youth demonstrate a decline in β-cell function relative to insulin sensitivity with IL infusion, indicative of acute lipotoxicity. The greater decline in C compared with AA youth may suggest that either C youth are more susceptible to β-cell lipotoxicity than AA youth, or alternatively, AA youth are hypersensitive to FFA stimulation of β-cell insulin secretion consistent with our theory. Whether the latter is reflective of a greater dependency of glucose-stimulated insulin secretion on circulating FFA in AAs remains to be determined. Lastly, it remains to be investigated if dietary FFAs might induce similar adverse changes in β-cell function in youth.

**Author Contribution**

SFM first authored the manuscript, and contributed to the data analyses and interpretation; RCB provided C-peptide modeling analysis and critically reviewed/edited the manuscript; LAS, SJL contributed to data interpretation, reviewed and edited the manuscript; LF maintained the database and contributed data analysis; SA provided the study concept and design, acquired data,
obtained funding, provided administrative, technical and material support, supervised the study and critically reviewed/edited the manuscript.

Acknowledgements

SA is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. None of the authors report any conflict of interest with respect to this work. We would like to thank all the children and their parents who participated in this study, without whom science would not advance. We are grateful to the nursing staff of the Pediatric Clinical and Translational Research Center for their outstanding care of the participants and meticulous attention to the research, to Nancy Guerra, Children’s Hospital of Pittsburgh, University of Pittsburgh Medical Center, for her assistance with clamp experiments and to Resa Stauffer, Children’s Hospital of Pittsburgh, University of Pittsburgh Medical Center, for her laboratory analytical contributions.
References


4. Sako Y, Grill VE. A 48-hour lipid infusion in the rat time-dependently inhibits glucose-induced insulin secretion and B cell oxidation through a process likely coupled to fatty acid oxidation. Endocrinology 1990;127:1580-9


Table 1—Physical characteristics of the participants

<table>
<thead>
<tr>
<th></th>
<th>African American</th>
<th>Caucasian</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>12</td>
<td>13</td>
<td></td>
</tr>
<tr>
<td>Age (yrs)</td>
<td>9.5 ± 0.3</td>
<td>9.9 ± 0.4</td>
<td>ns</td>
</tr>
<tr>
<td>Sex (male/female)</td>
<td>6/6</td>
<td>5/8</td>
<td>ns</td>
</tr>
<tr>
<td>Family history of T2DM (+/-)</td>
<td>7/4</td>
<td>6/6</td>
<td>ns</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>138.8 ± 2.4</td>
<td>136.5 ± 2.1</td>
<td>ns</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>31.9 ± 1.3</td>
<td>32.8 ± 1.6</td>
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<tr>
<td>BMI (kg/m²)</td>
<td>16.5 ± 0.4</td>
<td>17.5 ± 0.4</td>
<td>ns</td>
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<tr>
<td>BMI (%)</td>
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<td>59.0 ± 5.3</td>
<td>ns</td>
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<tr>
<td>FFM (kg)</td>
<td>23.8 ± 0.7</td>
<td>23.2 ± 1.1</td>
<td>ns</td>
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<tr>
<td>Fat mass (kg)</td>
<td>6.2 ± 0.7</td>
<td>7.8 ± 0.7</td>
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<tr>
<td>Body fat (%)</td>
<td>19.5 ± 1.5</td>
<td>24.1 ± 1.7</td>
<td>ns</td>
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<tr>
<td>Visceral adipose tissue (cm²)</td>
<td>11.9 ± 2.0</td>
<td>19.5 ± 2.3</td>
<td>0.03</td>
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<tr>
<td>Subcutaneous adipose tissue (cm²)</td>
<td>49.2 ± 4.6</td>
<td>73.5 ± 7.1</td>
<td>0.01</td>
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<td>Visceral:Subcutaneous adipose tissue</td>
<td>0.26 ± 0.05</td>
<td>0.27 ± 0.03</td>
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</table>

T2DM = type 2 diabetes; FFM = fat free mass; ns = not significant; one AA is missing family history of T2DM and visceral/subcutaneous adipose tissue data. All subjects are Tanner I puberty.
Table 2—Hormone and substrate concentrations

