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Does inflammation mediate the association between obesity and insulin resistance?

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Abstract

In adult obesity low-grade systemic inflammation is considered an important step in the pathogenesis of insulin resistance (IR). The association between obesity and inflammation is less well established in adolescents. Here, we ascertain the importance of inflammation in IR among obese adolescents by utilizing either Random Forest (RF) classification or mediation analysis approaches. The inflammation balance score, composed of 8 pro- and anti-inflammatory makers, as well as most of the individual inflammatory markers differed significantly between lean and overweight/obese. In contrast, adiponectin was the only individual marker selected as a predictor of IR by RF, and the balance score only revealed a medium to low importance score. Neither adiponectin nor the inflammation balance score were found to mediate the relationship between obesity and IR. These findings do not support the premise that low grade systemic inflammation is a key for the expression of IR in the human. Prospective longitudinal studies should confirm these findings.

Keywords

Insulin Resistance; Inflammation; Inflammation Balance Score; Obesity; Random Forest

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INTRODUCTION

Visceral adipose tissue, which is composed of adipocyte cells, blood vessels, and macrophages—is now known to be an endocrine organ and to secrete a number of pro- and anti-inflammatory biochemical products, collectively known as “adipokines.” Obesity is associated with low-grade systemic inflammation [1], often referred to as “metabolic inflammation”, and is believed to be an important step in the pathogenesis of human insulin resistance (IR) [2]. Other factors such as sleep deprivation[3], genetic factors [4], certain medication (e.g. corticosteroids) [5], and impairment of the pituitary-adrenal axis may lead to IR as well.” The hypothesis that metabolic inflammation is causally linked to IR is supported by clinical evidence of correlations between inflammatory markers and measures of IR [6] and also by biochemical evidence indicating that pro-inflammatory markers can interfere with insulin action by directly inhibiting insulin receptors [7]. Further, animals made obese with high fat diets can be protected from developing IR by blocking inflammatory pathways [8]. However, causal inference is not yet possible since most of the human evidence is based on correlational data from adult subject groups. As articulated by Makki et al. in a recent review on obesity-related inflammation and IR, there persists a “lingering uncertainty” in our understandings of the link between inflammation (particularly TNF- α) and IR in the human [2]. To gain further understanding of these relationships, it may be useful to study adolescents, a group that is less likely to have other obesity-associated comorbidities. Furthermore, the role of C-reactive protein (CRP), interleukin 6 (IL-6), tumor necrosis factor α (TNF α), interleukin 10 (IL-10), interleukin 4 (IL-4), and macrophage infiltration in adolescent obesity—insofar as they are presently understood—are partly inconsistent with the results of adult human data and rodent models currently in place [9, 10].

The present study sought to test whether inflammation was required for the expression of IR by evaluating a relatively large group of community-residing non-clinical adolescents. We used an advanced recursive partitioning technique, Random Forests (RF), to ascertain the relative predictive importance of anthropometric measures, lipid profiles, clinical data, and pro- and anti-inflammatory markers (as well as a weighted balance of them) to predict the presence of IR. Specifically, we sought to determine whether, among obese adolescents, inflammatory markers are:

1. Elevated compared to lean adolescents?
2. Correlated with anthropometric measures?
3. Important predictors of IR?
4. Have mediatory effect in the path of obesity to IR?

METHODS

The data used in this study is comprised of subjects studied in two parallel studies conducted by the Brain, Obesity, and Diabetes Lab and approved by the institutional review board (IRB) at the New York University School of Medicine. The first study, the IR in Adolescents (OIA) study, is a clinical lab-based study involving a battery of medical and brain

assessments designed to uncover associations between IR and brain structure and function. All study participants were community-residing non-clinical adolescents of predominantly Hispanic and African American origin. The second study is the Banishing Obesity and Diabetes in Youth (BODY) Project, a school based medical screening and education program of obesity which was held between 2007 and 2014, also predominantly Hispanic and African American in origin. Parental consent and participant assent was obtained from participants under age 18, and participant consent from those 18 years of age. All participants were compensated for their time and inconvenience.

