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Association between red cell distribution width and glycemic control in adults with type 2 diabetes mellitus: a cross-sectional study in northern Iran

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Abstract

Red cell distribution width (RDW) is a routine and inexpensive hematological parameter that has recently been explored as a potential biomarker of metabolic and inflammatory disorders. This study investigated the associations between RDW, platelet distribution width (PDW), neutrophil-to-lymphocyte ratio (NLR), and glycemic control in adults with type 2 diabetes mellitus (T2DM). This cross-sectional study included 160 adults with T2DM attending diabetes clinics affiliated with Guilan University of Medical Sciences, Iran. Demographic characteristics, clinical variables, and hematological indices were collected from medical records. Glycemic control was assessed using fasting blood sugar (FBS) and glycated hemoglobin (HbA1c). Associations were evaluated using Spearman's or Pearson's correlation coefficients, and comparisons between controlled (HbA1c < 7%) and uncontrolled (HbA1c ≥ 7%) groups were performed using appropriate parametric or non-parametric tests. RDW demonstrated a weak but statistically significant positive unadjusted correlation with HbA1c ($r = 0.280$, $P < 0.001$). However, the difference in RDW between patients with controlled and uncontrolled glycemic status was not statistically significant ($P = 0.188$), and multivariable adjustment was not performed. Therefore, RDW should be considered an exploratory rather than a confirmatory marker. RDW showed a weak positive association with HbA1c, but its modest correlation and non-significant between-group difference indicate that it should be considered an adjunctive, rather than a substitute, marker for glycemic control. PDW and NLR were not consistently associated with glycemic control. Further prospective studies with larger populations and multivariable adjustments are warranted.

Keywords: Type 2 diabetes mellitus, Red cell distribution width, RDW, Glycated hemoglobin, HbA1c, Complete blood count

1. Introduction

In Iran, the prevalence of type 2 diabetes is estimated at 10.8% and is expected to more than double over the next three decades, representing a substantial public health burden, particularly among middle-aged and older adults (1, 2). Given the growing prevalence and the long-term complications associated with poor glycemic control, identifying practical and accessible tools for disease monitoring remains a clinical priority.

Glycated hemoglobin (HbA1c) is the established indicator of long-term glycemic control and plays a central role in the management of type 2 diabetes mellitus (T2DM). Nevertheless, its routine assessment may be limited in some healthcare settings because of cost, availability, or laboratory infrastructure. Consequently, increasing attention has been directed toward inexpensive hematological parameters that are routinely measured as part of the complete blood count (CBC) and may provide complementary information regarding metabolic status (3, 4).

Red cell distribution width (RDW), a quantitative measure of the variability in erythrocyte size, has recently emerged as a potential biomarker in several chronic metabolic and inflammatory disorders. Persistent hyperglycemia may alter erythrocyte membrane structure, increase oxidative stress, and shorten red blood cell survival, resulting in greater anisocytosis and elevated RDW values. Several studies have reported positive associations between RDW and HbA1c or other indicators of glycemic status, although the magnitude and clinical significance of these associations remain inconsistent across different populations (5, 6).

Other CBC-derived indices have also attracted attention. Platelet distribution width (PDW) and mean platelet volume (MPV) reflect platelet activation and have been associated with vascular dysfunction and diabetic complications (7, 8). Likewise, the neutrophil-to-lymphocyte ratio (NLR), a readily available marker of systemic inflammation, has been investigated as a potential indicator of metabolic dysregulation and diabetes-related complications (9, 10). However, findings regarding the relationship between these hematological parameters and glycemic control have been inconsistent, and comparative evidence evaluating these biomarkers simultaneously remains limited.

Despite increasing interest in CBC-derived biomarkers, published findings remain inconsistent regarding the relationship of RDW, PDW, and NLR with glycemic control. Most available studies have evaluated only one hematological parameter or were conducted in limited populations. Furthermore, evidence from Middle Eastern populations remains scarce. Therefore, additional population-specific studies are needed to clarify the potential clinical utility of these routinely available biomarkers.

2. Methods

2.1. Study design and participants

This analytical cross-sectional study was conducted among adults with T2DM attending an outpatient diabetes clinic in northern Iran during 2024.

2.2. Eligibility criteria

Patients aged ≥18 years with a confirmed diagnosis of T2DM and available laboratory measurements, including fasting blood sugar (FBS), HbA1c, and CBC, were eligible for inclusion.

