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ARE ROUTINE HAEMATOLOGICAL PARAMETERS- MEAN PLATELET VOLUME (MPV), NEUTROPHIL-TO-LYMPHOCYTE RATIO (NLR), AND WHITE BLOOD CELL COUNT (WBC) RELIABLE BIOMARKERS FOR RECURRENT APHTHOUS STOMATITIS (RAS)? - A SYSTEMATIC REVIEW AND META-ANALYSIS

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ABSTRACT

Background: Recurrent Aphthous Stomatitis (RAS) is a common, painful inflammatory disorder of the oral mucosa with a multifactorial etiology. Routine haematological parameters such as Mean Platelet Volume (MPV), Neutrophil-to-Lymphocyte Ratio (NLR), and White Blood Cell Count (WBC) have been proposed as potential biomarkers reflecting systemic inflammation in RAS. However, individual studies report inconsistent findings.

Objective: Cumulative assessment of the haematological parameters like mean platelet volume (MPV), neutrophil-to lymphocyte ratio (NLR), and total white blood cell count (WBC) across studies in recurrent aphthous stomatitis (RAS).

Methods: A systematic literature search was conducted in PubMed, Cochrane Central, and Google Scholar up to 16 September 2025. Case-control and cross-sectional studies reporting MPV, NLR, and/or WBC in RAS patients versus controls were included. Study quality was assessed using the AXIS tool. Random-effects meta-analysis was performed using Review Manager 5.4, with outcomes reported as standardized mean differences (SMD) and 95% confidence intervals (CI). A p-value of <0.05 was considered statistically significant for the overall pooled effect.

Results: Six studies (total n=828 participants) met the inclusion criteria. Meta-analysis showed a non-significant trend towards higher MPV in RAS (SMD = 1.09; 95% CI: -0.04 to 2.22; p=0.06) with extreme heterogeneity ($I^2 = 98\%$). NLR was significantly elevated in RAS patients (SMD = 0.39; 95% CI: 0.04 to 0.75; p=0.03), though heterogeneity was substantial ($I^2 = 79\%$). No significant difference was found in WBC count (SMD = 0.23; 95% CI: -0.45 to 0.90; p=0.51; $I^2 = 94\%$).

Conclusion: NLR appears to be a promising inflammatory marker linked to RAS, hinting at low-grade, ongoing inflammation. Right now, there's just not enough solid evidence to say that MPV or WBC are useful biomarkers due to the inconsistent results. Well-designed studies will be required to establish the clinical importance of these biomarkers.

INTRODUCTION

Recurrent Aphthous Stomatitis (RAS) is an ulcerative condition of the oral mucosa which affects around 5% to 66% of the general population particularly children and adolescents [1]. It is recognized by the presence of recurrently occurring painful ulcers which are round to ovoid in shape with a necrotic centre and an erythematous halo. It occurs predominantly on the buccal mucosa, lips, and tongue [2]. RAS is classified into three main types: Minor (80%) Major (10%), and Herpetiform (1 to 10%) [3].

The etiopathogenesis of RAS is multifactorial and not completely understood. It involves a complex interaction of genetic factors, immunological disturbances, microbial factors, nutritional deficiencies (e.g., iron, vitamin B12, folic acid), hormonal changes, psychological stress, and local trauma [4]. An elevated Th1-dominant cytokine profile (elevated IL-2, IFN-γ, TNF-α), is responsible for the inflammatory process which leads to mucosal breakdown [5].

