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Association of Emerging Biomarkers with HbA1c and Glycaemic Control Among Patients with Type 2 Diabetes Mellitus: A Comparative Observational Study.

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Abstract
Background: Type 2 diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycaemia and progressive development of microvascular and macrovascular complications. Glycated haemoglobin (HbA1c) remains the standard biomarker for assessment of long-term glycaemic control; however, its interpretation may be influenced by factors affecting erythrocyte lifespan and haemoglobin metabolism. Emerging biomarkers such as fructosamine, glycated albumin, high-sensitivity C-reactive protein (hs-CRP), and adiponectin may provide additional information regarding short-term glycaemic variation, inflammation, and metabolic dysfunction. ***Objectives:*** To evaluate the association of HbA1c with selected emerging biomarkers and assess their clinical relevance in determining glycaemic control and metabolic abnormalities among patients with type 2 diabetes mellitus. ***Materials and Methods:*** This hospital-based observational comparative study was conducted in the Department of Biochemistry in collaboration with the Department of Medicine at Rajendra Institute of Medical Sciences (R.I.M.S), Ranchi, Jharkhand, from November 2024 to November 2025. A total of 120 patients with established type 2 diabetes mellitus were enrolled according to predefined inclusion and exclusion criteria. Clinical parameters and biochemical measurements including HbA1c, fructosamine, glycated albumin, hs-CRP, and adiponectin were evaluated using standardized laboratory methods. Correlation between HbA1c and other biomarkers was assessed using Pearson or Spearman correlation analysis as appropriate. Receiver operating characteristic (ROC) curve analysis was performed to evaluate the predictive performance of selected biomarkers for identifying poor glycaemic control. ***Results:*** HbA1c demonstrated a significant positive correlation with fructosamine and glycated albumin levels, indicating their association with glycaemic status. Patients with higher HbA1c values showed significantly increased fructosamine, glycated albumin, and hs-CRP levels compared with those having better glycaemic control ($p<0.05$). Adiponectin levels demonstrated an inverse association with HbA1c and markers of metabolic dysfunction. ROC analysis demonstrated that selected biomarkers showed potential predictive performance for identifying poor glycaemic control among patients with established type 2 diabetes mellitus. Combined evaluation of glycaemic and inflammatory biomarkers provided additional information regarding metabolic alterations among patients with type 2 diabetes mellitus. ***Conclusion:*** HbA1c remains an important marker for long-term glycaemic monitoring; however, complementary biomarkers including fructosamine, glycated albumin, hs-CRP, and adiponectin may improve assessment of short-term glycaemic variation and associated metabolic abnormalities. Combined biomarker evaluation may enhance clinical characterization and risk assessment among patients with type 2 diabetes mellitus.

Keywords: HbA1c; fructosamine; glycated albumin; hs-CRP; adiponectin; type 2 diabetes mellitus; glycaemic control; biomarkers.

INTRODUCTION
Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycaemia resulting from impaired insulin secretion, insulin resistance, or a combination of both. It is associated with progressive metabolic disturbances and an increased risk of microvascular and macrovascular complications. Accurate assessment of glycaemic control plays a crucial role in preventing disease progression and optimizing therapeutic strategies among affected individuals.[1]

Glycated haemoglobin (HbA1c) is an established biomarker for evaluating long-term glycaemic control as it reflects the average blood glucose concentration over approximately the preceding 2–3 months. Due to its clinical utility and standardization, HbA1c is widely used for monitoring patients with diabetes mellitus.[2] However, HbA1c values may be influenced by factors affecting erythrocyte turnover, haemoglobin variants, anaemia, and other clinical conditions, which may limit its interpretation in certain situations.[3]

Fructosamine and glycated albumin are alternative glycation biomarkers that reflect shorter periods of glycaemic exposure, generally representing glucose fluctuations over the previous 2–4 weeks. These markers may provide additional information regarding recent changes in glycaemic status and may be particularly useful when HbA1c does not completely reflect current metabolic conditions.[4]