Patients were excluded if they had acute or chronic kidney disease, anemia, thrombocytopenia, leukopenia, polycythemia, thrombocytosis, or leukocytosis. Individuals who had received erythropoietin, insulin initiation, hematinic agents, or blood transfusion within the previous three months were also excluded. In addition, pregnant women, smokers, alcohol consumers, and patients with hypertension were not eligible for participation. Patients younger than 18 years or older than 65 years were also excluded.

2.3. Sample size and sampling

Sample size was calculated using G*Power software (version 3.1.9.7) for comparison of two independent means (RDW between controlled and uncontrolled glycemic groups). Assuming a moderate effect size (Cohen's $d = 0.45$) based on previous studies, $\alpha = 0.05$ (two-tailed), and power = 80%, the minimum required sample size was 156 participants. Accounting for a potential 5% attrition or incomplete data, we enrolled 160 participants.

2.4. Data collection

Demographic and clinical data were extracted from patients' medical records using a standardized data collection form. The collected variables included age, sex, FBS, HbA1c, hemoglobin (Hb), RDW, PDW, MPV, platelet count, neutrophil count, lymphocyte count, and the neutrophil-to-lymphocyte ratio (NLR). The NLR was calculated by dividing the absolute neutrophil

count by the absolute lymphocyte count.

2.5. Definition of glycemic control

Participants were categorized according to their HbA1c levels into two groups: controlled glycemic status (HbA1c < 7.0%) and uncontrolled glycemic status (HbA1c ≥ 7.0%). FBS was used as a complementary measure in descriptive analyses but was not part of the primary grouping criteria.

2.6. Statistical analysis

Statistical analyses were performed using IBM SPSS Statistics version 27 (IBM Corp., Armonk, NY, USA).

Continuous variables were summarized as mean ± standard deviation (SD), whereas categorical variables were presented as frequencies and percentages. The normality of continuous variables was assessed before analysis. Comparisons between study groups were performed using the independent-samples t-test or the Mann–Whitney U test, as appropriate. Categorical variables were compared using the chi-square test or Fisher's exact test.

Correlations between HbA1c and hematological parameters were evaluated using Pearson's or Spearman's correlation coefficients, depending on data distribution. All statistical tests were two-sided, and a P value <0.05 was considered statistically significant.

The normality of continuous variables was assessed using the Kolmogorov–Smirnov test. For variables with normal distribution, independent t-test and Pearson correlation were used. For non-normally distributed variables, Mann–Whitney U test and Spearman's rho correlation were applied.

3. Results

3.1. Baseline characteristics

A total of 160 adults with type 2 diabetes mellitus were included in the final analysis. The mean age of the participants was 50.64 ± 13.58 years (range, 18–65 years), and 91 (56.9%) were women. Based on HbA1c levels, 113 participants (70.6%) had controlled glycemic status (HbA1c <7.0%), whereas 47 (29.4%) had uncontrolled glycemic status (HbA1c ≥7.0%). The baseline demographic and laboratory characteristics of the study population are summarized in **(Table 1)**.

3.2. Relationship between HbA1c and hematological parameters

Correlation analysis showed a weak but statistically significant positive correlation between HbA1c and RDW (r = 0.280, P <0.001). RDW was also positively correlated with age (r = 0.227, P = 0.004). In addition, FBS showed a weak positive correlation with NLR (r = 0.197, P = 0.012), while NLR was positively correlated with white blood cell (WBC) count (r = 0.288, P <0.001). Platelet count was inversely correlated with MPV (r = −0.462, P <0.001), whereas red blood cell (RBC) count was positively correlated with hemoglobin concentration (r = 0.558, P <0.001) **(Table 3)**.

Comparison of hematological parameters according to glycemic status demonstrated that the mean RDW was higher in patients with uncontrolled diabetes than in those with controlled diabetes (13.74 ± 1.77 vs. 13.37 ± 1.87); however, this difference did not reach statistical significance (P = 0.188). Likewise, no statistically significant differences were observed for PDW, MPV, NLR, or hemoglobin concentration between the two groups (all P >0.05) **(Table 2)**.

3.3. Subgroup analyses

Sex-stratified analysis showed that male participants with uncontrolled diabetes had significantly higher NLR values than those with controlled diabetes (2.18 ± 0.48 vs. 1.82 ± 0.63, P = 0.023), whereas no significant difference was observed among female participants (P = 0.431). Age-stratified analysis demonstrated that participants aged 18–40 years with uncontrolled diabetes had significantly higher RDW values than those with controlled diabetes (16.05 ± 3.74 vs. 12.91 ± 1.25, P = 0.038). No statistically significant difference was observed among participants aged 41–65 years (P = 0.664) **(Table 4)**.