Due to the inflammatory nature of RAS, there has been a growing interest in identifying easily available and cost-effective systemic biomarkers that could help in diagnosis, reflect disease activity, and explain the pathophysiology. Routine complete blood count (CBC) parameters such as Mean Platelet Volume (MPV), Neutrophil-to-Lymphocyte Ratio (NLR), and total White Blood Cell Count (WBC), have emerged as potential candidates. MPV is a marker of platelet activation and size and it is often elevated in pro-inflammatory and pro-thrombotic states [6]. NLR is calculated from differential

counts and it gives information on innate immunity (neutrophils) and adaptive immune regulation (lymphocytes), serving as a marker of systemic inflammation [7]. WBC count is a classic indicator of inflammation.
Individual studies investigating these parameters in RAS have given conflicting results. Some report significant elevations in MPV and NLR during active phases [8, 9], while others find no differences compared to healthy individuals [10, 11]. Due to this inconsistency, there is a need for a synthesized evidence-based assessment.
Therefore, this systematic review and meta-analysis aim to evaluate and summarize the published evidence on difference in mean platelet volume (MPV), neutrophil-to lymphocyte ratio (NLR), and total white blood cell count (WBC) between recurrent aphthous stomatitis (RAS) and healthy controls.

MATERIALS AND METHODS

Once the desired aims & objectives has been set, a rigorous search on online databases, (PubMed, Cochrane central and Google Scholar) was performed by (R.K and R.M) & relevant clinical trials were selected with filters applied in relation to below mentioned inclusion and exclusion criteria. The available studies were further scrutinized following the PRISMA guidelines and final studies were selected for systematic review. This review was conducted according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2021 guidelines and was registered in the PROSPERO international prospective register (CRD420251144530).

ELIGIBILITY CRITERIA

Inclusion Criteria:

1. Non-randomized control trials (prospective or retrospective), case-control, or cross-sectional studies.
2. Studies comparing haematological parameters (MPV, NLR, and/or WBC) between clinically diagnosed RAS patients and healthy controls.
3. Studies involving all age groups, genders, and geographical locations.
4. Full-text articles published in English from 1 January 2000 to 16 September 2025.

Exclusion Criteria:

1. Studies on patients with Behçet's disease, systemic inflammatory conditions (e.g., Crohn’s disease, HIV), or a history of smoking/alcohol abuse.
2. Studies published before 2000 or in languages other than English.
3. Articles where full-text was unavailable or did not report the parameters of interest.
4. Case reports, case series, reviews, and editorials

Table No.1: Search strategy

Sr. No.	Category	Search Terms
1.	Population	Recurrent Aphthous Stomatitis * [Title/Abstract] OR Oral Recurrent Aphthous Stomatitis *[Title/Abstract] OR Oral Ulcers* [Title/Abstract]
2.	Exposure	Idiopathic*[Title/Abstract] OR Multifactorial* [Title/Abstract]
3.	Comparison	Healthy Controls *[Title/Abstract] OR Healthy Individuals *[Title/Abstract]
4.	Outcome	Mean Platelet Volume *[Title/Abstract] AND Neutrophil-to-Lymphocyte Ratio* [Title/Abstract] AND White Blood Cell Count *[Title/Abstract]
5.	Study Design	Non randomized* trial* OR Case Control Study OR Cohort Study
		1AND 2 AND 3AND 4AND 5

STUDY SELECTION

Two independent reviewers (R.K. and R.M.) screened titles, abstracts, and full texts. Discrepancies were resolved by consensus or consultation with a third reviewer. A standardized data extraction form was used to collect: author, year, country, study design, sample size, demographic details of RAS and control groups, and mean values with standard deviations for MPV, NLR, and WBC.

DATA EXTRACTION

Data collection process:

Data collection was performed using a customized data extraction form.

Data extraction form:

Contents-

1.Author Name
2.Publication Year
3.Country
4.Journal Name
5.Language Of Publication
6.Study Setting
7.Study Period
8.Study Design
9.Sample Size
10.RAS group
11.Female In RAS
12.Male in RAS
13.Control Group
14.Female Control
15.Male Control
16.Mean Age in RAS
17.Mean Age in Healthy Controls
18.MPV value in RAS
19.MPV value in Healthy Controls
20.NLR in RAS
21.NLR in Healthy Controls
22.Other observations in RAS group
23.Other observations in control group