Apart from hyperglycaemia, chronic low-grade inflammation and altered adipose tissue function contribute significantly to the pathogenesis and progression of type 2 diabetes mellitus. High-sensitivity C-reactive protein (hs-CRP), an inflammatory biomarker, has been associated with insulin resistance, metabolic dysfunction, and increased risk of diabetes-related complications.[5] Adiponectin, an adipocyte-derived hormone involved in glucose and lipid metabolism, enhances insulin sensitivity, and reduced circulating adiponectin levels have been linked with obesity, insulin resistance, and metabolic abnormalities.[6]

The combined assessment of HbA1c with emerging biomarkers may provide a broader understanding of glycaemic control, inflammatory status, and metabolic alterations among patients with diabetes mellitus. Correlation analysis between these parameters may help determine whether these biomarkers provide clinically relevant information beyond conventional glycaemic assessment.[7]

Receiver operating characteristic (ROC) curve analysis is a statistical approach used to evaluate the predictive performance of biomarkers in identifying specific clinical states. In patients with diabetes mellitus, ROC analysis may help determine the ability of emerging biomarkers to identify individuals with poor glycaemic control rather than serving as diagnostic tools for diabetes itself.[8]

Although HbA1c remains the cornerstone biomarker for long-term glycaemic monitoring, additional markers such as fructosamine, glycated albumin, hs-CRP, and adiponectin may provide complementary information regarding short-term glycaemic variation and associated metabolic abnormalities. Therefore, the present study was undertaken to evaluate the association of HbA1c with selected emerging biomarkers and assess their clinical relevance in relation to glycaemic control among patients with type 2 diabetes mellitus.

MATERIALS AND METHODS

Study Design

The present study was designed as a hospital-based observational comparative study to evaluate the association between glycated haemoglobin (HbA1c) and selected emerging biomarkers, including fructosamine, glycated albumin, high-sensitivity C-reactive protein (hs-CRP), and adiponectin, among patients with type 2 diabetes mellitus.

Study Setting and Duration

The study was conducted in the Department of Biochemistry in collaboration with the Department of Medicine, Rajendra Institute of Medical Sciences (R.I.M.S), Ranchi, Jharkhand. The study was carried out from 15 November 2024 to 14 November 2025.

Study Population

The study population included patients with previously diagnosed type 2 diabetes mellitus attending the outpatient and inpatient services of the Department of Medicine. Patients fulfilling the predefined inclusion and exclusion criteria were enrolled in the study.

Sample Size

The sample size was calculated based on the prevalence of poor glycaemic control among patients with type 2 diabetes mellitus reported in previous studies. Fekadu et al. reported that 64.9% of patients with type 2 diabetes mellitus had poor glycaemic control [20]. Considering a 95% confidence level and an absolute precision of 9%, the minimum required sample size was calculated using the formula:

$$n = Z^2PQ / d^2$$

where $Z=1.96$, $p=64.9\%$, $q=35.1\%$, and $d=9\%$. The calculated sample size was approximately 109. To improve the precision and reliability of the study findings, a total of 120 patients were included in the study.

Inclusion Criteria
Participants were included based on the following criteria:
- Patients diagnosed with type 2 diabetes mellitus according to standard clinical and biochemical criteria.
- Age ≥18 years.
- Patients willing to participate and provide written informed consent.
- Patients with available clinical history and relevant laboratory investigations.

Exclusion Criteria
Participants were excluded if they had:
- Acute infection or active inflammatory conditions at the time of sample collection.
- Chronic kidney disease, chronic liver disease, or other systemic disorders likely to affect biomarker levels.
- Haematological disorders or conditions affecting HbA1c interpretation.
- Recent blood transfusion.
- Incomplete clinical or biochemical data.
- Refusal to participate in the study.

Ethical Considerations
The study was conducted in accordance with the principles of the Declaration of Helsinki. Written informed consent was obtained from all participants before enrolment, and confidentiality of patient information was maintained throughout the study.

