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Inflammatory Markers Among African American Adolescents with Type 2 Diabetes Mellitus and Obesity: A Cross-Sectional Study

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Abstract

Background/Objectives: Type 2 diabetes (T2D), a chronic metabolic disorder characterized by systemic inflammation, disproportionately affects African American adolescents. Metformin may reduce inflammation beyond glycemic control; however, its impact on inflammatory markers in adolescents remains unclear. We hypothesized that inflammatory markers would differ across groups defined by diabetes, obesity, and metformin use. Methods: In this cross-sectional analysis, inflammatory markers (C-reactive protein [CRP], interleukin-6 [IL-6], and tumor necrosis factor-alpha [TNF-α]), metabolic panels, and metformin exposure were assessed in African American adolescents from Children's of Alabama. Simple and multivariate linear regression models were used to test associations between metformin use and inflammatory markers, adjusting for age, sex, and BMI z-score. Results: Among 78 adolescents, metformin use was reported in 73.6% of those with T2D + obesity and 27.3% of those with obesity-only. In the T2D + obesity group, metformin use was associated with TNF-α ($\beta = 0.34$, $p = 0.02$). Conclusions: Metformin use was associated with higher levels of specific inflammatory markers, potentially reflecting that metformin was more likely utilized in more severe diseases. Longitudinal studies are needed to disentangle treatment effects from underlying disease progression.

Keywords: type 2 diabetes; inflammatory markers; adolescents; metformin

1. Introduction

Type 2 diabetes (T2D) is a metabolic disorder arising from insulin resistance (IR) and relative insulin deficiency in the absence of autoimmune beta-cell destruction [1]. While IR and hormonal changes of puberty can precipitate the development of T2D, they do not fully explain the rapid disease progression observed in adolescents compared to adults. The increasing prevalence of T2D, particularly among individuals with obesity, underscores the need to understand how therapeutic agents, such as metformin, influence inflammatory processes that may drive disease progression [2]. Identifying biological mechanisms beyond lifestyle determinants is essential to improve long-term outcomes in this high-risk population.

Inflammation plays a pivotal role in the development and progression of T2D [3]. For example, high-sensitivity C-reactive protein (CRP) has been identified in both adult and adolescent populations [4,5]. Pro-inflammatory cytokines such as tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6) are increased in individuals with IR before the onset of

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T2D and gestational diabetes [6–10]. TNF-α promotes IR by phosphorylating the insulin receptor [11], while IL-6 triggers hepatic synthesis of CRP and inhibits insulin signaling [11]. Several studies have confirmed these findings, highlighting the role of pro-inflammatory cytokines in IR and T2D development [12–16]. Given the varying presentations of T2D, particularly among individuals with and without obesity, understanding how inflammation differs across these groups is essential for targeted interventions.

Metformin, the first-line pharmacologic therapy for T2D, impacts glucose levels by decreasing gluconeogenesis from the liver [17–19] and improving tissue insulin sensitivity [20,21]. It has been shown to decrease nuclear factor κβ, a key regulator of inflammation [22,23]. Other studies indicate that metformin use is associated with reductions in CRP levels [24–26], suggesting that its benefits extend beyond glycemic control to modulating inflammatory pathways. It has been shown to improve cardiovascular disease (CVD) outcomes via improving inflammation as well [27,28]. The magnitude and consistency of these anti-inflammatory effects remain unclear, particularly in adolescents and across obesity phenotypes. Investigating inflammatory markers in African American adolescents with varying metabolic risk profiles yields critical insights into their role in mitigating disease progression and comorbidities in this population.

While African American individuals represent 14.4% of the population [29], African Americans are underrepresented with a prevalence to participation ratio of 0.36–0.72 [30]. This underrepresentation makes it difficult to generalize existing findings of inflammation and T2D in African American adolescents. The prevalence of T2D in African American adolescents has increased from 0.98 in 1000 people to 1.8 per 1000 people in the past 20 years, representing a dramatic increase [2]. Including a population highly impacted by the disease is therefore critical to ensure that insights into the disease pathology and response to treatment apply to all populations. Our study aimed to address this knowledge gap by focusing exclusively on African American adolescents, providing valuable data on inflammatory and metabolic characteristics in a group often excluded from mechanistic studies of diabetes.

In this study, the primary objective was to evaluate whether inflammatory markers differed across three groups (African American adolescents with either T2D + obesity or obesity only, or normal-weight controls). We hypothesized that inflammatory markers would differ across groups defined by obesity phenotype and that metformin use would be associated with lower levels of inflammatory markers after adjusting for age and sex. Understanding these relationships may help guide future longitudinal studies of metformin use and its impact on disease progression and inflammatory response to treatment in this high-risk population.

2. Materials and Methods

2.1. Study Design and Settings

The study was conducted using data collected during a single study visit for each participant. All procedures were completed at Children's of Alabama, between January 2022 and February 2023, as previously described [31]. Eligible participants completed the study assessments during one in-person visit with no longitudinal follow-up.

2.2. Inclusion/Exclusion Criteria

Participants were self-reported race-identified African American adolescents aged 12–18 years who were classified into three groups: (1) adolescents with T2D + obesity (n = 19), (2) adolescents with obesity only (n = 43), and (3) normal-weight, non-diabetic controls (n = 15). Inclusion criteria in the T2D + obesity group were body mass index (BMI) > 95th percentile for age and sex, a confirmed diagnosis of T2D based on a history

of hemoglobin A1c (HbA1c) > 6.5% at diagnosis and negative pancreatic antibodies, and no history of a monogenic cause of obesity. Participants in the obesity only group met the following criteria: HbA1c < 6.5%, BMI > 90th percentile for age and sex, and no history of a monogenic cause of obesity. Normal-weight controls had HbA1c < 5.6%, BMI < 90th percentile, and no history of a monogenic cause of obesity.