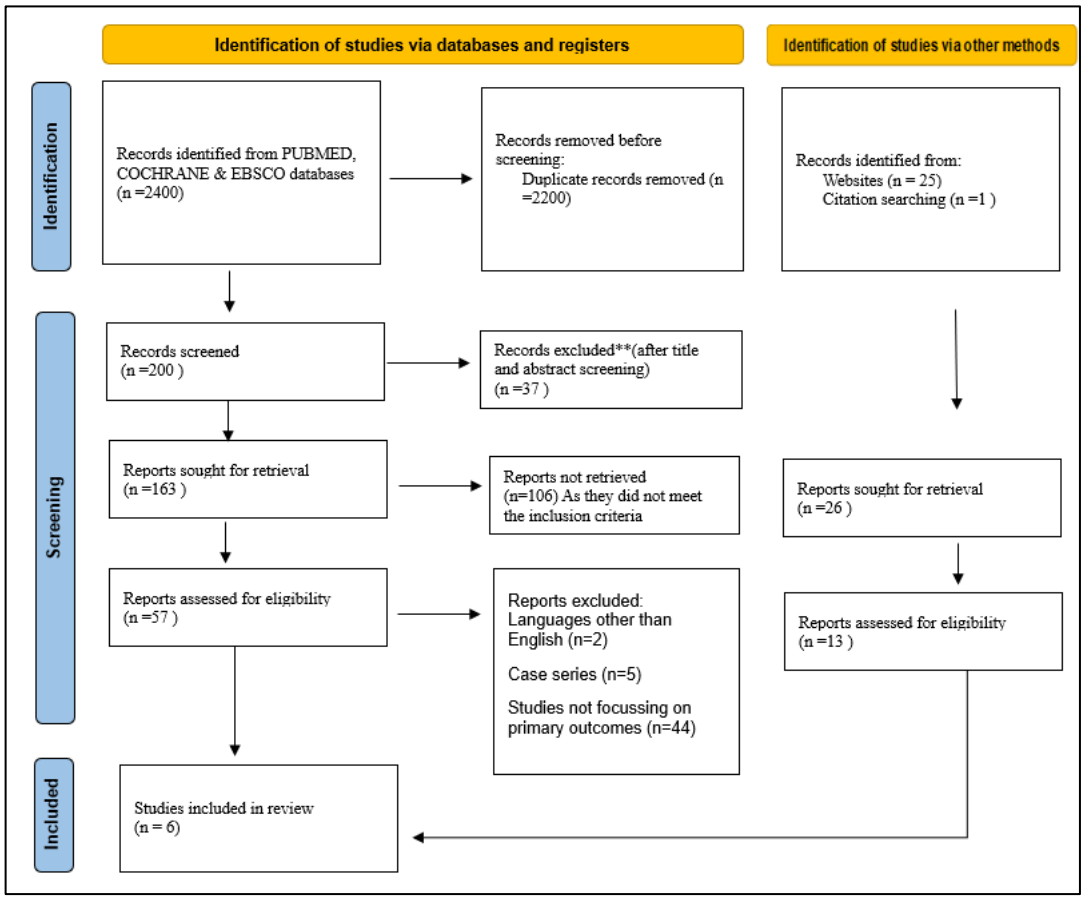


Figure 1: PRISMA Flow Diagram of Study Selection

2400 papers in all were found after PUBMED and COCHRANE screening. There were 200 papers remaining from the original count of 2400 after duplicate entries (2200) were eliminated. Following the screening of titles and abstracts, the eligibility of 57 full-text publications was assessed. After the entire content was read, 51 articles were deleted since they did not meet the conditions to be included. In the end, six articles underwent qualitative analysis and quantitative synthesis analysis.