<table>
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<th>African American</th>
<th>Caucasian</th>
<th>P, ANOVA</th>
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<tbody>
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<td></td>
<td>Normal Saline</td>
<td>Intralipid</td>
<td>Normal Saline</td>
</tr>
<tr>
<td><strong>Fasting Measurements</strong></td>
<td></td>
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<tr>
<td>FFA (mmol/L)</td>
<td>0.23 ± 0.04</td>
<td>0.72 ± 0.10</td>
<td>0.20 ± 0.04</td>
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<tr>
<td>Glucose (mmol/L)</td>
<td>5.0 ± 0.1</td>
<td>5.2 ± 0.1</td>
<td>5.2 ± 0.1</td>
</tr>
<tr>
<td>Insulin (pmol/L)</td>
<td>77.5 ± 6.9</td>
<td>104.1 ± 9.0</td>
<td>59.9 ± 7.7</td>
</tr>
<tr>
<td>C-peptide (nmol/L)</td>
<td>0.43 ± 0.04</td>
<td>0.56 ± 0.04</td>
<td>0.48 ± 0.05</td>
</tr>
<tr>
<td>Triglycerides (mmol/L)</td>
<td>0.63 ± 0.12</td>
<td>3.2 ± 0.3</td>
<td>0.81 ± 0.08</td>
</tr>
<tr>
<td><strong>Basal Insulin Clearance</strong></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>(liter min⁻¹ m⁻² BSA)</td>
<td>0.95 ± 0.10</td>
<td>0.93 ± 0.08</td>
<td>1.43 ± 0.10</td>
</tr>
<tr>
<td><strong>Clamp Measurements</strong></td>
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<td></td>
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<tr>
<td>1ˢᵗ ph FFA (mmol/L)</td>
<td>0.21 ± 0.04</td>
<td>0.61 ± 0.10</td>
<td>0.21 ± 0.04</td>
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<tr>
<td>1ˢᵗ ph Glucose (mmol/L)</td>
<td>12.3 ± 0.2</td>
<td>12.3 ± 0.1</td>
<td>12.8 ± 0.2</td>
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<tr>
<td>1ˢᵗ ph Insulin (pmol/L)</td>
<td>495.2 ± 44.6</td>
<td>679.0 ± 73.8</td>
<td>330.8 ± 42.9</td>
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<tr>
<td>1ˢᵗ ph C-peptide (nmol/L)</td>
<td>1.45 ± 0.1</td>
<td>1.78 ± 0.2</td>
<td>1.30 ± 0.1</td>
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<tr>
<td>1ˢᵗ ph Triglycerides (mmol/L)</td>
<td>0.63 ± 0.13</td>
<td>3.0 ± 0.27</td>
<td>0.78 ± 0.08</td>
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<tr>
<td>2ⁿᵈ ph FFA (mmol/L)</td>
<td>0.08 ± 0.02</td>
<td>0.53 ± 0.10</td>
<td>0.04 ± 0.02</td>
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<tr>
<td>2ⁿᵈ ph Glucose (mmol/L)</td>
<td>12.4 ± 0.05</td>
<td>12.4 ± 0.04</td>
<td>12.3 ± 0.04</td>
</tr>
<tr>
<td>2ⁿᵈ ph Insulin (pmol/L)</td>
<td>595.8 ± 65.8</td>
<td>738.4 ± 77.9</td>
<td>418.3 ± 63.2</td>
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<tr>
<td>2ⁿᵈ ph C-peptide (nmol/L)</td>
<td>2.19 ± 0.2</td>
<td>2.53 ± 0.3</td>
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<tr>
<td>2ⁿᵈ ph Triglycerides (mmol/L)</td>
<td>0.57 ± 0.10</td>
<td>3.0 ± 0.3</td>
<td>0.79 ± 0.12</td>
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<tr>
<td>Clamp Insulin Clearance*</td>
<td>0.81 ± 0.03</td>
<td>0.74 ± 0.04</td>
<td>1.16 ± 0.08</td>
</tr>
</tbody>
</table>

1ˢᵗ ph = first-phase; 2ⁿᵈ ph = second-phase; FFA = free fatty acids; ns = not significant. * P<0.01 Clamp insulin clearance vs. basal insulin clearance
Figure legends

Figure 1. Basal insulin secretion rate (A), glucose sensitivity of first-phase insulin secretion (B), and glucose sensitivity of second-phase insulin secretion (C) in African American (AA) and Caucasian (C) children during infusion of normal saline (NS) (empty bars) or 20% intralipid (IL) (filled bars). Condition=Main effect of condition, normal saline vs. intralipid. Race=Main effect of race, AA vs. C. Condition x Race= Condition x Race interaction. NS = not significant

Figure 2. Insulin sensitivity (A), disposition index from first-phase insulin secretion (B), and disposition index from second-phase insulin secretion (C) in African American (AA) and Caucasian (C) children during infusion of normal saline (NS) (empty bars) or 20% intralipid (IL) (filled bars). Condition=Main effect of condition, normal saline vs. intralipid. Race=Main effect of race, AA vs. C. Condition x Race= Condition x Race interaction. NS = not significant
Glucose Sensitivity of 1st phase Insulin Secretion (pmol/kg per mmol/L/min)

Glucose Sensitivity of 2nd phase Insulin Secretion (pmol/min/kg per mmol/L)

Basal Insulin Secretion Rate (pmol/kg/min)

Condition P<0.001
Race P=0.65
Condition x Race P=0.39

Condition P=0.61
Race P=0.02
Condition x Race P=0.03

Condition P=0.95
Race P=0.36
Condition x Race P=0.002
Insulin Sensitivity (µmol/kg/min per pmol/L)  

Condition P<0.001  
Race P=0.81  
Condition x Race P=0.69

First Phase DI

Condition P<0.001  
Race P=0.04  
Condition x Race P=0.27

Second Phase DI

Condition P<0.001  
Race P=0.60  
Condition x Race P=0.01