






Pyroptosis-Mediator GSDMD in Plasma: A Promising Biomarker for Early Diagnosis of Type 2 Diabetes

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Objective: This study investigates the potential of plasma Gasdermin D (GSDMD) as a novel biomarker for early diagnosis and monitoring of type 2 diabetes mellitus.

Methods: We conducted a comparative analysis of clinical indicators among newly diagnosed diabetes patients, those with prediabetes, and healthy controls, finding significantly elevated plasma GSDMD levels in the diabetes group ($P < 0.05$).

Results: Correlation analyses revealed that GSDMD levels were positively associated with inflammatory markers and indicators of insulin resistance, while negatively correlating with HOMA- β ($P < 0.001$). Logistic regression analysis identified GSDMD, IL-6, and CRP as independent risk factors for type 2 diabetes ($P < 0.001$). The area under the ROC curve for plasma GSDMD was 0.988, indicating superior diagnostic capability compared to traditional markers like CRP (0.902) and IL-6 (0.857). With a threshold of 17.67 pg/mL, plasma GSDMD exhibited a sensitivity of 93.9% and specificity of 98.0%.

Conclusion: These findings suggest that plasma GSDMD not only reflects early metabolic abnormalities associated with type 2 diabetes but also holds promise as a therapeutic target to mitigate inflammatory responses and improve insulin resistance. Further large-scale clinical studies are warranted to validate its diagnostic utility and enhance clinical applications.

Keywords: biomarker discovery, gasdermin D, early diagnosis, type 2 diabetes mellitus, plasma biomarker, prediabetes

Introduction

According to the International Diabetes Federation (IDF), the number of individuals aged 18 and older with diabetes is projected to reach 693 million globally by 2045.¹ Diabetes is a chronic metabolic inflammatory disease characterized by insufficient insulin secretion or insulin resistance, resulting in an imbalance between anabolism and catabolism, and elevated blood glucose levels.² Type 2 diabetes mellitus (T2DM) has been described as a lack of insulin and insulin resistance in target cells associated with these islet cell dysfunctions.³ This condition is often linked to complications such as diabetic cardiomyopathy, diabetic nephropathy, and atherosclerosis.⁴ The progression of these microvascular diseases involves multiple mechanisms, such as elevated polyol and hexosamine pathway activity, excessive advanced glycosylation end products (AGEs) and protein kinase C (PKC) isoforms, and inadequate antioxidant defenses.⁵

Dyslipidemia is common in patients with type 2 diabetes mellitus (T2DM) and is mainly a mixed dyslipidemia [increased triglycerides (TGs), low high-density lipoprotein cholesterol (HDL-C), small dense (atherogenic), and low-density lipoprotein cholesterol (LDL-C) particles]. Numerous studies have highlighted chronic non-infectious inflammation as a critical factor in the onset and progression of diabetes,⁶ marked by elevated levels of inflammatory cytokines, such as interleukin-6 (IL-6) and interleukin-1 β (IL-1 β).⁷

Pyroptosis, a novel form of inflammatory cell death, was initially observed in macrophages infected by *Salmonella* and is mediated by the activation of caspase-1/4/5/11 in response to damage-associated molecular patterns (DAMPs) and pathogen-associated molecular patterns (PAMPs). This activation triggers the release of IL-1 β and IL-18, eliciting a robust inflammatory response.^{8,9} Gasdermin D (GSDMD), a member of the gasdermin family, serves as a substrate for caspase-1 and caspase-4/5/11, acting as the executor of pyroptosis. Upon cleavage by these caspases, GSDMD generates C-terminal and N-terminal fragments, with the N-terminus oligomerizing at the cell membrane to form non-selective pore channels. This process facilitates the release of mature IL-18 and IL-1 β , thereby inducing pyroptosis.^{10,11} Research indicates that GSDMD contributes to the progression of various diseases, including diabetes, by regulating immune cell death and the release of inflammatory factors such as IL-1 β .^{12,13} Chronic hyperglycemia and chronic inflammatory states cause insulin resistance in pancreatic islet cells. The release of inflammatory factors during cellular pyroptosis not only exacerbates the inflammatory response within pancreatic islets, but also directly affects the insulin signaling pathway, leading to insulin resistance. Under high-glucose and high-fat conditions, ROS production in pancreatic islet β -cells was increased, activating NLRP3 inflammatory vesicles, which in turn activated Caspase-1, contributing to the cleavage of GSDMD and inducing cellular pyroptosis. This process not only exacerbates islet β -cell loss, but also further damages islet structure and function through the release of inflammatory factors.

Currently, commonly employed diagnostic methods for type 2 diabetes in clinical practice include fasting blood glucose, fasting insulin, and glycated hemoglobin, among others.¹⁴ However, these indicators exhibit notable limitations; they are significantly influenced by factors such as diet and exercise, and often fail to detect the disease in its early stages. Therefore, the identification of novel, more sensitive, and specific diagnostic markers is critical for the early diagnosis and effective management of type 2 diabetes. The objective of this study is to investigate the differences in plasma GSDMD expression among individuals with varying glucose metabolism profiles, analyze the correlation between plasma GSDMD levels and disease severity, and assess its role as an independent risk factor for the onset of T2DM. Additionally, the study aims to evaluate the diagnostic value of plasma GSDMD for T2DM, providing a theoretical foundation for early screening to mitigate the risk of disease onset.