No formal power calculation was performed because this was an exploratory analysis. Recruitment targeted the available population within the study period to assess feasibility and identify potential associations to inform potential for future longitudinal work. The study was conducted following the ethical principles recommended by the Declaration of Helsinki. The Institutional Review Board (IRB) at the University of Alabama at Birmingham (UAB) approved this study, IRB approval number 3000010930. Written informed consent and assent were obtained from all participants and their legal guardians.

2.3. Recruitment Protocol

Recruitment was conducted by the research team at the UAB. The primary investigator trained the research team on the procedures and communication with patients and families. Potential eligible participants were identified through electronic medical records and contacted via phone to collect information based on age, ethnicity, and medical diagnosis. Adolescents were recruited from outpatient clinics at Children's of Alabama, either in the general adolescent medicine clinic, weight management clinic, or pediatric endocrine clinic. Once eligibility was confirmed, participants and their guardians received detailed study information and instructions for participation.

2.4. Data Collection

Demographic and anthropometric data was collected including age, sex, weight z-score, height z-score, and BMI z-score, which were collected from the electronic medical record after being calculated based on the normative data from the Center for Disease Control and Prevention for age and sex-specific weight, height, and BMI, HbA1c, lipid panel, AST, ALT, IL-6, TNF-alpha, and CRP. HbA1c was measured to evaluate glycemic control using a DCA Vantage Analyzer and Hemoglobin A1c reagent kit, which estimates average blood glucose during the past 2–3 months. Lipid panels, including total cholesterol, low-density lipoprotein (LDL), triglycerides, and high-density lipoprotein (HDL), were analyzed using the Abbot Architect ci8200 system. The limit of detection for total cholesterol was 6.2 mg/dL, with an assay imprecision <3% of the Total Coefficient of Variation (CV). For LDL, the detection limit was <10 mg/dL, with interassay and intraassay CVs of 2% and 1.1–14%, respectively. The limit of quantitation was 5.0 mg/dL, and the detection limit for HDL was 2.5 mg/dL with an interassay CV of 0.5–1.1%, and intraassay CV of 1.0–1.7%. IL-6 (Interleukin-6), TNF-alpha, and CRP were measured at the UAB Nutrition Obesity Research Center Metabolism Core with ELISA immunoassays.

2.5. Statistical Analysis

Descriptive statistics were used to summarize the baseline characteristics of participants by all three groups (normal weight, obesity only, and T2D + obesity and by two groups (obesity only and T2D + obesity). Categorical variables were presented as frequencies and percentages, while continuous variables were reported as means and standard deviation (SD). Differences in baseline characteristics among groups were assessed using one-way analysis of variance (ANOVA) for continuous variables and chi-square tests for categorical variables. Pairwise comparisons were conducted using Tukey's post hoc test to identify specific group differences.

Inflammatory markers (CRP, IL-6, and TNF-α) were assessed for normality using the Shapiro–Wilk test. Homoscedasticity was also assessed using Levene's and Brown–

Forsythe tests. Due to the highly skewed nature of biomarkers (<0.05), each biomarker was transformed using the natural log formula log(value + 0.0001) to improve normality.

Simple and multivariate linear regression models were used to determine the association between metformin use and each log-transformed inflammatory marker among participants with obesity only and T2D + obesity, given the study objective to assess whether T2D modifies the relationship between metformin use and inflammatory markers. Simple linear regression models were employed to assess the relationship with inflammatory markers individually. Stratified multivariate and simple linear regression analyses were conducted to further investigate the association based on group differences (obesity only and T2D + obesity). Correlation analyses were conducted to assess the relationship between potential confounders of the association between metformin use and inflammatory markers among the obesity only and T2D + obesity groups. All regression models were adjusted for age, sex, and BMI z-score. The normal-weight group was excluded from all regression analyses, given that we were interested in looking at the impact of the diagnosis of T2D on the changes seen in inflammatory markers. No prior power calculation was performed due to the exploratory nature and limited sample size. Given the small study sample size, subgroup analysis will be underpowered to allow for the detection of moderate-sized differences. Statistical significance was set at alpha <0.05, and all analyses were conducted using SAS 9.4.

2.6. Outcomes and Variables

The outcomes of interest were circulating levels of IL-6, TNF-α, and CRP. Key exposure variables included weight category (normal weight, obesity only, and T2D + obesity) and use of metformin. Additional confounding variables considered in adjusted models included sex and age. Secondary clinical variables for descriptive and exploratory analysis may include HbA1c, AST, ALT, and lipid panel components.

3. Results

The total study population included 78 African American adolescents, with baseline characteristics summarized in Table 1. The mean age across all participants was 14.8 years (±1.8), and 64.1% of participants were female. Metformin use was reported in 27.3% of participants in the obesity only group and 73.6% in the T2D + obesity group; no participants in the normal-weight control group reported metformin use.

ANOVA with post hoc Tukey's group comparisons of significant and/or clinically important characteristics and markers are shown in Table 2. Participants in the obesity only and T2D + obesity groups had significantly higher weight z-scores, height z-scores, BMI z-scores, and lower HDL compared to those in the normal-weight group ($p < 0.001$). Additionally, triglycerides and fasting glucose were significantly different between the T2D + obesity group and the normal-weight group. HbA1c levels were significantly different between the obesity only and normal-weight groups. However, no significant differences were observed in inflammatory markers or metabolic markers such as AST, ALT, A1C, total cholesterol, and LDL.