Clinical and Demographic Data Collection
Detailed clinical and demographic information was collected using a structured data collection proforma.
The following parameters were recorded:
- Age
- Sex
- Duration of diabetes mellitus
- Treatment history
- Anthropometric parameters including body mass index (BMI), wherever available
- Relevant clinical history and laboratory findings

Sample Collection
Approximately 5 ml of peripheral venous blood was collected from each participant under aseptic precautions.
The collected blood sample was appropriately processed for estimation of:
- HbA1c
- Fructosamine
- Glycated albumin
- High-sensitivity C-reactive protein (hs-CRP)
- Adiponectin
- Serum/plasma samples were separated and analysed according to standard laboratory procedures.

Laboratory Analysis
HbA1c estimation was performed using a standardized laboratory method. Serum fructosamine, glycated albumin, high-sensitivity C-reactive protein (hs-CRP), and adiponectin were measured using commercially available validated assay kits according to the manufacturer's instructions.

The assay principle, kit manufacturer details, analyzer model, calibration procedures, and intra- and inter-assay coefficients of variation were documented according to laboratory quality control protocols.

All biochemical analyses were performed under standardized conditions with appropriate internal quality control measures to ensure analytical accuracy and reproducibility.

Categorization According to Glycaemic Control
Participants were categorized according to HbA1c values into:
- Good glycaemic control group: HbA1c <7%
- Moderate glycaemic control group: HbA1c 7–9%
- Poor glycaemic control group: HbA1c >9%
- These categories were used for comparative analysis of biomarker levels.

Outcome Measures

Primary Outcome

To evaluate the correlation between HbA1c and selected emerging biomarkers among patients with type 2 diabetes mellitus.

Secondary Outcomes

- To compare fructosamine, glycated albumin, hs-CRP, and adiponectin levels according to glycaemic control categories.
- To assess the association between inflammatory/metabolic biomarkers and HbA1c levels.
- To evaluate the predictive performance of selected biomarkers for identifying poor glycaemic control using ROC curve analysis.

Statistical Analysis

Data were entered into Microsoft Excel and analysed using appropriate statistical software.

Continuous variables were expressed as mean ± standard deviation or median with interquartile range depending on data distribution. Categorical variables were presented as frequency and percentage.

Normality of distribution was assessed using appropriate statistical tests.

Correlation between HbA1c and other biomarkers was evaluated using Pearson or Spearman correlation analysis depending on data distribution.

Comparison of biomarker levels between groups was performed using Student's t-test, Mann–Whitney U test, or analysis of variance (ANOVA) as appropriate.

ROC curve analysis was performed to determine the predictive performance of selected biomarkers for identifying poor glycaemic control. Area under the curve (AUC), sensitivity, specificity, and optimal cut-off values were calculated wherever applicable.

Multiple linear regression analysis was performed to evaluate independent associations between biomarkers and HbA1c after adjustment for potential confounding factors including age, sex, BMI, duration of diabetes, and treatment status.

A p-value <0.05 was considered statistically significant.

RESULTS

A total of 120 patients with established type 2 diabetes mellitus were included in the present observational study. The demographic characteristics, glycaemic parameters, emerging biomarkers, and correlation analyses were evaluated. Participants were categorized according to HbA1c levels into good glycaemic control (<7%), moderate glycaemic control (7–9%), and poor glycaemic control (>9%) groups.

Demographic and Clinical Characteristics of Study Participants

The mean age of the study population was 56.8 ± 10.4 years. Among the participants, 68 (56.7%) were males and 52 (43.3%) were females. The mean duration of diabetes was 8.2 ± 4.6 years. Based on HbA1c categories, 32 (26.7%) participants had good glycaemic control, 48 (40.0%) had moderate glycaemic control, and 40 (33.3%) had poor glycaemic control. The demographic and clinical characteristics of study participants are presented in **Table 1**.

Table 1. Demographic and Clinical Characteristics of Study Participants

Parameter	Value
Total participants	120
Mean age (years)	56.8 ± 10.4
Male	68 (56.7%)
Female	52 (43.3%)
Mean duration of diabetes (years)	8.2 ± 4.6
HbA1c <7%	32 (26.7%)
HbA1c 7–9%	48 (40.0%)
HbA1c >9%	40 (33.3%)

Comparison of Biomarkers According to Glycaemic Control

The levels of HbA1c and emerging biomarkers showed significant variation across different glycaemic control categories. Participants with poor glycaemic control demonstrated significantly higher levels of fructosamine, glycated albumin, and hs-CRP compared with participants having good glycaemic control (p<0.001). Adiponectin levels showed a decreasing trend with worsening glycaemic control, indicating an inverse association with metabolic dysfunction. The comparison of biomarker levels among HbA1c groups is presented in **Table 2**.