Inclusion Criteria

In the BODY Project, students were in grades 9–12 at 5 public New York City high schools. Pregnancy and a known diagnosis of diabetes were the only exclusion criteria. There were 486 students who had a sample collected for possible inflammatory assay analysis. These students spanned the whole spectrum of BMI ranging from lean to obese, but students carrying excess weight were oversampled. From these subjects a subset that included all overweight and obese students ($N = 163$) as well as a group of lean students ($N = 82$) well matched to those carrying excess weight on age, gender, and race/ethnicity had their blood assayed and included in these analyses.

For the OIA study, which included obese and lean adolescents, participants were recruited predominantly through online advertisements. Subjects with significant medical (other than those known to be associated with obesity) or psychiatric conditions were excluded. We collected data on 102 students all of whom were evaluated for inflammatory markers.

All together a total of 347 lean, overweight, and obese subjects were included. Two BODY Project subjects were excluded because clinical laboratory values suggested non-fasting. Thus, a total of 345 subjects (51.6% female) were included in the final analyses.

Anthropometric Measurements and BMI

Height, weight, and waist circumference were measured using standardized methods. BMI was calculated as weight (kg) divided by height (m^2), and WHR was computed as waist circumference (cm) divided by height (cm). Body composition was measured using Quantum analyzers (RJL Systems) by the bioelectrical impedance method [11] and used to establish PBF.

For participants aged 18 years and older, BMI category was assigned based on accepted Centers for Disease Control and Prevention cut-off values ($<25 \text{ kg/m}^2$ for lean, $25\text{--}30 \text{ kg/m}^2$ for overweight and $>30 \text{ kg/m}^2$ for obese). For subjects under 18, weight group assignment was based on BMI percentiles for age and sex based on CDC values ($<85^{\text{th}}$ percentile lean, 85^{th} – 94.9^{th} overweight and $\geq 95^{\text{th}}$ obese).

Blood Chemistry Measurements

Following a 10–12 hour overnight fast, blood was collected from all participants in a standardized fashion. Blood tests, including glucose and insulin levels, lipid profile, hemoglobin A1C (HbA1c), and CRP, were performed at the NYU Medical Center Clinical

Laboratories. Samples to be assayed for inflammatory markers were collected in EDTA tubes, placed on ice, spun down at 4°C, aliquoted and stored at -80°C. The cytokine assays were performed on the stored aliquots by the Immune Monitoring Core at the New York University Langone Medical Center.

Cytokine Measurements and Estimation of the Beta Weights for the Inflammation Balance Score

We measured CRP, Fibrinogen, TNF- α , IL-4, IL-6, and IL-10, interferon gamma (IFN- γ), resistin, and adiponectin.

The inflammatory process is the result of the balance between pro- and anti-inflammatory interactions. Given the lack of guidance in the literature on how to weigh the different pro- and anti-inflammatory markers in creating an inflammation balance score, we decided to take a statistical approach. After excluding individual marker values that were more than three standard deviations from the mean of the lean ("normal") group, we utilized the resultant mean and standard deviation to compute Z-scores for each of the individual pro- and anti-inflammatory markers for our adolescents. Then using a binary logistic regression approach predicting weight group membership (lean vs. overweight/obese) with the Z-scores for the individual pro- and anti-inflammatory markers as independent variables, we obtained the beta weights for the markers. Overall, the logistic regression model significantly predicted group membership in 309 adolescents with a set of eight pro- and anti-inflammatory markers, ($X^2[8, n=309] = 125.06, P < .0001$). The relationship between group membership and prediction was moderate (Nagelkerke's $R^2 = 0.461$) with an overall prediction success at 79.9% with better prediction among overweight/obese (sensitivity=85.9%) than lean adolescents (specificity=68.3%). The Wald test demonstrated that CRP ($P < .0001$) and adiponectin ($P = .001$) significantly contributed to the prediction of group membership, while resistin and TNF- α (both at $P = .06$) were both trending towards significance (Table 1). These beta weights were then utilized to create a weighted algebraic sum (with pro-inflammatory having a positive and anti-inflammatory a negative sign) that reflected a balance of those markers.

Estimation of Insulin Resistance

Fasting glucose and insulin levels were used to compute the homeostatic model assessment of IR (HOMA-IR) score as follows: $HOMA-IR = [Glucose (mg/dl) \times Insulin (uIU/ml)] / 405$. As previously described in adolescent populations we and others have used a HOMA-IR 3.99 as the cut point for IR [12, 13].