4. Discussion

The principal finding of this study was a weak but statistically significant positive correlation between RDW and HbA1c. However, the lack of a significant difference in RDW between the controlled and uncontrolled glycemic groups suggests that this correlation should be interpreted cautiously and does not support RDW as a reliable independent marker of glycemic control. Although the observed association was modest, it suggests that RDW may provide complementary information regarding glycemic status when interpreted alongside established measures of glycemic control. In contrast, no significant associations were observed between HbA1c and either PDW or NLR in the overall analysis, although subgroup analyses revealed notable

Table 1. Baseline characteristics

Variable	Total (N=160)
Sex, n (%)	
Male	69 (43.1)
Female	91 (56.9)
Age (years), Mean ± SD (Range)	50.64 ± 13.58 (18 – 65)
Glycemic parameters	
FBS (mg/dL), Mean ± SD (Range)	112.1 ± 31.96 (72 – 284)
HbA1c (%), Mean ± SD (Range)	6.2 ± 1.34 (4.30 – 12.0)
Glycemic control status, n (%)	
Controlled (HbA1c < 7%)	113 (70.6)
Uncontrolled (HbA1c ≥ 7%)	47 (29.4)
Hematological indices, Mean ± SD (Range)	
WBC ($\times 10^3/\mu L$)	7.62 ± 1.85 (4.03 – 13.38)
RBC ($\times 10^6/\mu L$)	4.82 ± 0.61 (3.17 – 7.32)
Platelet ($\times 10^3/\mu L$)	245.63 ± 55.02 (77 – 414)
Hb (g/dL)	13.76 ± 1.70 (6.5 – 17.6)
MCV (fL)	86.3 ± 7.32 (60 – 102)
RDW (%)	13.48 ± 1.84 (11.2 – 23.8)
PDW (fL)	12.94 ± 2.11 (9.2 – 19.6)
MPV (fL)	10.76 ± 0.90 (9.2 – 13.5)
NLR	1.93 ± 0.75 (0.37 – 6.47)

Abbreviations: FBS, Fasting blood sugar; HbA1c, Glycated hemoglobin; WBC, White blood cell; RBC, Red blood cell; Hb, Hemoglobin; MCV, Mean corpuscular volume; RDW, Red cell distribution width; PDW, Platelet distribution width; MPV, Mean platelet volume; NLR, Neutrophil-to-lymphocyte ratio. Data are presented as Mean ± SD (Min-Max) or n (%).

Table 2. Comparison of indices by glycemic control

Variable	Controlled (n=113)	Uncontrolled (n=47)	*P*-value	Test applied
RDW (%)	13.37 ± 1.87	13.74 ± 1.77	0.188†	Mann-Whitney U
PDW (fL)	12.88 ± 2.07	13.08 ± 2.22	0.592	Independent t-test
MPV (fL)	10.76 ± 0.93	10.74 ± 0.84	0.873	Independent t-test
NLR	1.86 ± 0.81	2.10 ± 0.55	0.059	Independent t-test
Hb (g/dL)	13.86 ± 1.65	13.52 ± 1.80	0.246	Independent t-test

Data are presented as Mean ± SD. † Non-parametric test applied due to non-normal distribution (Kolmogorov–Smirnov test, $P<0.05$ for RDW). Abbreviations: RDW, Red cell distribution width; PDW, Platelet distribution width; MPV, Mean platelet volume; NLR, Neutrophil-to-lymphocyte ratio; Hb, Hemoglobin.

Table 3. Correlation matrix between hematological indices and clinical parameters

Variable	RDW (%)	PDW (fL)	NLR	MPV (fL)	Hb (g/dL)
FBS	r = -0.026	r = 0.045	**r = 0.197**	r = 0.043	r = -0.040
	P = 0.746	P = 0.572	**P = 0.012**	P = 0.591	P = 0.611
HbA1c	**r = 0.280**	r = 0.047	r = 0.111	r = 0.027	r = -0.078
	***P < 0.001**	P = 0.557	P = 0.161	P = 0.737	P = 0.328
WBC	r = 0.075	r = -0.103	**r = 0.288**	r = -0.115	r = -0.025
	P = 0.343	P = 0.195	***P < 0.001**	P = 0.147	P = 0.751
Platelet	r = 0.086	**r = -0.462**	r = -0.041	**r = -0.462**	r = -0.113
	P = 0.280	***P < 0.001**	P = 0.606	***P < 0.001**	P = 0.153
RBC	r = 0.147	r = 0.006	r = 0.008	r = -0.122	**r = 0.558**
	P = 0.064	P = 0.935	P = 0.925	P = 0.125	***P < 0.001**
Age	**r = 0.227**	r = 0.000	r = 0.084	r = 0.011	r = -0.136
	***P = 0.004**	P = 0.996	P = 0.288	P = 0.886	P = 0.086