Table 2. Biomarker Levels According to HbA1c Categories

Biomarker	HbA1c <7% (n=32)	HbA1c 7–9% (n=48)	HbA1c >9% (n=40)	p-value
HbA1c (%)	6.4 ± 0.4	8.1 ± 0.6	10.4 ± 1.1	<0.001
Fructosamine (µmol/L)	258.4 ± 34.6	318.7 ± 42.5	372.6 ± 48.8	<0.001
Glycated albumin (%)	14.2 ± 2.1	19.3 ± 3.0	24.8 ± 4.1	<0.001
hs-CRP (mg/L)	2.1 ± 0.9	3.8 ± 1.5	6.2 ± 2.4	<0.001
Adiponectin (µg/ml)	12.6 ± 3.4	9.8 ± 2.7	7.1 ± 2.3	<0.001

Figure 1 illustrates the comparison of mean biomarker levels across different HbA1c-based glycaemic control categories.

Figure 1. Comparison of Mean Biomarker Levels According to HbA1c Categories Among Patients with Type 2 Diabetes Mellitus

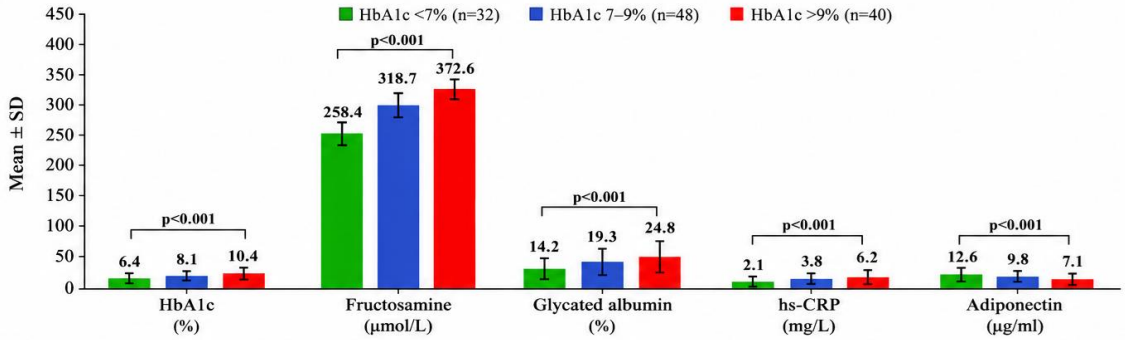

Figure 1: Comparison of Mean Biomarker Levels According to HbA1c Categories Among Patients with Type 2 Diabetes Mellitus.

Correlation Between HbA1c and Emerging Biomarkers

Correlation analysis demonstrated a significant positive correlation between HbA1c and fructosamine ($r=0.78$, $p<0.001$) and between HbA1c and glycated albumin ($r=0.74$, $p<0.001$).

HbA1c also showed a significant positive correlation with hs-CRP ($r=0.46$, $p<0.001$), suggesting an association between poor glycaemic control and inflammatory status.

A significant negative correlation was observed between HbA1c and adiponectin levels ($r=-0.41$, $p<0.001$).
The correlation analysis is presented in **Table 3**.

Table 3. Correlation of HbA1c with Emerging Biomarkers

Biomarker	Correlation coefficient (r)	95% CI	p-value
HbA1c vsFructosamine	0.78	0.69–0.85	<0.001
HbA1c vsGlycated albumin	0.74	0.63–0.82	<0.001
HbA1c vshs-CRP	0.46	0.31–0.59	<0.001
HbA1c vsAdiponectin	−0.41	−0.55 to −0.25	<0.001

Predictive Performance of Biomarkers for Poor Glycaemic Control

ROC curve analysis was performed to evaluate the predictive performance of selected biomarkers for identifying patients with poor glycaemic control (HbA1c >9%) among individuals with established type 2 diabetes mellitus. Fructosamine and glycated albumin demonstrated good predictive ability, whereas hs-CRP showed moderate predictive performance. The ROC analysis parameters are presented in **Table 4** and **Figure 2**.