In multivariate regression models, metformin use was not associated with inflammatory markers in participants with obesity only or in those with concurrent T2D + obesity (Table 3). In separate analyses of individual inflammatory markers, metformin use was not associated with CRP, TNF-α, or IL-6 (Supplementary Materials: Tables S1–S3).

Table 1. Characteristics of participants by group (Normal Weight, Obesity, T2D + Obesity) (n = 78).

	Total (n = 78)	Normal Weight (n = 15)	Obesity Only (n = 44)	T2D + Obesity (n = 19)	*p*-Value [1]
Demographics					
Age (years)	14.8 (1.8)	14.8 (1.5)	14.8 (1.9)	14.7 (1.9)	0.96
Female, n (%)	50 (64.1)	9 (60.0)	28 (63.6)	13 (68.4)	0.87
Weight z-score	2.3 (1.3)	−0.2 (0.9)	2.8 (0.7)	2.7 (0.5)	<0.01
BMI z-score	2.2 (1.0)	0.0 (0.9)	2.6 (0.3)	2.4 (0.3)	<0.01
Metabolic Markers					
ALT (U/L)	21.5 (21.7)	12.8 (4.6)	20.7 (11.4)	30.0 (39.0)	0.07
Total cholesterol (mg/dL)	156.2 (33.3)	148.4 (27.6)	160.2 (35.5)	153.1 (32.2)	0.45
LDL (mg/dL)	103.0 (38.0)	87.9 (24.6)	109.5 (42.8)	100.4 (32.6)	0.16
HDL (mg/dL)	41.6 (11.5)	51.1 (10.6)	40.3 (10.5)	37.1 (10.8)	<0.01
Triglycerides (mg/dL)	85.0 (65.4)	55.4 (16.5)	77.7 (34.7)	124.8 (111.9)	<0.01
Glycemic Markers					
Fasting glucose (mg/dL)	106.9 (53.8)	91.3 (7.8)	90.1 (11.5)	158.0 (91.4)	<0.01
Hemoglobin A1c (%)	6.2 (1.6)	5.5 (0.4)	5.7 (0.5)	7.8 (2.6)	<0.01
Inflammatory Markers					
CRP (mg/L)	8.2 (7.6)	7.4 (6.7)	9.2 (8.4)	5.6 (4.5)	0.29
TNF-α (pg/mL)	1.6 (0.6)	1.9 (0.9)	1.6 (0.4)	1.4 (0.4)	0.07
IL-6 (pg/mL)	1.8 (2.1)	1.2 (1.3)	1.9 (1.9)	1.9 (2.9)	0.46
Use of Metformin					
Yes, n (%)	26.0 (33.3)	0.0 (0.0)	12.0 (27.3)	14.0 (73.6)	<0.01
No, n (%)	52.0 (66.7)	15.0 (19.2)	32.0 (72.7)	5.0 (26.4)	

Notes: Mean (SD) unless stated otherwise. [1] Significance level at alpha <0.05. Abbreviations: BMI, body mass index; AST, aspartate transaminase; ALT, alanine transaminase; LDL, low-density lipoprotein; HDL, high-density lipoprotein; CRP, C-reactive protein; TNF-α, tumor necrosis factor-alpha; IL-6, interleukin-6; T2D, type 2 diabetes.

Table 2. One-way ANOVA with Tukey's post hoc group comparison of metabolic changes and inflammatory markers relative to normal-weight control, obesity only, and T2D + obesity groups (n = 78).

Variables	One-Way ANOVA		Post Hoc Test (Tukey)		
	F-Value	*p*-Value	Group Comparison	Mean Difference	95% CI
Weight z-score	84.5	<0.01	2–1 3–1	3.0 2.9	2.4, 3.5 2.3, 3.6
Height z-score	4.0	0.02	3–1	1.2	0.3, 2.2
BMI z-score	149.8	<0.01	2–1 3–1	2.5 2.4	2.2, 2.9 2.0, 2.8
HDL (mg/dL)	8.0	<0.01	2–1 3-1	−10.7 −14.0	−18.4, −3.1 −22.7, −5.2
Triglycerides (mg/dL)	6.0	<0.01	2–3 3–1	−47.1 69.4	−87.6, −6.6 18.8, 120.2
Fasting glucose (mg/dL)	15.7	<0.01	2–3 3–1	−67.9 66.7	−97.9, −37.9 28.9, 104.5
Hemoglobin A1c (%)	18.9	<0.01	2–3	−2.1	−3.0, −1.2
			3–1	2.3	1.2, 3.4

Abbreviations: CI, confidence interval; BMI, body mass index; HDL, high-density lipoprotein. Notes: Group codes: 1 = normal-weight control, 2 = obesity only, 3 = T2D + obesity.

In linear regression analyses, metformin use was negatively associated with TNF-α in the T2D + obesity group (Table 4). Metformin use was not associated with CRP, TNF-α, or IL-6 in the obesity only group; data note shown (Supplementary Materials: Tables S4–S6). No significant associations were observed for CRP or IL-6 in the adolescents with T2D + obesity group (Supplementary Materials: Tables S7 and S8). A multivariable linear regression including metformin use showed no association between AST, ALT, and inflammatory markers, looking at the obesity-only group and T2D + obesity together,

but CRP was positively associated with AST and ALT in the obesity only group. This difference could be secondary to the fact that the groups are small (Supplementary Materials: Table S9). If metformin is not included in the model, CRP was positively associated with AST and ALT in the obesity only group (Supplementary Materials: Table S10). No significant associations were observed in any of the three groups between lipid profiles (total cholesterol, LDL, and triglycerides) and inflammatory markers (Supplementary Materials: Tables S11 and S12). There was an association between CRP and lipid panel components in the T2D + obesity groups (Supplementary Materials: Table S12). Looking at Pearson correlation (Supplementary Materials: Table S13), there was no significant correlation with metformin use and inflammatory markers, but there was a significant correlation with A1c and liver enzymes.