Materials and Methods

Study Population

Between January 2021 and December 2022, a total of 100 cases of newly diagnosed type 2 diabetes and 100 cases of prediabetes were selected from the Department of Endocrinology at Hebei General Hospital. Additionally, 101 healthy individuals from the Physical Examination Center of the same hospital, frequency-matched for age and gender with the diabetes and prediabetes patients, were included as the healthy control group. This control group had no history of diabetes, nephropathy, or other significant diseases and did not meet any diagnostic criteria for diabetes. This study received approval from the Ethics Committee of Hebei General Hospital [Approval Number: 2024(268)], and all participants provided informed consent.

Diagnostic Criteria

The diagnostic criteria for the prediabetes group were defined as fasting blood glucose levels between 6.1 mmol/L and 7.0 mmol/L. For the diabetes group, the criteria were established as a fasting blood glucose level of 7.0 mmol/L or higher.¹⁵

Inclusion Criteria

(1) Age between 18 and 80 years; (2) Newly diagnosed patients with type 2 diabetes mellitus (T2DM) and prediabetes who meet the diagnostic criteria established by the American Diabetes Association (ADA) in 2020. (3) Availability of complete clinical data; (4) Voluntary signing of an informed consent form.

Exclusion Criteria

(1) Participants with other types of diabetes; (2) Individuals with diabetes complicated by acute or chronic infections; (3) Individuals with diabetes complicated by acute cardiovascular or cerebrovascular diseases; (4) Individuals with diabetes

complicated by malignant tumors, severe liver dysfunction, or kidney dysfunction; (5) History of acute severe diabetes complications and use of steroid medications.

Collection of Clinical Data

Data were collected on patient gender, age, body mass index (BMI), fasting blood glucose (FBG), fasting insulin (Fins), glycated hemoglobin (HbA1c), total cholesterol (TC), triglycerides (TG), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and high-sensitivity C-reactive protein (hsCRP). The homeostasis model assessment of insulin secretion index (HOMA- β) was calculated using the formula: $\text{HOMA-}\beta = 20 \times \text{Fins} (\mu\text{IU/L}) / [\text{FBG} (\text{mmol/L}) - 3.5]$. The insulin resistance index (HOMA-IR) was determined using the formula: $\text{HOMA-IR} = \text{FBG} (\text{mmol/L}) \times \text{Fins} (\mu\text{IU/L}) / 22.5$.

Sample Collection

All subjects were required to fast for more than 8 hours. The following morning, 5 mL of non-anticoagulant venous blood was collected from fasting participants and allowed to coagulate at room temperature for 2 hours. After centrifugation at $1000\times g$ for 15 minutes at 4°C , plasma was collected and stored at -80°C until analysis. Levels of plasma interleukin-6 (IL-6) and interleukin-1 β (IL-1 β) were determined using enzyme-linked immunosorbent assay (ELISA) (Hangzhou Linktech Bioengineering Co., Ltd). The plasma level of Gasdermin D (GSDMD) was measured via chemiluminescence assay (Beijing Meide Taikang Biotechnology Co., Ltd). The kit item number is MDTK8001. All procedures were conducted in strict accordance with the instructions provided in the assay kits. The calibration and validation steps for these techniques are detailed in [supplementary material 1](#).

Statistical Analysis

Measurement data conforming to a normal distribution are presented as mean \pm standard deviation ($X \pm S$), while non-normally distributed data are expressed as median (Q1, Q3). Given the study's focus on multiple comparisons, a homogeneity of variances test was performed. For data with homogeneous variances, one-way ANOVA was utilized, followed by multiple comparisons using the Least Significant Difference (LSD) method. In cases of non-homogeneous variances, non-parametric tests were employed, including the Kruskal–Wallis test for multiple group comparisons and the Bonferroni correction for pairwise comparisons. Qualitative data were analyzed using chi-square tests. Spearman's rank correlation analysis assessed the relationship between plasma GSDMD levels and clinical indicators. Additionally, multiple logistic regression analysis was conducted to identify risk factors for the onset of type 2 diabetes mellitus (T2DM). The diagnostic value of plasma GSDMD for T2DM was evaluated using the area under the receiver operating characteristic curve (AUC). We performed a correlation analysis of the study indicators and did not find multicollinearity between the variables. See [supplementary material 2](#). All statistical tests were two-tailed, with a significance threshold set at $P < 0.05$. All analyses were performed using SPSS 25.0 software.