Table 4. ROC Analysis of Biomarkers for Identifying Poor Glycaemic Control

Biomarker	Cut-off value	AUC (95% CI)	Sensitivity	Specificity	p-value
Fructosamine	>340 µmol/L	0.86 (0.78–0.92)	82.5%	78.8%	<0.001
Glycated albumin	>21%	0.84 (0.76–0.91)	80.0%	76.3%	<0.001
hs-CRP	>4 mg/L	0.71 (0.61–0.80)	70.0%	65.0%	0.002
Adiponectin	<8 µg/ml	0.69 (0.59–0.79)	67.5%	63.8%	0.004

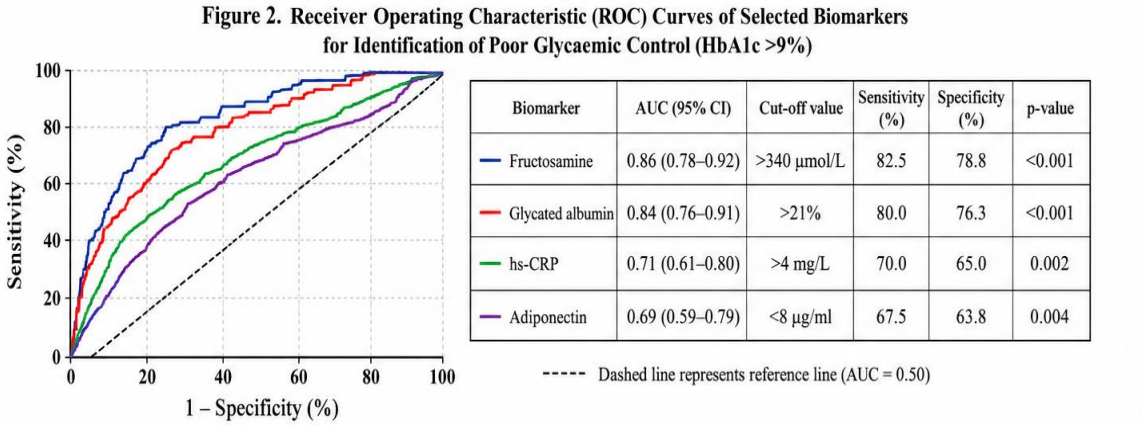

Figure 2. Receiver Operating Characteristic (ROC) Curves of Selected Biomarkers for Identification of Poor Glycaemic Control

Multiple Linear Regression Analysis

Multiple Linear Regression Analysis was performed to identify independent factors associated with HbA1c levels after adjustment for age, sex, BMI, duration of diabetes, and biomarker levels. Fructosamine and glycated albumin remained significantly associated with HbA1c after adjustment.

The regression analysis is presented in **Table 5**.

Table 5. Multiple Linear Regression Analysis for Factors Associated with HbA1c

Variable	Adjusted β coefficient	95% CI	p-value
Age	0.08	−0.02–0.18	0.12
Sex	0.05	−0.09–0.19	0.48
BMI	0.11	0.02–0.21	0.02
Diabetes duration	0.16	0.05–0.27	0.004
Fructosamine	0.52	0.39–0.65	<0.001
Glycated albumin	0.38	0.24–0.51	<0.001
hs-CRP	0.18	0.06–0.31	0.005
Adiponectin	−0.14	−0.25 to −0.03	0.01

Overall, fructosamine and glycated albumin showed the strongest association with HbA1c, supporting their role as complementary biomarkers for assessment of glycaemic control among patients with type 2 diabetes mellitus.