Table 3. The association between metformin use and inflammatory markers among those with obesity and T2D + obesity (n = 63).

Effect	df	Wilks' Lambda	f Value	*p*
[1] Obesity and T2D + Obesity				
Metformin use	3	0.9	1.2	0.33
Age	3	1.0	0.2	0.88
Sex *	3	0.8	4.0	0.01
BMI z-score	3	1.0	0.8	0.48
[2] Obesity only				
Metformin use	3	0.9	1.9	0.15
Age	3	1.0	0.3	0.82
Sex *	3	0.7	4.3	0.01
BMI z-score	3	0.9	0.7	0.56
[3] T2D + Obesity				
Metformin use	3	0.4	3.9	0.06
Age	3	0.6	1.4	0.33
Sex *	3	0.6	1.7	0.25
BMI z-score	3	0.5	2.0	0.20

* Male as the referent group. Multivariate regression models were used to test these associations. [1] Model: Metformin use (among those with obesity only and T2D + obesity) and inflammatory markers (CRP, TNF-α, and IL-6), controlling for age, sex, and BMI z-score. [2] Model: Metformin use (among those with obesity only) and inflammatory markers (CRP, TNF-α, and IL-6) controlling for age, sex, and BMI z-score. [3] Model: Metformin use (among those with T2D + obesity) and inflammatory markers (CRP, TNF-α, and IL-6), controlling for age, sex, and BMI z-score. Abbreviations: CRP, C-reactive protein; TNF- α, tumor necrosis factor-alpha; IL-6, interleukin-6; T2D, Type 2 diabetes.

Table 4. The association between metformin use and TNF-α among those with T2D + obesity (n = 19).

Effect	*β*	SE	95% CI		*p*
			LL	UL	
Metformin use	0.34	0.12	0.08	0.59	0.02
Age	0.05	0.03	−0.01	0.11	0.12
Sex *	0.11	0.14	−0.17	0.41	0.40
BMI z-score	0.25	0.19	−0.16	0.66	0.21

* Male as the referent group. Linear regression models were used to test these associations. Model: Metformin use and TNF-α, controlling for age, sex, and BMI z-score. Bold *p*-values denote an alpha < 0.05. Abbreviations: Body mass index, BMI; TNF-α, tumor necrosis factor-alpha; T2D, Type 2 diabetes.

4. Discussion

Our study examined clinical and metabolic differences among adolescents with T2D + obesity, adolescents with obesity only, and normal-weight controls, an age group for which there are limited studies. Participants with T2D + obesity had significantly lower HDL and higher triglycerides compared to normal-weight controls. This study demonstrated no significant difference in biomarkers of inflammation across the three groups. (Table 1).

Differences in dyslipidemia between the three groups were consistent with prior studies reporting dyslipidemia in youth with insulin resistance and T2D [32–34]. We also assessed differences in inflammatory markers among the three groups, but found no significant differences. These findings contrast with previous studies that reported elevated markers of inflammation, including CRP, TNF-α, IL-5, IL-1β, and IL-6 in individuals with insulin resistance [35–40] and Type 2 diabetes [41–45]. The lack of significant differences in our study may reflect our limited sample size or the heterogeneity of disease severity. The discrepancy between our findings and prior studies may also be due to confounding by the indication of starting metformin, as metformin is more likely to be prescribed to individuals with more advanced metabolic dysfunction. Given that metformin is prescribed to patients with more severe insulin resistance, those patients may inherently have higher levels of inflammation regardless of treatment. These findings may reflect that adolescents with higher inflammation or worse metabolic profiles were more likely to receive metformin.

Metformin has been shown in both adult and animal models to have anti-inflammatory effects, suppressing IL-1β, IL-6, and TNF-α [46–48] and enhancing IL-10 and activation of AMP-activated protein kinase [49,50]. In metabolic syndrome, metformin has been shown to improve adipokines and lower TNF-α, IL-6, and CRP [51–53]. In patients with obesity on metformin, there has also been improvement in BMI and insulin sensitivity, as well as improvement in serum IL-6 and TNF-α [54]. Interestingly, metformin was not shown to decrease inflammatory markers in early-onset T2D in the Lancet trial, which is in line with our findings, given that our recruited participants were early in their course of disease [55]. To further investigate the role of metformin in inflammation in patients with T2D, we compared inflammatory markers in adolescents with T2D + obesity and obesity only, stratified by metformin use. In our study, there was no significant association between inflammatory markers in the obesity only or the T2D + obesity group (Supplementary Materials: Table S11). These differential findings suggest that the relationship between metformin and inflammatory markers may be multi-factorial and depend on diabetes status and duration of use of metformin and dosing, underscoring the need for future research. The lack of statistically significant differences in inflammatory markers across groups should not be interpreted as evidence of no clinically relevant differences, given the small sample size of this study. The study was a pilot study and thus should be interpreted as a hypothesis-generating study, indicating the need for larger prospective studies.

Higher inflammatory markers in patients with T2D + obesity place them at an increased risk of CVD. Patients with increased CRP have been associated with increased waist circumference and increased cardiovascular risk [56]. Elevated levels of inflammatory markers, such as TNF-α, fetuin A, and FGF-21, have also been shown to be tied to pancreatic beta-cell failure [35] and other complications [57]. A meta-analysis of 13 randomized controlled trials showed lower CRP but did not show sustained lower levels of IL-6 and TNF-α with metformin use [24]. Longitudinal studies are needed to assess whether inflammatory markers can serve as predictors of beta-cell decline and future comorbidities. Establishing protocols using these markers may help identify adolescents at greater risk for disease progression or need for insulin therapy.