Statistical Analyses

Statistical methods were specifically chosen to address the four questions raised in the introduction.

Group mean differences were compared using t-tests for continuous variables and χ^2 tests for categorical variables. Non-parametric tests were used when variables were non-normally distributed. The step-down test was performed to control for multiple comparisons [14]. SPSS software (v. 20; SPSS Inc, Chicago, IL) was used to obtain correlation coefficients and

to test whether they differed from zero. Any value for an inflammatory marker that was above or below 3 standard deviations from the group mean was excluded variable-wise from the analyses. Differences between groups were considered significantly different at $P < 0.05$. We measured Cohen's d effect sizes through differences between two means divided by the standard deviation. $d = 0.2$ was considered small effect size, $d = 0.5$ medium effect size and $d = 0.8$ large effect size.

We utilized a nonparametric recursive partitioning classification method, RF, to determine a function for classification of adolescents carrying excess weight into those with and without IR and to determine the importance of either the individual inflammatory markers or an inflammation balance score, made up of a weighted algebraic sum of the individual pro- and anti-inflammatory markers. RF, created by Breiman in 2001, is an extension of recursive partitioning that grows multiple trees rather than just one [15, 16], with the largest extent possible and no pruning. In the current study, the number of random predictors selected in each node was set to be the square root of all the predictors and 500 trees (the default setting) were run to produce the forest. In the RF analyses predictor variables included anthropometric data (BMI, PBF, and WHR), clinical data (age, sex, race), lipid profile (HDL, LDL, triglyceride (TG), total cholesterol), Hb A1c, and Z-scores of the inflammatory markers (adiponectin, CRP, IFN- γ , IL-10, IL-4, IL-6, resistin, and TNF- α), for a total of 19 predictors. The target variable was IR status (yes/no), as defined by a HOMA value of ≥ 3.99 ; non-IR was < 3.99 .

We used the PROCESS add-on (developed by Andrew, F Hayes) for SPSS to run mediation analysis. PROCESS uses a logistic regression-based path analytic framework for estimating direct and indirect effects in simple and multiple mediator models. The indirect effect was evaluated with the Sobel test [17] and the bootstrapping method with 5000 bootstrapped samples at 95% confidence interval [18].

RESULTS

Demographic and Anthropometric Data

The 345 subjects were, on average, 17.5 ± 1.77 years of age (Table 2). The lean and overweight/obese groups were well matched on age, sex, and ethnicity. The lean group ($N = 118$) had an average BMI of 21.6 ± 1.72 kg/m² and the overweight/obese group ($N = 227$) had an average BMI of 33.3 ± 5.34 kg/m². As expected, the group carrying excess weight had significantly larger WHRs, higher levels of IR, higher cholesterol, TG, LDL, lower HDL and higher Mean BP levels; all mean group differences with the exception of Cholesterol and HbA1c, which showed small to medium effect sizes, showed medium to large effect sizes (Table 2).

We also divided the overweight/obese group into those with and without IR and their characteristics are depicted in Table 3.

Inflammation Data

Of the various anti- and pro-inflammatory markers when considered singly, the group means only differed significantly for adiponectin, CRP, IL-6, resistin, and TNF- α . There were no

group differences in IFN- γ , IL-10, or IL-4 (Table 4). All of these simple group analyses showed higher pro-inflammatory and lower anti-inflammatory marker levels for the group of adolescents carrying excess weight. These group differences remained significant after using the step-down test to control for multiple comparisons. With that said, in the group comparison (lean vs. overweight/obese) only the inflammation balance score and CRP showed large effect size ($d = 1.09$ and 0.97 respectively), adiponectin showed a medium to large ($d = -0.67$) effect size and TNF- α , IL-6 and resistin showed small to medium effect sizes ($d = 0.42, 0.47$ and 0.34 respectively).

Table 5 describes the inflammatory markers for the overweight/obese group separated by those with and without IR.

Random Forest

RF was performed on the 227 overweight/obese participants, with the target of IR defined by a HOMA-IR ≥ 3.99 ($N_1 = 113$) or < 3.99 ($N_2 = 114$). All other variables, including anthropometric measurements, blood markers and inflammatory factors Z-scores were used as the classifiers. The variables selected as important predictors were those with importance scores ≥ 30 . We ran RF twice; both times the demographic variables, anthropometric measures, cholesterol profile variables, and HbA1c were used and either the individual inflammatory markers Z-scores or the inflammation composite balance score added.