Results

Comparison of Clinical Indicators Among Newly Diagnosed Diabetes, Prediabetes, and Healthy Controls

No statistically significant differences were observed in age, gender, or body mass index (BMI) among the three groups ($P > 0.05$, [Table 1](#)). However, significant differences were noted in several clinical indicators across the groups ($P < 0.05$, [Tables 1 and 2](#)). The Plasma levels of triglycerides (TG) values were highest in the pre-diabetic group, followed by those in the diabetic group, while tg values were lowest in the healthy control group. The Plasma levels of total cholesterol (TC), low-density lipoprotein (LDL), FPG, HbA1c (glycated hemoglobin A1c), FINs (fasting insulin), HOMA-IR (homeostasis model assessment of insulin resistance), GSDMD (Gasdermin D), IL-6 (Interleukin-6), IL-1 β (Interleukin-1 β), CRP (C-reactive protein) were highest in the diabetes group, followed by the prediabetes group, and lowest in the healthy control group ($P < 0.05$, [Figures 1A–C, E–H and 2A–D](#)). Conversely, the levels of high-density lipoprotein (HDL) and homeostasis model assessment of β -cell function (HOMA- β) were lowest in the diabetes group, intermediate in the

Table 1 Comparison of Clinical Data Among Three Groups

Projects	T2DM Group	Pre-T2DM Group	Healthy Control Group	$\chi^2/F/H$	P value
Gender (male/female)	44/55	44/56	54/47	2.301	0.317
Age (year)	56.47±12.59	55.63±7.56	54.50±12.31	0.807	0.447
BMI (Kg/m ²)	23.71(20.93–26.49)	23.54 (20.87–26.21)	23.53 (20.67–26.39)	1.384	0.912
TC (mmol/l) (mmol/l)	5.35 (4.61~5.98) [#]	5.08 (4.24~5.77) ^Δ	4.51 (4.03~5.01)	48.288	<0.001
TG (mmol/l)	1.40 (0.89~1.96) [#]	1.68 (1.28~2.21) ^Δ	0.98 (0.77~1.18)	65.100	<0.001
LDL (mmol/l)	3.35 (2.67~3.87) [#]	2.89 (2.39~3.54)	2.73 (2.41~3.16)	12.114	<0.001
HDL (mmol/l)	1.00 (0.89~1.15) [#]	1.24 (1.08~1.38) ^Δ	1.33 (1.21, 1.50)	51.485	<0.001
FPG (mmol/l)	8.73 (7.71~9.95) [#]	6.46 (6.31~6.64) ^Δ	5.16 (4.88~5.42)	242.262	<0.001
HbA1c (%)	8.30 (7.00~10.00) [#]	6.00 (5.76~6.36) ^Δ	5.69 (5.56~5.83)	172.478	<0.001
FINS (mU/l)	16.93 (15.15~18.79) [#]	14.78 (10.62~18.35) ^Δ	9.94 (7.46~14.05)	70.416	<0.001
HOMA-IR	6.51 (5.20~8.47) [#]	4.19 (2.96~5.38) ^Δ	2.28 (1.77~3.15)	157.887	<0.001
HOMA-β	65.86 (51.30~79.53) [#]	93.28 (69.49~117.83) ^Δ	130.67 (81.39~178.28)	75.674	<0.001

Note: * shows the comparison between the diabetic group and the pre-diabetic group; # indicates the comparison between the diabetic group and the healthy control; Δ represents the comparison between the pre-diabetic group and the healthy control.

Table 2 Comparison of Serum GSDMD and Inflammatory Factors Between the Three Groups

Projects	T2DM Group	Pre-T2DM Group	Healthy Control Group	$\chi^2/F/H$	P value
GSDMD (pg/mL)	74.35 (37.62~107.57) [#]	11.47 (8.52~13.57) ^Δ	5.06 (2.99~7.29)	214.775	<0.001
IL-6 (pg/mL)	6.39 (3.63~14.60) [#]	3.49 (2.25~5.12) ^Δ	2.36 (1.74~3.75)	88.698	<0.001
IL-1β (pg/mL)	2.59 (1.62~6.50) [#]	1.96 (1.15~2.54) ^Δ	0.94 (0.29~1.30)	115.194	<0.001
CRP (mg/l)	9.77 (3.93~16.30) [#]	4.49 (4.18~4.83) ^Δ	2.13 (1.14~3.07)	144.831	<0.001

Note: * shows the comparison between the diabetic group and the pre-diabetic group; # indicates the comparison between the diabetic group and the healthy control; Δ represents the comparison between the pre-diabetic group and the healthy control.

prediabetes group, and highest in the healthy control group (Figures 1D and I). For confidence intervals for plasma GSDMD levels between groups, see [supplementary material 3](#).

Analysis of the Correlation Between Plasma GSDMD and Inflammatory Markers, Islet Secretion Function, and Insulin Resistance Indicators

Spearman analysis showed a correlation between GSDMD and FPG ($r=0.634$, $P<0.05$), between GSDMD and HbA1c ($r=0.583$, $P<0.05$), between GSDMD and IL-6 ($r=0.471$, $P<0.05$), between GSDMD and IL-1β ($r=0.353$, $P<0.05$), between GSDMD and CRP ($r=0.242$, $P<0.05$), between GSDMD and HOMA-IR ($r=0.398$, $P<0.05$) and between GSDMD and FINS ($r=0.149$, $P<0.05$); Conversely, a significant negative correlation between GSDMD and HOMA-β ($r=-0.370$, $P<0.05$) (Table 3, Figure 3).

Analysis of Influencing Factors for the Onset of Newly Diagnosed Type 2 Diabetes Mellitus

In investigating factors influencing the onset of newly diagnosed type 2 diabetes mellitus, GSDMD, IL-6, IL-1β, CRP, total cholesterol (TC), triglycerides (TG), low-density lipoprotein (LDL), high-density lipoprotein (HDL), fasting plasma glucose (FPG), HbA1c, insulin levels (FINS), HOMA-IR, and HOMA-β were evaluated as independent variables, with newly diagnosed type 2 diabetes as the dependent variable. Univariate logistic regression analysis revealed that all observed indicators served as independent influencing factors ($P<0.001$). Subsequent multivariate logistic regression analysis identified GSDMD, IL-6, and CRP as independent risk factors for newly diagnosed type 2 diabetes ($P<0.001$, Table 4).