A strength of this study is its focus on an understudied population—African American adolescents with T2D + obesity—from a pediatric endocrinology clinic in the southern United States. This population is disproportionately affected by early-onset T2D, making it critical to understand metabolic and inflammatory differences in this group. However, our study has limitations. The cross-sectional design limits causal inference, and the small sample size may have reduced power to detect significant differences. There is also a potential for the indication for use of metformin with dysglycemia. Information regarding metformin duration was also not collected, given that the study was a cross-sectional study design. This should be explored in further research studies. Physical activity, dietary intake, environmental factors, and psychosocial stressors should also have been considered in the study, as they could also influence inflammatory markers; therefore, in further larger studies, these factors should also be included [58]. Another limitation of the analysis was that the study was underpowered to detect modest differences, and multiple comparisons increase the risk of type I error. Therefore, these analyses should be considered hypothesis-generating rather than confirmatory. Additionally, the single-site recruitment may restrict generalizability. Future studies with larger, multi-site cohorts and longitudinal designs are needed to explore the temporal relationships between metformin use, inflammation, and disease progression.

This study provides insights into the clinical and metabolic characteristics of African American adolescents with T2D + obesity, obesity only, and normal-weight controls. While no significant differences in inflammatory markers were observed between groups, we identified associations between CRP and metformin use in the obesity only group and TNF-α and metformin use in the T2D + obesity group. These results highlight the complexity of inflammatory regulation in youth with metabolic dysfunction and emphasize the importance of future longitudinal studies to clarify metformin's role in managing inflammation in this population.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/diabetology7010013/s1. Table S1: The association between metformin use and CRP among those with Obesity and T2D + Obesity (n = 63); Table S2: The association between metformin use and TNF-α among those with Obesity and T2D + Obesity (n = 63); Table S3: The association between metformin use and IL-6 among those with Obesity and T2D + obesity (n = 63); Table S4: The association between metformin use and CRP among those with Obesity only (n = 44); Table S5: The association between metformin use and TNF-α among those with Obesity only (n = 44); Table S6: The association between metformin use and IL-6 among those with Obesity only (n = 44); Table S7: The association between metformin use and CRP among those with T2D + Obesity (n = 19); Table S8: The association between metformin use and IL-6 among those with T2D + Obesity (n = 19). Table S9: The association between metformin use and inflammatory markers and metabolic markers (AST and ALT); Table S10: The association between inflammatory markers and metabolic markers (AST and ALT); Table S11: The association between inflammatory markers and metabolic markers (cholesterol, LDL, and triglycerides); Table S12: The association between inflammatory markers and metabolic markers (cholesterol, LDL, and triglycerides); Table S13: Pearson correlation matrix for metformin use, metabolic markers, glycemic markers, and inflammatory markers among those with Obesity only and T2D + Obesity (n = 63).

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Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki and approved by the Institutional Review Board of the University of Alabama at Birmingham (protocol code IRB-300010930, approved 7 May 2023).

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study. Written informed consent has been obtained from the patient(s) to publish this paper.

Data Availability Statement: The original contributions presented in this study are included in the article/Supplementary Materials. Further inquiries can be directed to the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

T2D	Type 2 diabetes
CRP	C-reactive protein
TNF-α	Tumor-necrosis factor-α
IL-6	Interleukin-6