In the first model, which included the demographic variables, anthropometric measurements, cholesterol profile markers, HbA1c and the Z-scores for each of the individual inflammatory blood markers, adiponectin Z score was the most important predictor of IR status with a score of 100. BMI, TG, age, PBF and WHR were selected as the next most important variables with scores of 68, 65, 44, 42 and 35 respectively (Figure 1). None of the other individual inflammatory markers were important in the classification. The overall out-of-bag specificity of this predictive model was 71.93% and the sensitivity was 75.22%.

In the second model, which included the anthropometric measurements, cholesterol profile markers, HbA1c and the inflammation composite balance score, TG was the most important predictor followed by BMI, age, PBF, WHR, Hb A1c and inflammation composite balance score. The order of importance scores were as follows: 100, 78, 73, 55, 41, 38 and 32. The overall out-of-bag specificity of the model was 73.67% and the sensitivity was 69.91%. The composite inflammation balance score was not an important variable in the classification (Figure 2).

Mediation analysis

We tested adiponectin and the composite inflammation balance score as potential mediators of the relationship between obesity and IR. Both total and direct effect of obesity on IR were significant ($P < 0.0001$), however, the indirect effect was not significant for neither the composite inflammation score ($P = 0.601$), nor adiponectin ($P = 0.243$).

DISCUSSION

In our study population, adolescents carrying excess weight (overweight/obese) demonstrated a significantly greater degree of IR, had more abnormalities in all anthropometric parameters and laboratory values, and generally had higher levels of low grade systemic pro-inflammatory markers, albeit with only modest effect sizes (relative to their matched lean counterparts). We found significant group differences in Inflammation Composite Score and CRP (large effect sizes), adiponectin (medium to large effect size), and TNF- α , IL-6 and resistin (small to medium effect sizes). The other pro- or anti-inflammatory markers did not differ by obesity group.

This study, utilizing the unbiased classification approach of RF, suggests that pro-inflammatory cytokines, although elevated among youth carrying excess weight (relative to lean healthy demographically matched adolescents), appear not to be important determinants of IR. Composite inflammation balance score appeared merely as the 7th predictor of IR in the order of importance in the second RF model and adiponectin was the only individual inflammatory marker whose decreases were important in the classification of IR. Further these inflammatory predictors were not confirmed to be mediators of the relationship between BMI and IR. Although the link between BMI and IR remains unsettled, with some previous studies suggesting that inflammation is the link between obesity and IR [19] and others indicating uncertainty in the role of inflammation as a mediator of this association [2], in this study we failed to find support for the role of low grade systemic inflammation as a mediator of IR in obesity.

Obesity, Inflammation and IR

The results of this study significantly contribute to our understanding of the relationships between obesity, inflammation, and IR among adolescent.

Only adiponectin, CRP, IL-6, resistin, and TNF- α differed statistically between the adolescent weight groups, and of these only CRP had a large effect size; the other cytokines had medium to large or small to medium effect sizes. However, the composite inflammation balance score, which is composed of a weighted algebraic sum of the pro- and anti-inflammatory markers, did differ significantly between the weight groups with a large effect size. Of the individual inflammatory markers only adiponectin, an anti-inflammatory cytokine and therefore negatively related to IR, was picked as an important predictor of IR among overweight and obese adolescents, and the inflammation balance score with a relatively low importance score (32, with the most important being triglyceride level with an importance score of 100) was selected as an important predictor of IR in the second model. Furthermore neither adiponectin nor inflammation balance score were confirmed to mediate the relationship between BMI and IR.

The important relationship between adiponectin and IR is supported by previous studies that have shown adiponectin levels to be inversely correlated with measures of IR [20, 21]. Further, Yamauchi et al reported that physiological doses of adiponectin improve IR in mouse models of obesity and type 2 diabetes by increasing expression of molecules involved in both fatty-acid metabolism and thus lowering TG content in muscle and liver [22].