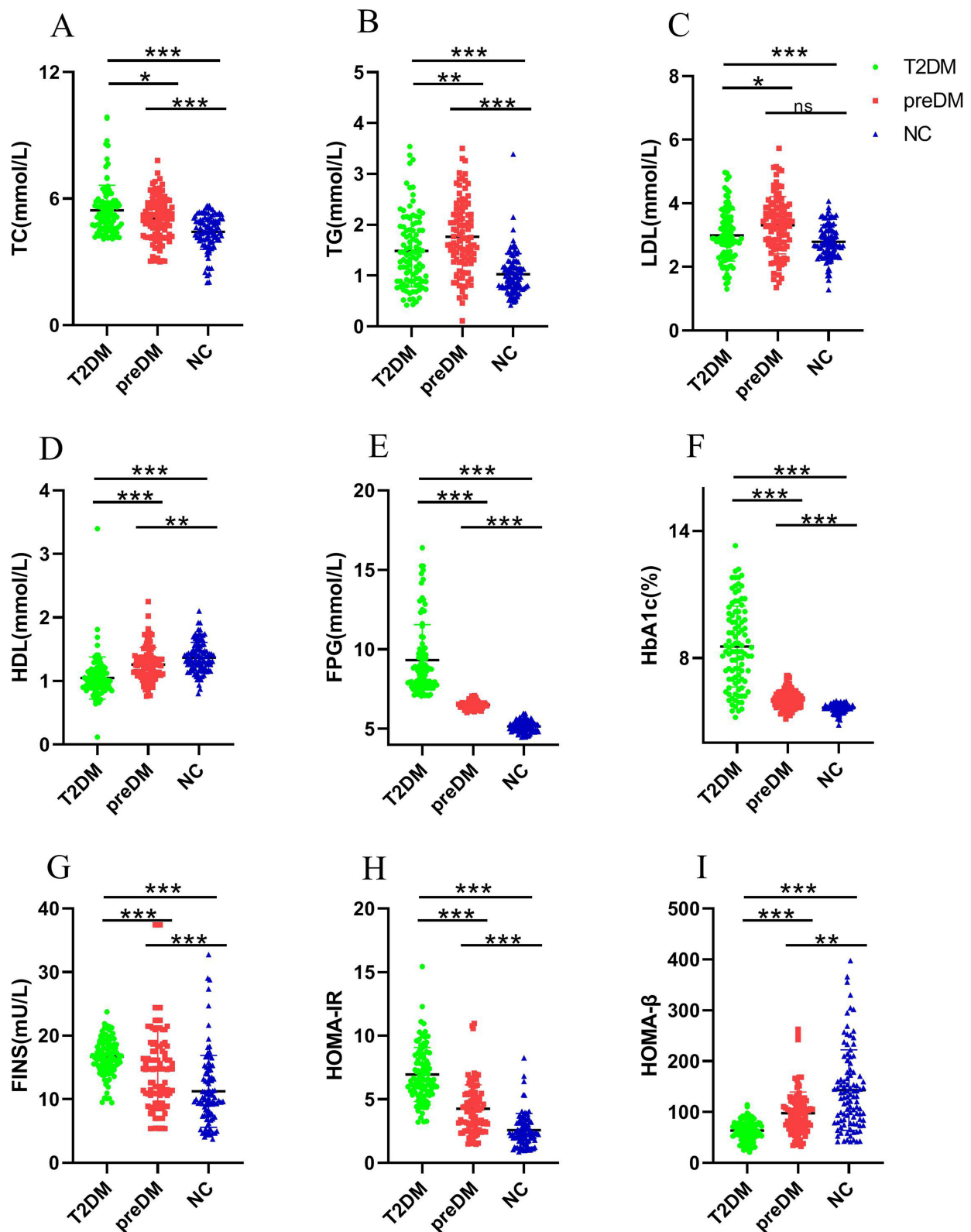


Figure 1 Plasma levels of TC(A), TG(B), LDL(C), HDL(D), FPG(E), HbA1c(F), FINS(G), HOMA-IR(H) and HOMA-β(I) in T2DM group, Pre-T2DM group and Healthy control group.

Note: * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

Abbreviations: TC, total cholesterol; TG, triglycerides; LDL, low-density lipoprotein; HDL, high density lipoprotein cholesterol; FPG, fasting blood glucose; HbA1c, glycated hemoglobin A1c; FINS, fasting insulin; HOMA-IR, homeostasis model assessment of insulin resistance; HOMA-β, homeostasis model assessment of β-cell function.