References

1. American Diabetes, A. Diagnosis and classification of diabetes mellitus. *Diabetes Care* **2010**, *33*, S62–S69. [CrossRef] [PubMed]
2. Lawrence, J.M.; Divers, J.; Isom, S.; Saydah, S.; Imperatore, G.; Pihoker, C.; Marcovina, S.M.; Mayer-Davis, E.J.; Hamman, R.F.; Dolan, L.; et al. Trends in Prevalence of Type 1 and Type 2 Diabetes in Children and Adolescents in the US, 2001-2017. *JAMA* **2021**, *326*, 717–727. [CrossRef] [PubMed]
3. Donath, M.Y.; Shoelson, S.E. Type 2 diabetes as an inflammatory disease. *Nat. Rev. Immunol.* **2011**, *11*, 98–107. [CrossRef]
4. Lucas, R.; Parikh, S.J.; Sridhar, S.; Guo, D.-H.; Bhagatwala, J.; Dong, Y.; Caldwell, R.; Mellor, A.; Caldwell, W.; Zhu, H.; et al. Cytokine profiling of young overweight and obese female African American adults with prediabetes. *Cytokine* **2013**, *64*, 310–315. [CrossRef]
5. Reinehr, T.; Karges, B.; Meissner, T.; Wiegand, S.; Stoffel-Wagner, B.; Holl, R.W.; Woelfle, J. Inflammatory Markers in Obese Adolescents with Type 2 Diabetes and Their Relationship to Hepatokines and Adipokines. *J. Pediatr.* **2016**, *173*, 131–135. [CrossRef]
6. Wang, X.; Bao, W.; Liu, J.; OuYang, Y.Y.; Wang, D.; Rong, S.; Xiao, X.; Shan, Z.L.; Zhang, Y.; Yao, P.; et al. Inflammatory markers and risk of type 2 diabetes: A systematic review and meta-analysis. *Diabetes Care* **2013**, *36*, 166–175. [CrossRef]
7. Abell, S.K.; De Courten, B.; Boyle, J.A.; Teede, H.J. Inflammatory and Other Biomarkers: Role in Pathophysiology and Prediction of Gestational Diabetes Mellitus. *Int. J. Mol. Sci.* **2015**, *16*, 13442–13473. [CrossRef]
8. Rehman, K.; Akash, M.S.H.; Liaqat, A.; Kamal, S.; Qadir, M.I.; Rasul, A. Role of Interleukin-6 in Development of Insulin Resistance and Type 2 Diabetes Mellitus. *Crit. Rev. Eukaryot. Gene Expr.* **2017**, *27*, 229–236. [CrossRef]
9. Pestel, J.; Blangero, F.; Watson, J.; Pirola, L.; Eljaafari, A. Adipokines in obesity and metabolic-related-diseases. *Biochimie* **2023**, *212*, 48–59. [CrossRef]
10. Akbari, M.; Hassan-Zadeh, V. IL-6 signalling pathways and the development of type 2 diabetes. *Inflammopharmacology* **2018**, *26*, 685–698. [CrossRef] [PubMed]
11. Smitka, K.; Maresova, D. Adipose Tissue as an Endocrine Organ: An Update on Pro-inflammatory and Anti-inflammatory Microenvironment. *Prague Med. Rep.* **2015**, *116*, 87–111. [CrossRef]
12. Gungor, N.; Thompson, T.; Sutton-Tyrrell, K.; Janosky, J.; Arslanian, S. Early signs of cardiovascular disease in youth with obesity and type 2 diabetes. *Diabetes Care* **2005**, *28*, 1219–1221. [CrossRef] [PubMed]
13. Roth, C.L.; Kratz, M.; Ralston, M.M.; Reinehr, T. Changes in adipose-derived inflammatory cytokines and chemokines after successful lifestyle intervention in obese children. *Metabolism* **2011**, *60*, 445–452. [CrossRef]
14. Su, S.C.; Pei, D.; Hsieh, C.-H.; Hsiao, F.-C.; Wu, C.-Z.; Hung, Y.-J. Circulating pro-inflammatory cytokines and adiponectin in young men with type 2 diabetes. *Acta Diabetol.* **2011**, *48*, 113–119. [CrossRef]
15. Nadeau, K.J.; Zeitler, P.S.; Bauer, T.A.; Brown, M.S.; Dorosz, J.L.; Draznin, B.; Reusch, J.E.B.; Regensteiner, J.G. Insulin resistance in adolescents with type 2 diabetes is associated with impaired exercise capacity. *J. Clin. Endocrinol. Metab.* **2009**, *94*, 3687–3695. [CrossRef]
16. Tanti, J.F.; Jager, J. Cellular mechanisms of insulin resistance: Role of stress-regulated serine kinases and insulin receptor substrates (IRS) serine phosphorylation. *Curr. Opin. Pharmacol.* **2009**, *9*, 753–762. [CrossRef]