Spearman's rho correlation was used for FBS/HbA1c with RDW/NLR/MPV/Hb; Pearson for others. Bold values indicate statistical significance. Abbreviations: FBS, Fasting blood sugar; HbA1c, Glycated hemoglobin; WBC, White blood cell; RBC, Red blood cell.

Table 4. Significant subgroup analyses

Subgroup	n (Controlled/Uncontrolled)	Variable	Controlled (Mean ± SD)	Uncontrolled (Mean ± SD)	*P*-value	Test
Sex: Male	48 / 21	NLR	1.82 ± 0.63	2.18 ± 0.48	**0.023**	t-test
Age: 18–40 years	34 / 2	RDW (%)	12.91 ± 1.25	16.05 ± 3.74	**0.038**†	Mann-Whitney U
FBS ≤ 100 mg/dL	72 / 4	RDW (%)	13.45 ± 2.03	15.80 ± 2.21	**0.009**†	Mann-Whitney U
WBC (4–12 ×10³/µL)	111 / 44	NLR	1.82 ± 0.68	2.11 ± 0.56	**0.013**	t-test

Non-parametric test. Only subgroups with statistically significant differences are shown (P < 0.05). Data are presented as Mean ± SD. Abbreviations: NLR, Neutrophil-to-lymphocyte ratio; RDW, Red cell distribution width; FBS, Fasting blood sugar; WBC, White blood cell. Note: Subgroup analyses with small sample sizes (n < 10 in uncontrolled group) should be interpreted with caution.

sex- and age-specific patterns that warrant further investigation.

4.1. RDW and glycemic control

A weak but statistically significant positive correlation was observed between RDW and HbA1c in the present study (r = 0.280), suggesting that RDW may reflect certain aspects of long-term glycemic status. However, given the modest correlation and the non-significant between-group difference, this association should be interpreted as complementary rather than standalone. In line with our findings, Adithya and Chitra demonstrated a significant positive correlation between RDW and HbA1c and suggested that RDW may provide additional information regarding glycemic status (11). Similarly, Mhirig et al. reported a significant association between RDW and HbA1c among Moroccan patients with type 2 diabetes and proposed that RDW may represent an inexpensive laboratory parameter that complements routine glycemic assessment (12). Likewise, Arkew et al. found significantly higher RDW values in patients with poor glycemic control than in those with adequate glycemic control, further supporting the po-

tential clinical relevance of this hematological parameter (13).

Several biological mechanisms may explain the observed association between RDW and glycemic control. Chronic hyperglycemia promotes non-enzymatic glycation of hemoglobin and erythrocyte membrane proteins, leading to reduced erythrocyte deformability, increased membrane rigidity, and shortened red blood cell survival (14, 15) . In addition, persistent oxidative stress and low-grade systemic inflammation contribute to increased heterogeneity in erythrocyte size, which is reflected by elevated RDW values (16). Supporting this concept, Wang et al. demonstrated in a large prospective cohort of 16,971 Chinese adults that each one-unit increase in RDW was associated with a 16% higher risk of incident diabetes (hazard ratio (HR) = 1.16, 95% confidence interval (CI): 1.06–1.26), indicating that RDW may reflect metabolic alterations associated with glucose dysregulation (5) .

Beyond glycemic control, RDW has also been associated with adverse clinical outcomes in patients with diabetes. Roumeliotis et al. reported that higher RDW was independently associated with increased risks of all-cause mortality, cardiovascular mortality, and progression to end-stage kidney disease among patients with diabetic kidney disease (HR = 1.75, 1.66, and 2.14, respectively), highlighting the broader prognostic significance of this readily available hematological parameter (17) .