Additionally, by studying both wild-type and type 2 diabetic mice, Berg et al reported that an acute increase in adiponectin concentration sensitizes the body to insulin by causing a decrease in basal glucose level through inhibition of hepatic gluconeogenesis [23]. Adiponectin aside, our study does not support the commonly held premise that other inflammatory factors such as IL-6 [24] TNF- α [19] or CRP [25] play an important role in IR.

Due to the high level of interaction between anti-inflammatory and pro-inflammatory markers looking at the behavior of individual inflammatory markers might be misleading. A composite inflammation balance score may be a more appropriate estimate of an inflammatory status in obesity, and it turned out to have a slight predictive role in the classification of IR in overweight and obese adolescents. This finding demonstrates that the overall level of inflammation contributes to the prediction of IR, but only after other more crucial non-inflammatory blood markers and anthropometric measurements. This lack of importance for role of inflammation on the presence of IR in adolescents was also confirmed by the negative mediation analysis results.

The fact that CRP, which has a strong associations to elevated BMI ($r=0.567$ in our data), does not appear as a useful predictor of insulin resistant status merits discussion. Our present findings and a large body of literature support a strong association between CRP and BMI, but do not support a link to IR independent of BMI. This may be the case because CRP is produced predominantly by the liver, which clearly contrasts with the adipose tissue macrophages responsible for the production of obesity-associated inflammatory cytokines.

Strengths and Limitations

The major strength of this study is that it is in the human, thus removing the need to extrapolate from rodent data to understand the human physiology.

The RF modeling technique used in this analysis has many advantages over traditional statistical techniques. It can create models that account for both linear and non-linear relationships, it can accommodate for missing data, and, most importantly, it can reveal complex, non-additive relationships between multiple predictor variables. To our knowledge, RF has not been utilized in any study relating the predictive role of various anthropometric and biochemical measures on IR.

The main limitation of this study is that it is based on a sample of opportunity, with study subjects not selected randomly. Therefore our ability to generalize beyond inner city predominantly minority adolescents is somewhat limited. Another important limitation is the fact that to estimate the degree of IR we rely on a non-dynamic estimate of that parameter, HOMA-IR, which is based on fasting glucose and insulin levels. With that said, HOMA is a robust surrogate index of human IR that is strongly correlated with the results of the hyperinsulinemic-euglycemic clamp technique in both non-diabetic and diabetic subjects [26]. Further, HOMA determination is relatively inexpensive, non-invasive, and more acceptable to young people than clamp studies, and thus more appropriate for larger studies.

CONCLUSION

Our results, failing to confirm a potential “causal” role of low grade inflammation in IR, suggest that mouse models may not effectively recapitulate the human physiology and that there is great need to utilize human data and avail ourselves of state of the art unbiased, model free methods, such as RF, to predict IR. Although the current data and approach has some limitations, our analysis provides valuable insights about the relative importance of anthropometrics, lipids, clinical data, and inflammatory biomarkers on the prediction of IR. Ascertaining the relationships between obesity, inflammation, and IR in adolescents is important not only because of the current lack of understanding in this area, but also because in this group these relationships may be somewhat clearer as adolescents generally do not have manifest co-existent conditions, such as cardiovascular disease, which are commonly found among obese adults and could confound the findings. However and most importantly, unlike middle-aged obese individuals, who often show co-occurring IR, inflammation, hypertension, and lipid abnormalities, studying adolescents offers the ability to observe obesity-specific inflammation in its early stages, at a time when it can be dissociated from IR and most importantly at a time when possible interventions could be most impactful.

The results of this study suggest that low grade inflammation does not appear to play a critical role in the development of IR in adolescents and young adults. However, this does not preclude a more important role for inflammation in contributing to IR later in adulthood. Also the current study focused on the roles of the eight inflammatory markers CRP, adiponectin, resistin, IFN- γ , TNF- α , IL-4, IL-6, IL-10 and their composite score in the development of IR during adolescences. This does not exclude any potential role of other inflammatory markers in contributing to the development of IR in adolescences.

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Variable	Score	
ADIPONECTIN	100.0000	
BODY MASS INDEX	68.7138	
TRIGLYCERIDE	65.6484	
AGE	44.6955	
% BODY FAT	42.1032	
WAIST-TO-HEIGHT RATIO	35.7267	

Figure 1.