STUDY CHARACTERISTICS

Table No.2: List of selected studies for systematic review & related findings

Sr. No.	Year / Author(s)	Sample Size	Study Design	Study Group	Control Group	Main Outcomes	Additional Outcomes
1	2017, Sinan Uluyol, Saffet Kilicaslan	107	Retrospective	Active RAS, Inactive RAS	Healthy controls	**MPV (fL):** Active RAS: 10.6 ± 2.9; Inactive RAS: 7.1 ± 2.4; Controls: 6.9 ± 2.1 **NLR:** Active RAS: 3.74 ± 1.9; Inactive RAS: 2.1 ± 1.43; Controls: 2.07 ± 0.96 WBC not assessed	Not assessed
2	2016, Murat Sereflican et al.	73	Case–Control	RAS	Healthy controls	**MPV (fL):** RAS: 8.96 ± 1.27; Controls: 8.29 ± 1.32 **NLR:** RAS: 2.12 ± 0.65; Controls: 2.26 ± 1.02 **WBC:** RAS: 6.64 ± 1.9; Controls: 7.21 ± 2.11	CRP = 0.2; Vitamin B12 = 287.5; Folic acid = 5.4
3	2023, Mujtaba Nadeem, Usman Manzoor, Uzma Tariq, Sufyan Ahmed, Awais Akbar, Syed Aijaz Ali Zaidi	112	Case–Control	RAS	Healthy controls	**MPV (fL):** RAS: 8.86 ± 0.33; Controls: 6.95 ± 0.20 NLR, WBC not assessed	Not assessed
4	2020, Deniz Avci	102	Case–Control	RAS	Healthy controls	**MPV (fL):** RAS: 8.82 ± 0.87; Controls: 8.82 ± 0.87 **NLR:** RAS: 1.94 ± 0.74; Controls: 1.80 ± 0.80 **WBC:** RAS: 6.96 ± 1.99; Controls: 6.65 ± 1.83	ANC = 3.78 ± 1.43; ALC = 2.19 ± 0.59; PLR = 121.98 ± 32.96
5	Isil Cakmak Karaer	274	Case–Control	RAS	Healthy controls	**MPV (fL):** RAS: 9.8 ± 0.17; Controls: 10 ± 0.19 **NLR:** RAS: 1.8 (1.4–2.5); Controls: 1.6 (1.2–2.1) **WBC:** RAS: 6.9 ± 0.34; Controls: 6.96 ± 0.35	PLR = 129; Vitamin B12 = 346.4 ± 24; CRP = 0.23; ESR = 10.7 ± 1.19
6	2016, Suat Terzi et al.	160	Case–Control	RAS	Healthy controls	**MPV (fL):** RAS: 8.1 ± 1.8; Controls: 8.70 ± 1.2 **NLR:** RAS: 3.37 ± 1.74; Controls: 1.95 ± 1.21 **WBC:** RAS: 8.65 ± 1.78; Controls: 6.69 ± 1.51	PLR = 120.7 ± 46.02

RISK OF BIAS ASSESSMENT

The methodological quality and risk of bias of the included cross-sectional studies were evaluated using the AXIS tool (Appraisal Tool for Cross-Sectional Studies) developed by Downes et al.

Table No 3: Risk of bias results

AXIS Item	Avcı 2020	Nadeem et al. 2023	Karaer 2020	Şereflican et al. 2016	Terzi et al. 2016	Uluyol & Kilicaslan 2019
1. Aims/objectives clear	Yes	Yes	Yes	Yes	Yes	Yes
2. Design appropriate	Yes	Yes	Yes	Yes	Yes	Yes
3. Sample size justified	No	No	No	No	No	No
4. Target population defined	Yes	Yes	Yes	Yes	Yes	Yes
5. Sampling frame appropriate	Yes	No	No	No	No	No
6. Selection representative	No	No	No	No	No	No
7. Non-responders addressed	Don't know	No	No	No	Don't know	No
8. Measurements appropriate	Yes	Yes	Yes	Yes	Yes	Yes
9. Measurements reliable	Yes	Yes	Yes	Yes	Yes	Yes
10. Statistical significance defined	Yes	Yes	Yes	Yes	Yes	Yes
11. Methods reproducible	Yes	Yes	Yes	Yes	Yes	Yes
12. Basic data described	Yes	Yes	Yes	Yes	Yes	Yes
13. Response rate concerns	Don't know	No	No	Don't know	Don't know	Don't know
14. Info on non-responders	Don't know	No	No	Don't know	Don't know	Don't know
15. Results internally consistent	Yes	Yes	Yes	Yes	Yes	Yes
16. Results complete	Yes	Yes	Yes	Yes	Yes	Yes
17. Conclusions justified	Yes	Yes	Yes	Yes	Yes	Yes