Despite the significant positive correlation observed between RDW and HbA1c, the comparison between patients with controlled and uncontrolled glycemic status did not reach statistical significance. This apparent discrepancy may be explained by the relatively weak strength of the observed correlation, the predominance of participants with well-controlled diabetes, and the unequal distribution between the two glycemic groups. These factors may have reduced the statistical power to detect between-group differences despite the presence of a statistically significant overall association. Therefore, our findings suggest that RDW reflects glycemic status only to a limited extent and should be interpreted as a complementary laboratory parameter that may assist routine clinical assessment rather than a standalone marker of glycemic control (18). Interestingly, the association between RDW and glycemic control was observed only among younger participants (18–40 years), whereas no significant association was identified in older adults. This finding should be interpreted cautiously because of the limited subgroup sample size. Nevertheless, age-related comorbidities and chronic low-grade inflammation (“inflammaging”) may attenuate the specific influence of hyperglycemia on erythrocyte morphology in older individuals. Further prospective studies with larger and more balanced populations are warranted to confirm this age-specific observation.

4.2. Platelet indices (PDW and MPV) and diabetes

Contrary to our expectations, we found no significant correlation between HbA1c and PDW or MPV in the overall analysis. This contrasts with several recent studies that have reported elevated platelet indices in diabetic patients. Waghale et al. demonstrated that platelet volume indices (PVIs), including MPV, PDW, and platelet-large cell ratio (P-LCR), were significantly higher in diabetic patients compared to controls and correlated positively with carotid intima-media thickness (CIMT), implying cardiovascular risk (7). Similarly, a study by Taderegew et al. on 352 type 2 diabetic patients in Ethiopia found that MPV, PDW, and P-LCR were significantly increased in patients with microvascular complications compared to those without (all $P < 0.001$), and these indices remained independently associated with complications after multivariate adjustment (adjusted odds ratio (AOR) = 1.68, 1.37, and 1.07, respectively) (19).

Abd El-Ghany et al. reported that MPV and PDW were significantly higher among diabetic subjects with retinopathy, nephropathy, and neuropathy, with PDW > 16.9 fL demonstrating 74.5% sensitivity and 100% specificity for detecting microvascular complications (20). Furthermore, a 2021 study found that MPV, PDW, and plateletcrit (PCT) were significantly higher in diabetic individuals than in matched controls, and these indices correlated positively with HbA1c levels (8).

The discrepancy between our findings and these studies may be explained by differences in study populations and inclusion criteria. Our study rigorously excluded patients with significant inflammatory conditions, renal failure, and hematological disorders — all of which are known potent stimulators of platelet activation. It is plausible that platelet indices such as PDW and MPV are more sensitive to the presence of advanced vascular complications or significant systemic inflammation than to glycemic control alone. As Waghale et al. noted, PVI may serve as markers of subclinical atherosclerosis and predictors of future cardiovascular events rather than as direct indicators of glycemic status (7). The negative correlation we observed between platelet count and both PDW ($r = -0.462$) and MPV ($r = -0.462$)

is consistent with the known inverse relationship between platelet size and number, reflecting increased platelet turnover in activated states (21).

4.3. Neutrophil-to-lymphocyte ratio (NLR) and glycemic control

Although NLR was not significantly correlated with HbA1c (P = 0.161), it showed a weak but statistically significant positive correlation with FBS (r = 0.197, P = 0.012). This finding suggests that NLR may be more reflective of acute hyperglycemic spikes or short-term inflammatory responses rather than average long-term glycemic status.

This nuanced finding contrasts with several recent studies that have reported positive associations between NLR and HbA1c. Mohammadi et al. concluded that NLR provides very good and similar results to HbA1c for identifying early complications of uncontrolled diabetes (22). Selvaraju et al. found that NLR was significantly elevated in type 2 diabetic patients (5.35 vs. 2.16 in controls, P < 0.001) and that increased NLR was associated with elevated HbA1c and poor glycemic control (23). A systematic review and meta-analysis also suggested an association between high NLR values and elevated HbA1c in type 2 diabetes patients (24). Duman et al. similarly reported a significant correlation between NLR and HbA1c (25).

The discrepancy between our findings and these reports may be attributable to several factors. First, our study excluded patients with significant inflammatory comorbidities, which may have attenuated the NLR-HbA1c relationship. Second, NLR is influenced by a wide range of factors beyond glycemic control, including age, smoking status, and underlying infections — all of which were controlled for in our exclusion criteria. Third, the relatively moderate sample size may have limited our statistical power to detect weaker associations.