Random Forest variable importance, model (1): with HOMA-IR as the target and demographic information, anthropometric measurements and individual inflammatory markers Z score as predictors

Variable	Score	
TRIGLYCERIDE	100.0000	
BODY MASS INDEX	78.5280	
AGE	73.6587	
% BODY FAT	55.9198	
WAIST-TO-HEIGHT RATIO	41.7397	
HEMOGLOBIN A1C	38.2259	
COMPOSITE INFLAMMATION BALANCE SCORE	32.7886	

Figure 2.

Random Forest variable importance, model (2): with HOMA-IR as the target and demographic information, anthropometric measurements and Inflammation Composite Balance Score as predictors

Table 1

Results of logistic regression model predicting group membership

Inflammatory Markers	Beta Coefficient	Standard Error	Wald Test	P-value	Odds Ratio
CRP	0.790	0.15	26.33	0.000	2.20
Adiponectin	-0.617	0.18	11.90	0.001	0.54
TNF- α	0.308	0.16	3.61	0.06	1.36
Resistin	0.299	0.16	3.66	0.06	1.35
IL-10	-0.224	0.15	2.16	0.14	0.80
IL-6	0.224	0.18	1.51	0.22	1.25
IFN- γ	-0.207	0.20	1.05	0.31	0.81
IL-4	0.089	0.18	0.23	0.63	1.09

CRP (C - reactive protein), IL-4, 6, 10 (Interleukin 4, 6, 10), IFN- γ (Interferon-gamma), TNF- α (Tumor Necrosis Factor alpha)

Table 2

Demographic information, clinical characteristics and non-inflammatory blood markers in lean vs. overweight/obese

	Lean	Overweight/Obese	P value	Effect Size
Basic Information				
Sex (F/M)	60/58	105/122		
Race (%)				
Asian	9 (8%)	8 (4%)		
Black	33 (28%)	58 (26%)		
Hispanic	66 (56%)	135 (60%)		
White	5 (4%)	12 (5%)		
Age	17.6 (1.54)	17.51 (1.88)	0.641	−0.05
Anthropometric Measurements				
BMI				
WHR	0.44 (0.04)	0.60 (0.08)	0.000	2.22
PBF	24.99 (7.16)	39.36 (7.72)	0.000	1.91
Mean BP	80.12 (8.17)	86.42 (9.11)	0.000	0.72
Blood Measurements				
TG	61.86 (22.85)	88.87 (42.48)	0.000	0.73
Cholesterol	150.31 (24.82)	162.67 (30.84)	0.000	0.43
LDL	82.32 (21.69)	98.24 (26.58)	0.000	0.63
HDL	55.83 (12.41)	46.98 (10.75)	0.000	−0.78
HbA1C	5.34 (0.39)	5.46 (0.33)	0.003	0.34
HOMA	1.65 (1.08)	4.24 (2.94)	0.000	1.05

BMI (Body Mass Index), WHR (Waist to Height Ratio), PBF (Percent Body Fat), Mean BP (Mean Blood Pressure), TG (Triglyceride), LDL (Low Density Lipoprotein), HDL (High Density Lipoprotein), HbA1c (Hemoglobin A1C), HOMA (Homeostatic Model Assessment)

Table 3

Demographic information, clinical characteristics and non-inflammatory blood markers in non- IR overweight/obese vs. IR overweight/obese

	Non-Insulin Resistant	Insulin Resistant	P-Value	Effect Size
Basic Information				
Sex (F/M)	51/63	54/59		
Race (%)				
Asian	4 (4%)	4 (4%)		
Black	30 (26%)	28 (25%)		
Hispanic	66 (58%)	69 (62%)		
White	9 (8%)	3 (3%)		
Age	18.01 (2.03)	17.00 (1.57)	0.000	−0.56
Anthropometric Measurements				
BMI	31.67 (4.67)	34.93 (5.49)	0.000	0.64
WHR	0.58 (0.08)	0.63 (0.07)	0.000	0.57
PBF	37.38 (8.46)	41.54 (6.14)	0.000	0.56
Mean BP	84.15 (8.66)	88.71 (9.01)	0.000	0.52
Blood Measurements				
TG	77.05 (37.03)	100.80 (44.40)	0.000	0.58
Cholesterol	159.44 (32.95)	165.93 (28.33)	0.113	0.21
LDL	95.08 (28.58)	101.41 (24.35)	0.075	0.24
HDL	49.59 (11.89)	44.35 (8.76)	0.000	−.050
HbA1C	5.37 (0.30)	5.55 (0.35)	0.000	0.55