18. Limitations discussed	Yes	Yes	No	No	No	Yes
19. Conflicts/funding declared	Yes	Yes	No	Don't know	Don't know	Yes
20. Ethics approval/consent	Yes	Yes	Yes	Yes	Yes	Yes
Overall Risk of Bias	**Low-to-moderate**	**Low-to-moderate**	**Low-to-moderate**	**Moderate**	**Moderate**	**Low-to-moderate**

Overall Summary of RoB Results

The majority of the included cross-sectional studies demonstrated clearly defined aims, appropriate study designs, valid and reliable measurements, reproducible methods, and complete results. Almost all studies obtained ethical approval and informed consent.

However, common limitations were identified across studies:

- Lack of sample size justification was consistent in all.
- Recruitment restricted to single centres reduced representativeness and generalizability.
- Non-responders were not addressed, and response rate information was largely absent.
- Conflicts of interest and funding declarations were inconsistently reported.
- Limitations were either brief or incomplete.

None of the studies could be considered having a low risk of bias because of these issues. Most studies were considered as having a low-to-moderate risk of bias, while a few (e.g., Şereflican 2016, Terzi 2016) were assessed as having a moderate risk of bias.

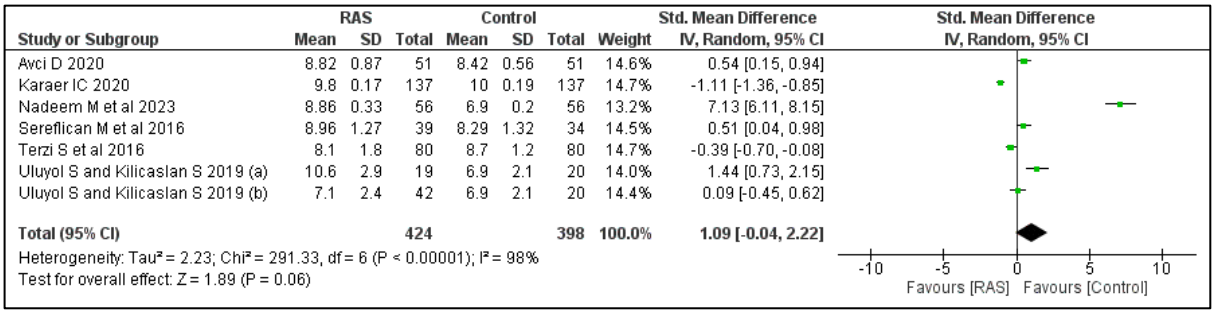
RESULTS

Statistical analysis:

The meta-analysis was done with Review Manager (RevMan, version 5.4, The Cochrane Collaboration, Copenhagen, Denmark). For continuous variables, the standardized mean difference (SMD) with 95% confidence intervals was used. A random-effects model was used to assess heterogeneity across studies. To check for statistical heterogeneity, Chi-square test (Cochran's Q) was used and the I^2 statistic—anything over 50% on I^2 meant there was a good deal of heterogeneity. If the p-value was under 0.05, the overall pooled effect was statistically significant.

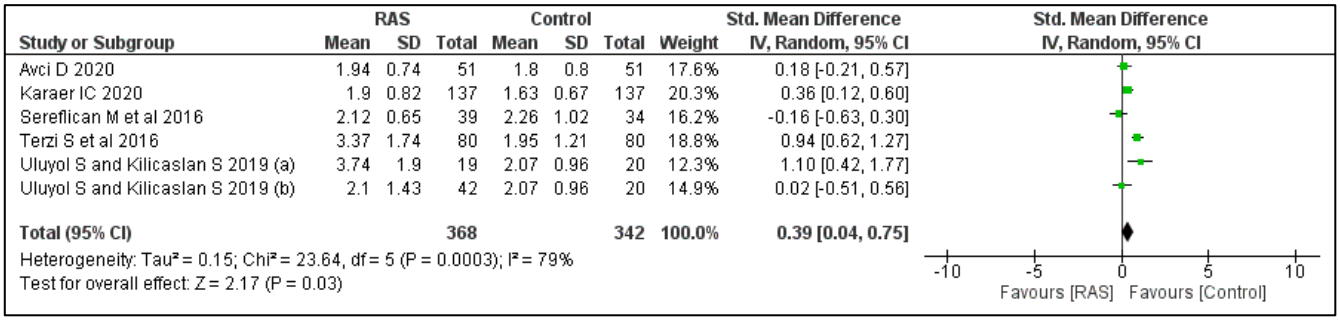
Mean Platelet Volume (MPV)