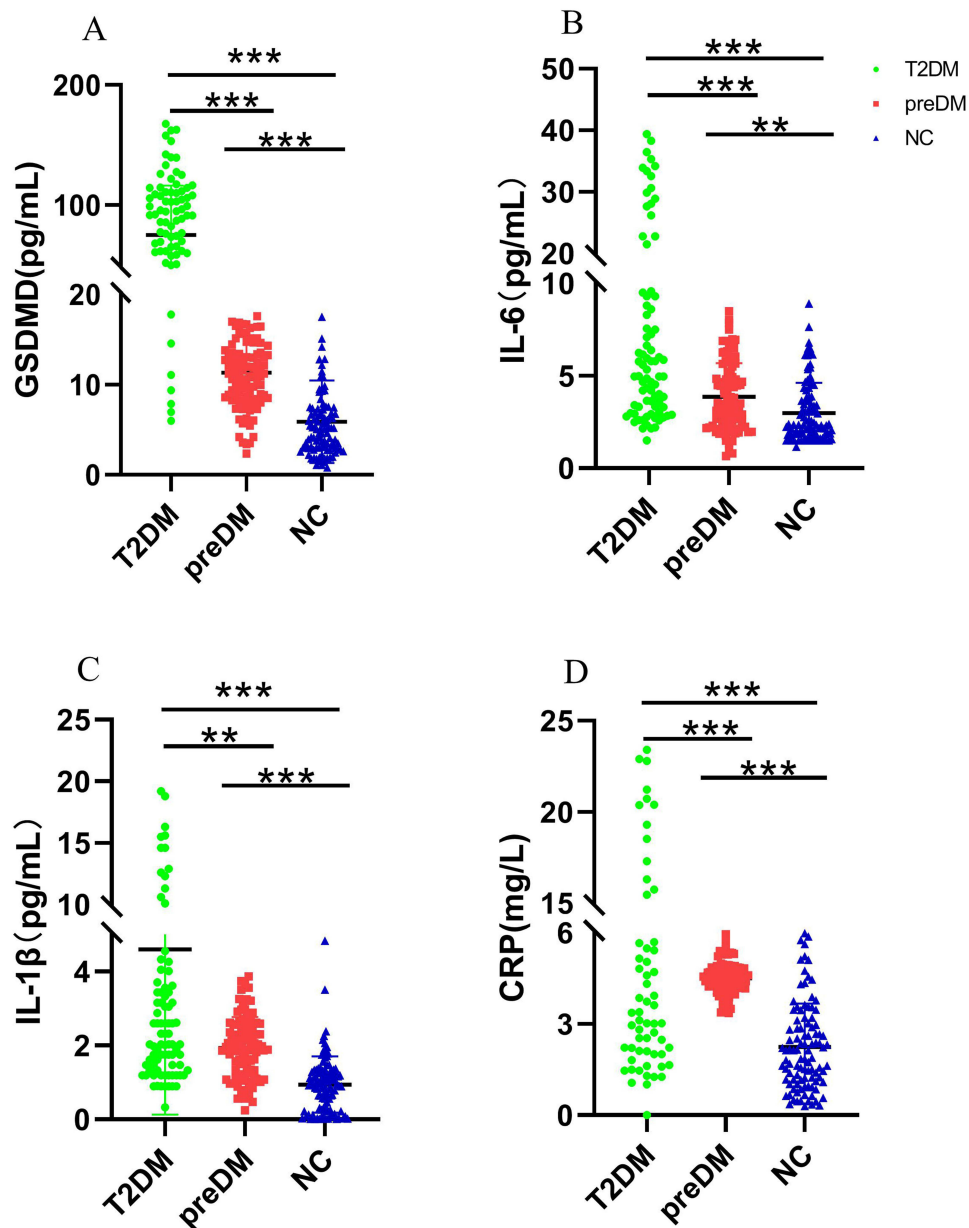


Figure 2 Plasma levels of GSDMD(A), IL-6(B), IL-1β(C), CRP(D) in T2DM group, Pre-T2DM group and Healthy control group.

Note: ** $p < 0.01$, *** $p < 0.001$.

Abbreviations: GSDMD, Gasdermin D; IL-6, Interleukin-6; IL-1β, Interleukin-1β; CRP, C-reactive protein.

The Predictive Value of Plasma GSDMD for Type 2 Diabetes

The area under the ROC curve (AUC) for plasma GSDMD (0.988) surpasses that of CRP (0.902) and IL-6 (0.857), indicating that plasma GSDMD possesses the highest diagnostic value among individual biomarkers for diabetes. At a plasma GSDMD threshold of 17.67 pg/mL, the sensitivity for diagnosis is 93.9%, with a specificity of 98.0% (Table 5, Figure 4).

Discussion

Type 2 diabetes is a metabolic disorder primarily characterized by insulin resistance and impaired insulin secretion. Insulin resistance, defined as the diminished sensitivity of tissues to insulin, results in decreased glucose uptake and utilization. New research identifies the potential of synthetic sulfonamide derivatives as potent multi-targeted inhibitors against key enzymes in metabolic diseases, offering valuable prospects for therapeutic intervention.¹⁶ This condition serves as

Table 3 Spearman Analysis Between GSDMD and Clinical Parameters (n=200)