17. Hundal, R.S.; Krssak, M.; Dufour, S.; Laurent, D.; Lebon, V.; Chandramouli, V.; Inzucchi, S.E.; Schumann, W.C.; Petersen, K.F.; Landau, B.R.; et al. Mechanism by which metformin reduces glucose production in type 2 diabetes. *Diabetes* **2000**, *49*, 2063–2069. [CrossRef] [PubMed]
18. DeFronzo, R.A.; Goodman, A.M. Efficacy of metformin in patients with non-insulin-dependent diabetes mellitus. The Multicenter Metformin Study Group. *N. Engl. J. Med.* **1995**, *333*, 541–549. [CrossRef]
19. Jackson, R.A.; Hawa, M.I.; Jaspan, J.B.; Sim, B.M.; DiSilvio, L.; Featherbe, D.; Kurtz, A.B. Mechanism of metformin action in non-insulin-dependent diabetes. *Diabetes* **1987**, *36*, 632–640. [CrossRef] [PubMed]
20. Gin, H.; Messerchmitt, C.; Brottier, E.; Aubertin, J. Metformin improved insulin resistance in type I, insulin-dependent, diabetic patients. *Metabolism* **1985**, *34*, 923–925. [CrossRef]
21. Hother-Nielsen, O.; Schmitz, O.; Andersen, P.H.; Beck-Nielsen, H.; Pedersen, O. Metformin improves peripheral but not hepatic insulin action in obese patients with type II diabetes. *Acta Endocrinol.* **1989**, *120*, 257–265. [CrossRef] [PubMed]
22. Saisho, Y. Metformin and Inflammation: Its Potential Beyond Glucose-lowering Effect. *Endocr. Metab. Immune Disord. Drug Targets* **2015**, *15*, 196–205. [CrossRef] [PubMed]
23. Hatunic, M.; Finucane, F.; Burns, N.; Gasparro, D.; Nolan, J.J. Vascular inflammatory markers in early-onset obese and type 2 diabetes subjects before and after three months' aerobic exercise training. *Diab Vasc. Dis. Res.* **2007**, *4*, 231–234. [CrossRef] [PubMed]
24. Karbalaee-Hasani, A.; Khadive, T.; Eskandari, M.; Shahidi, S.; Mosavi, M.; Nejadebrahimi, Z.; Khalkhali, L.; Sangdari, A.; Mohammadi, D.; Soltani, A.; et al. Effect of Metformin on Circulating Levels of Inflammatory Markers in Patients with Type 2 Diabetes: A Systematic Review and Meta-analysis of Randomized Controlled Trials. *Ann. Pharmacother.* **2021**, *55*, 1096–1109. [CrossRef]
25. Syed, S.U.; Cortez, J.I.; Wilson, S.J. Depression, Inflammation, and the Moderating Role of Metformin: Results From the Midlife in the United States Study and Sacramento Area Latino Study on Aging. *Psychosom. Med.* **2024**, *86*, 473–483. [CrossRef]
26. Goldberg, R.B.; Temprosa, M.G.; Mather, K.J.; Orchard, T.J.; Kitabchi, A.E.; Watson, K.E.; for the Diabetes Prevention Program Research Group. Lifestyle and metformin interventions have a durable effect to lower CRP and tPA levels in the diabetes prevention program except in those who develop diabetes. *Diabetes Care* **2014**, *37*, 2253–2260. [CrossRef]
27. Cameron, A.R.; Morrison, V.L.; Levin, D.; Mohan, M.; Forteath, C.; Beall, C.; McNeilly, A.D.; Balfour, D.J.; Savinko, T.; Wong, A.K.; et al. Anti-Inflammatory Effects of Metformin Irrespective of Diabetes Status. *Circ. Res.* **2016**, *119*, 652–665. [CrossRef]
28. Jafarzadeh, S.; Nemati, M.; Zandvakili, R.; Jafarzadeh, A. Modulation of M1 and M2 macrophage polarization by metformin: Implications for inflammatory diseases and malignant tumors. *Int. Immunopharmacol.* **2025**, *151*, 114345. [CrossRef]
29. MacEachern, T.; John-Baptiste, A.; Christie, A. The prevalence of Black/African American individuals in concussion literature: A systematic review and meta-analysis. *Front. Public. Health* **2024**, *12*, 1430428. [CrossRef]
30. Ahmed, R.; de Souza, R.J.; Li, V.; Banfield, L.; Anand, S.S. Twenty years of participation of racialised groups in type 2 diabetes randomised clinical trials: A meta-epidemiological review. *Diabetologia* **2024**, *67*, 443–458. [CrossRef]
31. Foster, C.A.; Barker-Kamps, M.; Goering, M.; Patki, A.; Tiwari, H.K.; Mrug, S. Epigenetic age acceleration correlates with BMI in young adults. *Aging* **2023**, *15*, 513–523. [CrossRef]
32. Sunil, B.; Ashraf, A.P. Dyslipidemia in Pediatric Type 2 Diabetes Mellitus. *Curr. Diab Rep.* **2020**, *20*, 53. [CrossRef]
33. Kim, G.; DeSalvo, D.; Guffey, D.; Minard, C.G.; Cephus, C.; Moodie, D.; Lyons, S. Dyslipidemia in adolescents and young adults with type 1 and type 2 diabetes: A retrospective analysis. *Int. J. Pediatr. Endocrinol.* **2020**, *2020*, 11. [CrossRef]
34. Jung, M.K.; Yoo, E.G. Hypertriglyceridemia in Obese Children and Adolescents. *J. Obes. Metab. Syndr.* **2018**, *27*, 143–149. [CrossRef]
35. Reinehr, T. Inflammatory markers in children and adolescents with type 2 diabetes mellitus. *Clin. Chim. Acta* **2019**, *496*, 100–107. [CrossRef]
36. Marcelino Rodriguez, I.; García, J.O.; Sánchez, J.J.A.; González, D.A.; Coello, S.D.; Díaz, B.B.; Gannar, F.; Pérez, M.d.C.R.; Elosua, R.; de León, A.C. Lipid and inflammatory biomarker profiles in early insulin resistance. *Acta Diabetol.* **2016**, *53*, 905–913. [CrossRef] [PubMed]
37. Muscari, A.; Antonelli, S.; Bianchi, G.; Cavrini, G.; Dapporto, S.; Ligabue, A.; Ludovico, C.; Magalotti, D.; Poggiopollini, G.; Zoli, M.; et al. Serum C3 is a stronger inflammatory marker of insulin resistance than C-reactive protein, leukocyte count, and erythrocyte sedimentation rate: Comparison study in an elderly population. *Diabetes Care* **2007**, *30*, 2362–2368. [CrossRef]
38. Natali, A.; Toschi, E.; Baldeweg, S.; Ciociaro, D.; Favilla, S.; Saccà, L.; Ferrannini, E. Clustering of insulin resistance with vascular dysfunction and low-grade inflammation in type 2 diabetes. *Diabetes* **2006**, *55*, 1133–1140. [CrossRef] [PubMed]
39. Chen, J.; Wildman, R.P.; Hamm, L.L.; Muntner, P.; Reynolds, K.; Whelton, P.K.; He, J. Association between inflammation and insulin resistance in U.S. nondiabetic adults: Results from the Third National Health and Nutrition Examination Survey. *Diabetes Care* **2004**, *27*, 2960–2965. [CrossRef] [PubMed]
40. Akash, M.S.; Rehman, K.; Chen, S. Role of inflammatory mechanisms in pathogenesis of type 2 diabetes mellitus. *J. Cell Biochem.* **2013**, *114*, 525–531. [CrossRef]