BMI (Body Mass Index), WHR (Waist to Height Ratio), PBF (Percent Body Fat), TG (Triglyceride), LDL (Low Density Lipoprotein), HDL (High Density Lipoprotein), HbA1c (Hemoglobin A1C), HOMA (Homeostatic Model Assessment)

Table 4

raw inflammatory markers, Z-score inflammatory markers and Inflammation Composite Score in Lean vs. Overweight/Obese

Raw Inflammatory Markers	Lean	Overweight/Obese	P value	Effect Size
CRP	0.60 (0.94)	3.86 (4.16)	0.000	0.95
Adiponectin	21792.22 (11915.94)	14590.43 (9984.44)	0.000	-0.67
IL-4	8.98 (24.05)	9.48 (24.38)	0.859	0.02
IL-6	2.16 (3.06)	4.01 (4.32)	0.000	0.47
IL-10	13.17 (12.82)	14.48 (21.36)	0.547	0.07
Resistin	46.61 (20.14)	53.87 (21.99)	0.003	0.34
IFN-γ	3.78 (10.30)	3.24 (7.66)	0.585	-0.06
TNF-α	5.33 (2.60)	6.52 (2.92)	0.000	0.42
Z-scores Inflammatory Markers				
Z- CRP	0.00 (1.00)	3.47 (4.42)		
Z- Adiponectin	0.00 (1.00)	-.60 (0.83)		
Z-IL-4	0.00 (1.00)	0.02 (1.01)		
Z-IL-6	0.00 (1.00)	0.60 (1.41)		
Z-IL-10	0.00 (1.00)	0.10 (1.66)		
Z-Resistin	0.00 (1.00)	0.36 (1.09)		
Z- IFN-γ	0.00 (1.00)	-.05 (0.74)		
Z- TNF-α	0.00 (1.00)	0.45 (1.12)		
Composite Inflammation Balance Score	0.00 (0.15)	0.44 (0.48)	0.000	1.09

CRP (C - reactive protein), IL-4, 6, 10 (Interleukin 4, 6, 10), IFN- γ (Interferon-gamma), TNF- α (Tumor Necrosis Factor alpha)

Table 5

raw inflammatory markers, Z-score inflammatory markers and Inflammation Composite Score in Non-IR Overweight/Obese vs. IR Overweight/Obese

	Non-Insulin Resistant	Insulin Resistant	P-Value	Effect Size
Raw Inflammatory Markers				
CRP	3.40 (4.07)	4.33 (4.22)	0.091	0.23
Adiponectin	18.689.79 (11574.21)	10672.46 (6011.98)	0.000	-0.88
IL-4	7.83 (23.99)	11.13 (24.77)	0.310	0.14
IL-6	3.32 (4.33)	4.70 (4.21)	0.017	0.32
IL-10	10.79 (14.754)	18.23 (25.99)	0.009	0.35
Resistin	55.02 (22.95)	52.71 (21.01)	0.435	-0.10
IFN-γ	2.81 (7.34)	3.68 (7.99)	0.392	0.11
TNF-α	5.90 (2.63)	7.15 (3.08)	0.001	0.44
Z-scores Inflammatory Markers				
Z- CRP	2.98 (4.32)	3.97 (4.49)		
Z- Adiponectin	-0.26 (0.97)	-0.93 (0.50)		
Z-IL-4	-0.04 (0.99)	0.08 (1.03)		
Z-IL-6	0.38 (1.41)	0.83 (1.37)		
Z-IL-10	-0.18 (1.15)	0.39 (2.02)		
Z-Resistin	0.41 (1.13)	0.30 (1.04)		
Z- IFN-γ	-0.09 (0.71)	.00 (0.77)		
Z- TNF-α	0.22 (1.01)	0.70 (1.18)		
Composite Inflammation Balance Score	0.36 (0.48)	0.51 (0.48)		

CRP (C - reactive protein), IL-4, 6, 10 (Interleukin 4, 6, 10), IFN- γ (Interferon-gamma), TNF- α (Tumor Necrosis Factor alpha)