Projects	GSDMD	
	<i>r</i>	<i>P</i> value
IL-6	0.471	<0.001
IL-1 β	0.353	<0.001
CRP	0.242	<0.001
FPG	0.634	<0.001
HbA1c	0.583	<0.001
FINS	0.149	<0.001
HOMA-IR	0.398	<0.001
HOMA- β	-0.370	<0.001

a common pathophysiological basis for metabolic diseases, including type 2 diabetes, obesity, and cardiovascular diseases. Recent studies have highlighted the significant role of pyroptosis in the development of insulin resistance. Pyroptosis is a form of programmed cell necrosis triggered by inflammasomes, which activate caspase-1 or caspase-11/4/5. This activation cleaves Gasdermin D (GSDMD), separating its N-terminal pore-forming domain (PFD) from its C-terminal inhibitory domain (RD). The PFD then oligomerizes within the cell membrane to form large pores, resulting in membrane swelling and rupture, which facilitates the release of pro-inflammatory cytokines such as interleukin-1 β (IL-1 β) that initiate downstream inflammatory responses. In this context, GSDMD acts as the executor of cell death.^{17,18}

GSDMD is a member of the gasdermin family, which comprises six members: GSDMA, GSDMB, GSDMC, GSDMD, GSDME, and PJVK.¹⁹ Cellular pyroptosis is a highly inflammatory form of programmed cell death involving an innate immune response capable of limiting the spread of pathogens by rapidly killing infected cells, but its overactivation may lead to chronic inflammation^{20,21} and exacerbate the progression of a variety of diseases. In the pathogenesis of type 2 diabetes, cellular pyroptosis has been shown to be closely associated with pathological processes such as pancreatic β -cell injury, and excessive inflammatory responses further exacerbate insulin resistance. The inflammatory response stimulates cleavage of the GSDMD protein molecule, releasing its N-terminal active fragment, which inserts into the cell membrane and forms a pore, disrupting the integrity of the cell membrane, leading to loss of intracellular water and ions, and ultimately triggering cell swelling and lysis. Caspase-1 also activates the pro-inflammatory cytokines IL-1 β and IL-18, which, along with the active fragment of the GSDMD protein molecule, are released into the cell membrane through the pores in the membrane are released extracellularly into the peripheral blood. The aim of this study is to observe the relationship between inflammation and insulin resistance in the pathogenesis of type 2 diabetes mellitus by detecting the active fragment (carboxy-terminal) of the GSDMD protein, a marker of cellular death, and inflammatory factors in peripheral blood, which can provide an important basis for the early diagnosis of type 2 diabetes mellitus.

Most gasdermin proteins are believed to possess pore-forming capabilities.¹⁹ Pro-inflammatory factors released by pyroptotic cells, including IL-1 β and interleukin-18 (IL-18), can activate inflammatory pathways, impairing insulin signaling and reducing insulin sensitivity, which in turn leads to tissue damage and insulin resistance.^{22,23} Notably, GSDMD can also induce pyroptosis in pancreatic β -cells, contributing to their dysfunction.²⁴ As the primary insulin-secreting cells, impaired pancreatic β -cells can result in inadequate insulin secretion, exacerbating the progression of type 2 diabetes.

Type 2 diabetes mellitus (T2DM) is characterized by a progressive decline in pancreatic beta cell function, which is manifested by inadequate insulin secretion and insulin resistance. Insulin resistance makes key tissues less responsive to insulin, resulting in chronically elevated blood glucose. Although β -cells initially compensate by increasing insulin secretion, this compensation gradually fails as the disease progresses. Type 2 diabetes stems not only from metabolic disorders²⁵ but is also closely associated with immune dysregulation²⁶ and chronic