41. Pradhan, A.D.; Imhof, A.; Berg, G.; Hutchinson, W.L.; Pepys, M.B.; Boeing, H.; Muche, R.; Brenner, H.; Koenig, W. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. *JAMA* **2001**, *286*, 327–334. [CrossRef]
42. Liu, C.; Feng, X.; Li, Q.; Wang, Y.; Li, Q.; Hua, M. Adiponectin, TNF-alpha and inflammatory cytokines and risk of type 2 diabetes: A systematic review and meta-analysis. *Cytokine* **2016**, *86*, 100–109. [CrossRef] [PubMed]
43. Cecerska-Heryc, E.; Engwert, W.; Michałów, J.; Marciniak, J.; Birger, R.; Serwin, N.; Heryć, R.; Polikowska, A.; Goszka, M.; Wojciuk, B.; et al. Oxidative stress markers and inflammation in type 1 and 2 diabetes are affected by BMI, treatment type, and complications. *Sci. Rep.* **2025**, *15*, 23605. [CrossRef]
44. Cao, C.; Yuan, J.; Gilbert, E.R.; Cline, M.A.; Lam, F.; Li, K.C.; Dilger, R.N. Increased Circulating Interleukin Concentrations in Type 2 Diabetes: A Systematic Review and Meta-Analysis. *Obes. Rev.* **2025**, *26*, e13971. [CrossRef]
45. Pellegrini, V.; La Grotta, R.; Carreras, F.; Giuliani, A.; Sabbatinelli, J.; Olivieri, F.; Berra, C.C.; Ceriello, A.; Prattichizzo, F. Inflammatory Trajectory of Type 2 Diabetes: Novel Opportunities for Early and Late Treatment. *Cells* **2024**, *13*, 1662. [CrossRef]
46. Kristofi, R.; Eriksson, J.W. Metformin as an anti-inflammatory agent: A short review. *J. Endocrinol.* **2021**, *251*, R11–R22. [CrossRef]
47. Sun, Y.; Li, J.; Xiao, N.; Wang, M.; Kou, J.; Qi, L.; Huang, F.; Liu, B.; Liu, K. Pharmacological activation of AMPK ameliorates perivascular adipose/endothelial dysfunction in a manner interdependent on AMPK and SIRT1. *Pharmacol. Res.* **2014**, *89*, 19–28. [CrossRef] [PubMed]
48. Han, Y.C.; Tang, S.-Q.; Liu, Y.-T.; Li, A.-M.; Zhan, M.; Yang, M.; Song, N.; Zhang, W.; Wu, X.-Q.; Peng, C.-H.; et al. AMPK agonist alleviate renal tubulointerstitial fibrosis via activating mitophagy in high fat and streptozotocin induced diabetic mice. *Cell Death Dis.* **2021**, *12*, 925. [CrossRef] [PubMed]
49. Bakkar, N.Z.; Mougharbil, N.; Mroueh, A.; Kaplan, A.; Eid, A.H.; Fares, S.; Zouein, F.A.; El-Yazbi, A.F. Worsening baroreflex sensitivity on progression to type 2 diabetes: Localized vs. systemic inflammation and role of antidiabetic therapy. *Am. J. Physiol. Endocrinol. Metab.* **2020**, *319*, E835–E851. [CrossRef]
50. Bharath, L.P.; Nikolajczyk, B.S. The intersection of metformin and inflammation. *Am. J. Physiol. Cell Physiol.* **2021**, *320*, C873–C879. [CrossRef]
51. Adeshirlarijaney, A.; Zou, J.; Tran, H.Q.; Chassaing, B.; Gewirtz, A.T. Amelioration of metabolic syndrome by metformin associates with reduced indices of low-grade inflammation independently of the gut microbiota. *Am. J. Physiol. Endocrinol. Metab.* **2019**, *317*, E1121–E1130. [CrossRef]
52. Dludla, P.V.; Nkambule, B.B.; Mazibuko-Mbeje, S.E.; Nyambuya, T.M.; Mxinwa, V.; Mokgalaboni, K.; Ziqubu, K.; Cirilli, I.; Marcheggiani, F.; Louw, J.; et al. Adipokines as a therapeutic target by metformin to improve metabolic function: A systematic review of randomized controlled trials. *Pharmacol. Res.* **2021**, *163*, 105219. [CrossRef]
53. Johansson, H.; Bellerba, F.; Macis, D.; Bertelsen, B.-E.; Guerrieri-Gonzaga, A.; Aristarco, V.; Viste, K.; Mellgren, G.; Di Cola, G.; Costantino, J.; et al. Effect of metformin and lifestyle intervention on adipokines and hormones in breast cancer survivors: A pooled analysis from two randomized controlled trials. *Breast Cancer Res. Treat.* **2024**, *205*, 49–59. [CrossRef]
54. Jing, Y.; Wu, F.; Li, D.; Yang, L.; Li, Q.; Li, R. Metformin improves obesity-associated inflammation by altering macrophages polarization. *Mol. Cell Endocrinol.* **2018**, *461*, 256–264. [CrossRef] [PubMed]
55. Pradhan, A.D.; Everett, B.M.; Cook, N.R.; Rifai, N.; Ridker, P.M. Effects of initiating insulin and metformin on glycemic control and inflammatory biomarkers among patients with type 2 diabetes: The LANCET randomized trial. *JAMA* **2009**, *302*, 1186–1194. [CrossRef]
56. Thomas, N.E.; Rowe, D.A.; Murtagh, E.M.; Stephens, J.W.; Williams, R. Associations between metabolic syndrome components and markers of inflammation in Welsh school children. *Eur. J. Pediatr.* **2018**, *177*, 409–417. [CrossRef]
57. Lampropoulou, I.T.; Stangou, M.; Papagianni, A.; Didangelos, T.; Iliadis, F.; Efstratiadis, G. TNF-alpha and microalbuminuria in patients with type 2 diabetes mellitus. *J. Diabetes Res.* **2014**, *2014*, 394206. [CrossRef] [PubMed]
58. Dolcini, J.; Landi, R.; Ponzio, E.; Picchini, L.; Luciani, A.; Formenti, L.; Baroni, S.S.; Gabrielli, A.; D'errico, M.M.; Barbadoro, P. Association between TNF-α, cortisol levels, and exposure to PM10 and PM2.5: A pilot study. *Environ. Sci. Eur.* **2024**, *36*, 141. [CrossRef]

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