The MPV analysis includes six investigations (Avci D 2020, Karaer IC 2020, Nadeem M et al 2023, Sereflican M et al 2016, Terzi S et al 2016, Uluyol S and Kilicaslan S 2019). Although RAS was favored overall, the impact magnitude was not statistically significant (SMD: 1.09; 95% CI: -0.04 to 2.22; p = 0.06). Significant discrepancy was suggested by high heterogeneity among trials (I^2 = 98%, p < 0.00001).



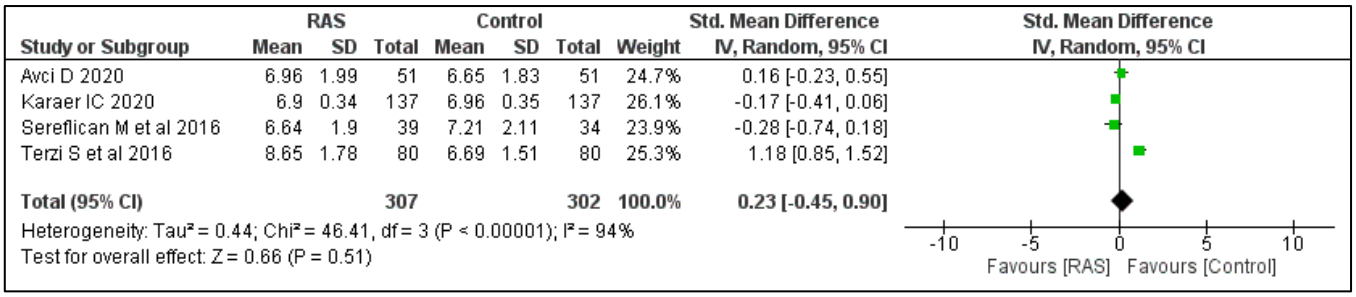
Neutrophil-to-Lymphocyte Ratio (NLR)

NLR was examined in six investigations (Avci D 2020, Karaer IC 2020, Sereflican M et al 2016, Terzi S et al 2016, Uluyol S, and Kilicaslan S 2019). NLR was significantly higher in RAS patients than in controls (SMD: 0.39; 95% CI: 0.04 to 0.75; p = 0.03). Significant heterogeneity (I^2 = 79%, p = 0.0003) revealed variations in the study's findings. The results indicate that NLR might be a possible inflammatory marker linked to RAS irrespective of this variability.



White Blood Cell Count (WBC)

There was no statistically significant difference among WBC levels between patients with RAS and healthy controls, according to a pooled analysis of four studies (Avci D 2020, Karaer IC 2020, Sereflican M et al 2016, Terzi S et al 2016) (SMD: 0.23; 95% CI: -0.45 to 0.90; p = 0.51). There was significant variation among the included studies, as shown by the extremely high heterogeneity (I^2 = 94%, p < 0.00001).



DISCUSSION

This systematic review and meta-analysis synthesized available evidence on three routine haematological parameters as potential inflammatory biomarkers in Recurrent Aphthous Stomatitis. The key finding is that NLR was significantly elevated in RAS patients compared to healthy controls, suggesting its role as a marker of systemic inflammation. In contrast, MPV showed a non-significant increasing trend, and WBC showed no significant difference, with both parameters exhibiting considerable heterogeneity.