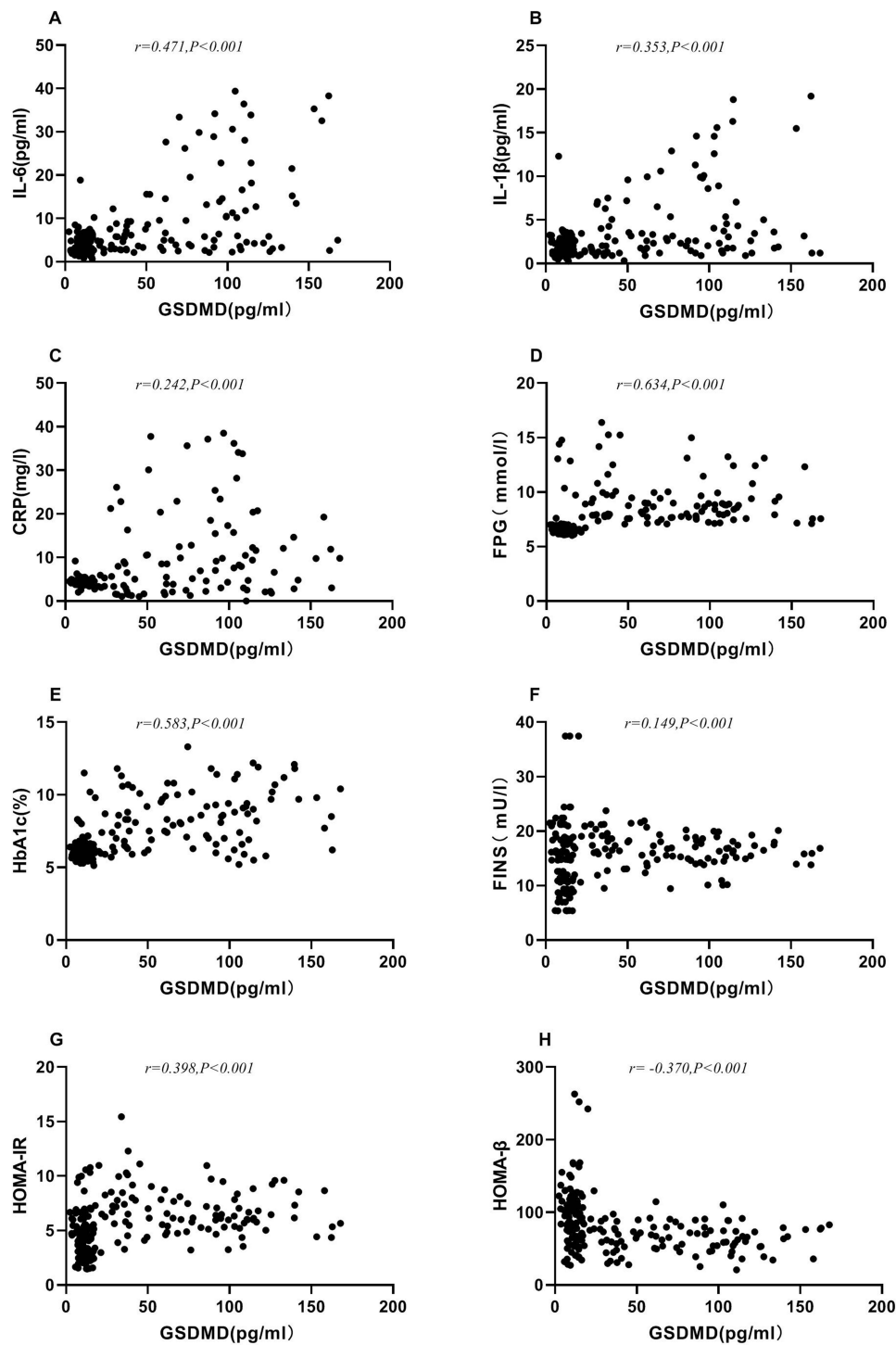


Figure 3 Analysis of the Correlation between plasma GSDMD and Inflammatory Markers, Islet Secretion Function, and Insulin Resistance Indicators. **(A)** scatterplot of plasma GSDMD levels associated with IL-6; **(B)** scatterplot of plasma GSDMD levels associated with IL-1 β ; **(C)** scatterplot of plasma GSDMD levels associated with CRP; **(D)** scatterplot of plasma GSDMD levels associated with FPG; **(E)** scatterplot of plasma GSDMD levels associated with HbA1c; **(F)** scatterplot of plasma GSDMD levels associated with FINS; **(G)** scatterplot of plasma GSDMD levels associated with HOMA-IR; **(H)** scatterplot of plasma GSDMD levels associated with HOMA- β .

inflammation.²⁷ Prolonged hyperglycemia leads to accumulation of AGEs²⁸ and activation of pro-inflammatory signaling pathways via RAGE receptors, releasing pro-inflammatory factors such as IL-6 and TNF- α ,²⁹ creating chronic low-grade inflammation and driving the development of diabetic insulin resistance. The search for new, more sensitive and specific diagnostic markers based on new targets for immune regulation and inflammation

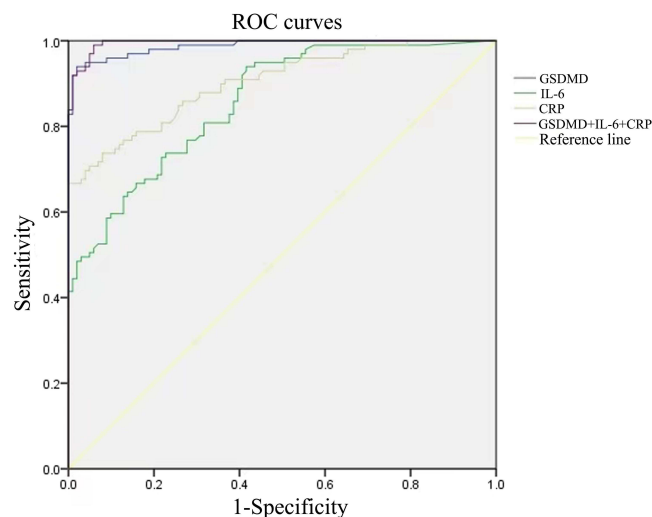
Table 4 Logistic Regression Analysis of Factors Developing Type 2 Diabetes

Independent Variable	Univariate		Multivariate	
	OR (95% CI)	P value	OR (95% CI)	P value
GSDMD	1.264 (1.162~1.375)	<0.001	1.200 (1.099~1.310)	<0.001
IL-6	1.671 (1.397~1.998)	<0.001	1.336 (1.037~1.721)	0.025
IL-1 β	5.294 (3.051~9.189)	<0.001	1.445 (0.965~2.165)	0.104
CRP	1.933 (1.560~2.396)	<0.001	1.764 (1.167~2.665)	0.007
TC	4.410 (2.668~7.289)	<0.001	2.888 (0.596~13.991)	0.188
TG	4.749 (2.525~8.933)	<0.001	0.785 (0.048~12.718)	0.865
LDL	2.868 (1.836~4.479)	<0.001	0.883 (0.163~4.799)	0.866
HDL	0.001 (0.000~0.007)	<0.001	0.000 (0.000~8.254)	0.120
FPG	2.282 (1.576~9.818)	<0.001	0.604 (0.180~1.925)	0.101
HbA1c	3.916 (2.853~5.049)	<0.001	4.381 (0.788~8.841)	0.159
FINS	1.320 (1.214~1.435)	<0.001	4.455 (0.731~27.155)	0.105
HOMA-IR	3.993 (2.739~5.819)	<0.001	1.469 (0.888~2.431)	0.134
HOMA- β	0.974 (0.966~0.983)	<0.001	0.864 (0.712~1.048)	0.138