DISCUSSION

Type 2 diabetes mellitus is a complex metabolic disorder characterized by chronic hyperglycaemia and progressive alterations in glucose metabolism, inflammatory pathways, and metabolic regulation. Accurate assessment of glycaemic control is essential for prevention of diabetes-related complications. Although glycated haemoglobin (HbA1c) remains the established marker for long-term glycaemic monitoring, additional biomarkers may provide valuable information regarding short-term glycaemic fluctuations and associated metabolic abnormalities.[9]

The present study demonstrated significant associations between HbA1c and selected emerging biomarkers, including fructosamine, glycated albumin, high-sensitivity C-reactive protein (hs-CRP), and adiponectin, among patients with type 2 diabetes mellitus.

In the present study, fructosamine and glycated albumin demonstrated a strong positive correlation with HbA1c, indicating their potential usefulness as complementary markers of glycaemic status. Fructosamine reflects glycation of serum proteins and represents shorter-term glycaemic exposure over approximately 2–4 weeks. Similarly, glycated albumin reflects recent glucose fluctuations and may provide additional information when HbA1c is affected by conditions altering red blood cell turnover.[10]

The significant association observed between HbA1c and glycated albumin supports previous evidence that glycated albumin may serve as a reliable indicator of glycaemic variability and may correlate with diabetes-related complications. Unlike HbA1c, glycated albumin is not influenced by erythrocyte lifespan, making it potentially useful in selected clinical situations.[11]

The present study also demonstrated a significant positive association between HbA1c and hs-CRP levels. This finding suggests that poor glycaemic control may be associated with increased low-grade inflammation. Chronic inflammation plays an important role in insulin resistance, endothelial dysfunction, and development of diabetes-related complications. Elevated inflammatory markers, including hs-CRP, have been linked with increased metabolic risk among individuals with type 2 diabetes mellitus.[12,13]

Adiponectin showed an inverse relationship with HbA1c in the present study. Lower adiponectin concentrations were associated with poorer glycaemic control, supporting its role as an important regulator of insulin sensitivity and glucose metabolism. Reduced adiponectin levels have been reported in insulin resistance, obesity, and metabolic syndrome, contributing to impaired glucose homeostasis.[14,15]

ROC curve analysis in the present study was performed to evaluate the predictive performance of biomarkers for identifying poor glycaemic control rather than for diagnosis of diabetes mellitus. Fructosamine and glycated albumin demonstrated better predictive performance compared with inflammatory and adipokine markers, suggesting their potential utility as supportive biomarkers for assessment of glycaemic status.[16]

The multivariable analysis further demonstrated that fructosamine and glycated albumin remained independently associated with HbA1c after adjustment for potential confounding factors. These findings suggest that these biomarkers may provide additional clinical information beyond conventional HbA1c measurement.[17]

The combined evaluation of glycaemic, inflammatory, and metabolic biomarkers may improve understanding of disease heterogeneity among patients with type 2 diabetes mellitus. While HbA1c remains essential for routine monitoring, additional biomarkers may help identify patients with altered metabolic profiles and those requiring closer clinical assessment.[18,19]

The present study has certain limitations. It was conducted at a single tertiary care centre with a relatively limited sample size. As an observational study, causal relationships between biomarkers and glycaemic control cannot be definitively established.The study may also be susceptible to selection bias and residual confounding. Additionally, follow-up data regarding development of diabetic complications were not available. Larger multicentric longitudinal studies are required to validate the clinical utility of these biomarkers.

Overall, the findings indicate that fructosamine, glycated albumin, hs-CRP, and adiponectin may serve as complementary biomarkers for evaluating glycaemic control and associated metabolic alterations among patients with type 2 diabetes mellitus.

CONCLUSION

HbA1c remains an important biomarker for long-term assessment of glycaemic control among patients with type 2 diabetes mellitus. In the present study, fructosamine and glycated albumin showed significant association with HbA1c, suggesting their usefulness as complementary indicators of recent glycaemic variations.

The association of hs-CRP and adiponectin with HbA1c highlights the contribution of inflammatory and metabolic alterations in diabetes progression. Combined assessment of glycaemic, inflammatory, and metabolic biomarkers may provide additional information regarding metabolic status and help in better clinical characterization of patients with type 2 diabetes mellitus.

Further large-scale longitudinal studies are required to establish the long-term clinical utility of these biomarkers in routine diabetes management.

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