Neutrophil-to-Lymphocyte Ratio (NLR) emerged as the most consistent biomarker. Elevated NLR aligns with the recognized immunopathology of RAS, which involves neutrophilic infiltration at ulcer sites and a dysregulated T-cell response [5]. NLR integrates two immune pathways: neutrophils represent the acute, pro-inflammatory innate response, while lymphocytes modulate adaptive immunity. An elevated NLR indicates a relative neutrophilia or lymphopenia, often seen in chronic inflammatory states [7]. This finding is consistent with studies in other inflammatory conditions like Behçet's disease, where NLR correlates with disease activity [16]. The clinical appeal of NLR lies in its simplicity, low cost, and routine availability from a standard CBC, making it a practical tool for assessing subclinical inflammation in RAS.However, the substantial heterogeneity (I^2 = 79%) cautions against overgeneralization. Variability may stem from differences in disease activity at sampling (active vs. remission), ulcer type (minor vs. major), patient age, and pre-analytical laboratory protocols. Future studies should standardize the timing of blood draw relative to ulcer activity and report NLR values stratified by clinical severity.

Mean Platelet Volume (MPV) showed a large but non-significant effect size with extreme heterogeneity (I^2 = 98%). MPV is a marker of platelet activation and size; larger platelets are more metabolically active and pro-thrombotic [6]. Some studies included in this review, particularly those sampling during active ulceration, reported significantly higher MPV [8, 9], suggesting platelet activation may occur during inflammatory flares. However, other well-controlled studies found no difference [10, 11]. The extreme heterogeneity likely reflects methodological differences (e.g., anticoagulant used, time to analysis) known to affect MPV measurements, as well as population-specific factors. Therefore, MPV cannot currently be recommended as a reliable standalone biomarker for RAS without standardized protocols.

White Blood Cell Count (WBC) did not differ between groups. This is unsurprising, as total WBC is a crude measure and can be influenced by numerous transient factors (e.g., minor infections, stress, diurnal variation) unrelated to RAS-specific pathology. The very high heterogeneity underscores its lack of specificity. The differential count (from which NLR is derived) appears more informative than the total count alone.

CLINICAL IMPLICATIONS AND RESEARCH GAPS

Elevated NLR values in RAS patients suggests that it is not a localized condition and that there is a possibility of underlying systemic inflammation. NLR levels can be used to assess diseases activity or treatment response in clinical practice after the diagnostic cut off values are determined.

Limited global representativeness was present as all the studies were only from two countries. There was lack of justification of the sample size and other factors like nutritional status, stress and subclinical infections. Temporal sequence between biomarker changes and ulcer onset/remission was not inferred as all studies were longitudinal.

STRENGTHS AND LIMITATIONS

This is the first meta-analysis, to our knowledge, to quantitatively synthesize evidence on MPV, NLR, and WBC in RAS. The review followed a pre-registered protocol and PRISMA guidelines, with a dual-reviewer process to minimize bias.
The limitations of this meta-analysis mirror those of the primary studies. The small number of included studies and high statistical heterogeneity are major constraints, precluding meaningful subgroup or meta-regression analyses. The cross-sectional nature of the evidence prevents causal inference. Publication bias could not be formally assessed due to the limited number of studies

CONCLUSION

This meta-analysis shows that NLR is significantly elevated in patients with RAS as compared to healthy individuals, hence it can be used as a biomarker in the assessment of systemic inflammation. The evidence for MPV and WBC count was not consistent.
There is not yet enough strong evidence to use NLR routinely in patient care even through it is appears to be a promising diagnostic and prognostic biomarker as more high-quality studies are needed to prove that it is truly useful and reliable.

TABLE LEGENDS

Table No.1: Search strategy
Table No.2: List of selected studies for systematic review & related findings
Table No 3: Risk of bias results

FIGURE LEGENDS

Figure 1: PRISMA Flow Diagram of Study Selection

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DECLARATIONS

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

Ethical Approval: Not applicable (systematic review of published data).\

Data Availability: The data extracted from included studies are available within the manuscript and its supplementary tables.

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