Table 5 ROC Curve Analysis of the Predictive Value of Serum GSDMD for the Occurrence of First Diagnosed T2DM

Subjects	AUC	P	95% CI	Optimal Cut-Off Value	Sensitivity (%)	Specificity (%)
GD	0.988	0.000	0.976~0.999	17.67	93.9	98.0
IL-6	0.857	0.000	0.808~0.907	3.86	73.7	77.2
CRP	0.902	0.000	0.861~0.944	4.58	73.7	92.1
GD+IL-6+CRP	0.995	0.000	0.990~1.000	-	97.0	95.0

control is important for the early diagnosis and treatment of type 2 diabetes. The study found that plasma GSDMD levels were significantly elevated in patients with type 2 diabetes compared to those with prediabetes and healthy controls, with a statistically significant difference ($P < 0.05$). Furthermore, plasma GSDMD exhibited a significant positive correlation with IL-6, IL-1 β , CRP, fasting plasma glucose (FPG), insulin levels (FINS), HbA1c, and

**Figure 4** Predictive value of plasma GSDMD for type 2 diabetes mellitus ROC curve.

HOMA-IR ($P < 0.001$), while showing a significant negative correlation with HOMA- β ($P < 0.001$). Thus, plasma GSDMD is a promising diagnostic marker for type 2 diabetes. Additionally, significant differences in GSDMD levels were observed between type 2 diabetes patients and healthy individuals, as well as between those with type 2 diabetes and prediabetes, and between prediabetic and healthy controls ($P < 0.05$). These findings indicate that plasma GSDMD is closely related to the progression of insulin resistance and can facilitate stratified diagnosis of disease progression.

Multivariate logistic regression analysis indicated that GSDMD, IL-6, and CRP are independent risk factors for newly diagnosed type 2 diabetes ($P < 0.001$). Notably, plasma GSDMD levels begin to rise during the prediabetes stage, highlighting its potential value for early diagnosis. In contrast, traditional diagnostic indicators such as fasting blood glucose and glycated hemoglobin typically become abnormal only after the disease has progressed to a certain extent. Plasma GSDMD, however, can reflect metabolic abnormalities in the early stages of diabetes, providing a critical basis for early diagnosis and intervention.

In the diabetes prediction model established in this study, the area under the ROC curve for plasma GSDMD (0.988) surpassed that of CRP (0.902) and IL-6 (0.857), indicating that plasma GSDMD offers the highest diagnostic value among single detection indicators for diabetes. At a plasma GSDMD threshold of 17.67 pg/mL, the sensitivity for diagnosis is 93.9%, and the specificity is 98.0%. When plasma GSDMD levels were at 17.67 pg/mL, the risk of developing type 2 diabetes was higher. Furthermore, the combined assessment of plasma GSDMD with IL-6 and CRP enhances diagnostic sensitivity to 97% and specificity to 95%, providing a more comprehensive reflection of the severity and prognosis of the patient's condition.

Plasma GSDMD, as a novel biomarker, presents significant clinical application potential. It can be employed for the early diagnosis of type 2 diabetes, monitoring disease progression, and assessing prognosis. Additionally, GSDMD offers a new therapeutic target for type 2 diabetes by potentially mitigating inflammatory responses and improving insulin resistance through the inhibition of GSDMD activity.

In summary, plasma GSDMD may significantly contribute to the development of type 2 diabetes and could serve as a novel, more sensitive, and specific diagnostic marker, facilitating early diagnosis and treatment. However, research on GSDMD in the context of type 2 diabetes is still in its preliminary stages, and further large-scale clinical studies are necessary to validate its diagnostic value and clinical applicability. Additionally, there is a need for the development of more accurate, rapid, and convenient detection methods to enhance its use in clinical practice.

Conclusion

Elevated plasma GSDMD is closely associated with the occurrence of T2DM. The diagnostic value of plasma GSDMD for T2DM is superior to that of IL-6 and CRP. Plasma GSDMD, as a novel biomarker, holds broad clinical application prospects. It can be used for early diagnosis, disease monitoring, and prognosis assessment of type 2 diabetes. We will add future tests such as GSDMD in urine specimens, and are already in preliminary studies of GSDMD in T2DM nephropathy.

Data Sharing Statement

All data are available in the main text or the Supplementary Information. Detailed anonymized clinical data that support the findings of this study are available on reasonable request.

Ethics Statement

The study was conducted in accordance with the Declaration of Helsinki, received approval from the Ethics Committee of Hebei General Hospital [Approval Number:2024(268)], and all participants provided informed consent.

Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically

reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors report no conflicts of interest in this work.

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