Importantly, our subgroup analysis revealed that NLR was significantly higher in the uncontrolled group only among male patients (P = 0.023), a finding not observed in females. This sex-specific difference is intriguing and may reflect hormonal influences on inflammatory responses. Testosterone has been shown to have immunomodulatory effects, and sex differences in body composition, insulin resistance patterns, and adipose tissue inflammation could contribute to this disparity (26). Further research is warranted to explore the mechanisms underlying this observation.

NLR has also been implicated in diabetic complications beyond glycemic control. Assulyn et al. demonstrated that NLR and RDW have positive predictive value for microalbuminuria in diabetic patients, suggesting their potential role in predicting diabetic nephropathy (10). A 2022 study found that NLR was significantly increased in type 2 diabetes patients with diabetic nephropathy, indicating that inflammation and endothelial dysfunction could be integral parts of the pathogenesis of diabetic nephropathy (27). Additionally, the presence of type 2 diabetes with elevated NLR has been associated with worse clinical outcomes in coronary artery disease patients undergoing percutaneous coronary intervention (28).

4.4. Clinical implications

The findings of this study have several practical implications. First, RDW is a routine, inexpensive, and widely available hematological parameter that may provide complementary clinical information when interpreted together with HbA1c. However, given the modest strength of the observed association, RDW should not be considered a substitute for established measures of long-term glycemic control. Second, the absence of significant associations between PDW or NLR and HbA1c suggests that these indices are unlikely to be reliable markers of glycemic control in the general diabetic population, although they may still have value in selected clinical settings or specific patient subgroups. However, given the exploratory nature of this study and the lack of multivariable adjustment, these implications should be considered preliminary and hypothesis-generating rather than definitive.

5. Limitations

Several limitations should be acknowledged. The cross-sectional design precludes causality. Restrictive exclusion criteria limit generalizability. Key confounders (iron, B12/folate, thyroid, liver) were not assessed. Despite excluding known anemia, Hb levels as low as 6.5 g/dL suggest undiagnosed anemia may confound RDW. Convenience sampling may cause selection bias. No multivariable adjustment was performed; therefore, we cannot claim an independent association. Subgroup analyses had very small samples (e.g., n=2), warranting caution. The predominance of well-controlled participants (mean HbA1c=6.2%) and unequal group distribution (113 vs. 47) reduced statistical power. No correction was applied for multiple comparisons; secondary findings are hypothesis-generating. Despite these limitations, this study provides valuable prelimi-

nary data from an underrepresented population.

6. Conclusion

In conclusion, this study demonstrated a weak but statistically significant positive correlation between RDW and HbA1c in adults with type 2 diabetes mellitus. However, the non-significant difference in RDW between controlled and uncontrolled glycemic groups, combined with the modest correlation coefficient, suggests that RDW should not be used as a standalone or substitute marker for glycemic control. At best, RDW may provide complementary information when interpreted alongside established measures such as HbA1c. PDW and NLR were not consistently associated with glycemic control and are unlikely to be reliable indicators of glycemic status in this population. Further large-scale prospective studies with multivariable adjustments and broader inclusion criteria are warranted to clarify the clinical utility of these readily available hematological indices.

7. Declarations

7.1. Ethical approval and consent to participate

The study was approved by the Ethics Committee of Guilan University of Medical Sciences **(Approval No. IR.GUMS.REC.1403.490)** and was conducted in accordance with the Declaration of Helsinki. Written informed consent was obtained from all participants prior to enrollment.

7.2. Availability of data and materials

The datasets generated and/or analyzed during the current study are available from the corresponding authors upon reasonable request.

7.3. Conflict of interest

The authors declare that they have no competing interests.

7.4. Financial support

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors. This research was self-funded.

7.5. Author's contributions

VL contributed to the study conceptualization, data collection, and investigation. SA contributed to the study design, clinical supervision, methodology, and manuscript review and editing. BM contributed to the clinical investigation, data interpretation, and manuscript review. ZAR contributed to the methodology, statistical analysis, and interpretation of the results. All authors have read and approved the final version of the manuscript and agree to be accountable for all aspects of the work.

7.6. Artificial intelligence statement

The authors declare whether AI or AI-assisted technologies were used during the preparation of this manuscript. If used, AI tools were employed solely to improve language quality, grammar, readability, and organizational structure. The authors carefully reviewed and edited all AI-generated content and take full responsibility for the accuracy, integrity, and originality of the final manuscript. No AI tool was used to generate, analyze, or interpret scientific data or images, or to draw scientific conclusions. The use of AI-assisted technologies complies with current publication ethics recommendations and journal policies.

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