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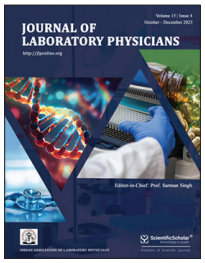

Original Article

Enhancing diagnostic precision in psoriasis skin lesions: The role of ki-67 and a three-tier grading approach

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ABSTRACT

Objectives: To evaluate the diagnostic utility of Ki-67 expression in distinguishing psoriasis from non-psoriatic psoriasiform dermatoses (NPPD) and to establish a cutoff value that supports diagnosis alongside histopathological features and to assess and categorize psoriasis and NPPD based on histomorphological characteristics and the proportion of inflammatory cells using a three-tier histological grading approach.

Materials and Methods: This study was conducted over a year, incorporating retrospective data from 6 months and prospective data collected between February 2024 and June 2024. A total of 32 cases were included, comprising 20 cases of psoriasis and 12 cases of NPPD. All skin biopsies were graded using a three-tier histological grading, and Ki-67 immunohistochemistry (IHC) was performed for each case. The results were then compared to assess diagnostic utility.

Statistical analysis: The results were statistically analyzed using the Chi-square test, and analysis of variance variables assessed for their association with Ki-67 expression, including histopathological features such as hyperkeratosis, parakeratosis, regular acanthosis, hypo- or agranulosis, and the grade of inflammatory cell infiltrate. Both the percentage and intensity of Ki-67 expression were compared between psoriasis and NPPD to evaluate their diagnostic utility.

Results: In the diagnosis of psoriasis and psoriasiform lesions, Ki-67 demonstrated a wide diagnostic cutoff range. In our study, Ki-67 immunohistochemistry showed that all 20 psoriasis cases exhibited Ki-67 expression with more than 60% positivity. In contrast, the majority of psoriasiform cases were negative for Ki-67, with only 16% showing expression above 50%, indicating a statistically significant difference ($P < 0.05$). Histopathological grading using our three-tier histological grading revealed that confluent parakeratosis, regular acanthosis, and agranulosis were significantly more common in psoriasis cases. Minimal dermal inflammation was also more frequently observed in psoriasis. However, hyperkeratosis did not show a significant difference between the two groups.

Conclusions: This study emphasizes the utility of our structured three-tier histological grading combined with Ki-67 immunostaining in distinguishing psoriasis from NPPD. A Ki-67 cutoff of 60% and above may enhance diagnostic accuracy in cases with overlapping clinical and histological features.

Keywords: Histopathology, Ki-67 antigen, Non-psoriatic psoriasiform dermatoses, Psoriasis

INTRODUCTION

Psoriasis is a chronic immune-mediated inflammatory skin disorder with a multifactorial etiology involving genetic, immunological, and environmental factors. Clinically, it manifests as

scaly, erythematous plaques commonly affecting the elbows, knees, scalp, and lower back. The most prevalent form is plaque psoriasis, while guttate, pustular, and erythrodermic variants are less common.[1]

Histopathologically, psoriasis is characterized by marked keratinocyte hyperproliferation, parakeratosis, acanthosis, and a dense dermal inflammatory infiltrate.[2,3] However, these features may also be seen in other non-psoriatic psoriasiform dermatoses (NPPD) such as lichen simplex chronicus, seborrheic dermatitis, pityriasis rubra pilaris, and certain drug reactions, leading to diagnostic overlap.[4-6]

Due to these overlapping clinical and histological features, particularly in cases presenting with erythroderma or longstanding lesions, reliable diagnostic markers are essential. Ki-67, a nuclear protein expressed during active phases of the cell cycle, serves as a robust immunohistochemical (IHC) marker for cellular proliferation. Its expression in psoriasis is typically elevated and extends beyond the basal layer into the suprabasal epidermis, contrasting with the limited basal staining seen in other NPPD.[7-10]

Several studies have demonstrated the diagnostic utility of Ki-67 IHC in distinguishing psoriasis from its mimickers.[7,11,12] High Ki-67 labeling indices in psoriasis may aid in narrowing differential diagnoses when histological features alone are inconclusive. Although definitive cutoff values for Ki-67 expression remain to be standardized, its adjunctive role in dermatopathology is increasingly recognized.[13,14]

This study evaluates Ki-67 expression in psoriasis and NPPD, correlating findings with histopathological patterns and inflammatory grading. The ultimate goal is to enhance diagnostic accuracy and contribute to establishing Ki-67 as a reliable marker in routine dermatopathological evaluation.

MATERIALS AND METHODS

This study was conducted over 1 year, incorporating retrospective data from 6 months and prospective data collected between February 2024 and June 2024.

Inclusion and exclusion criteria

The study included all skin biopsies with clinical and histopathological diagnosis of psoriasis and psoriasiform lesions.

The study excluded skin biopsies with psoriasis lesions with coexisting dermatological diseases.

In this study, cases included were those of clinically and histopathologically proven psoriasis, and a comparison of them with cases of NPPD.

Among the total of 32 cases included, 20 cases were of psoriasis and 12 cases were of NPPD.

Clinical diagnosis of psoriasis was made by a dermatologist based on features such as well-demarcated erythematous plaques with silvery-white scales, typical distribution over extensor surfaces, scalp, or trunk, and additional findings including nail changes, Auspitz's sign, and the Koebner phenomenon. Relevant clinical history, such as familial psoriasis, psoriatic arthritis, or known triggers, was also considered. These criteria align with those described by Ibad *et al.*[6]

Only those cases that fulfilled both clinical and histopathological criteria were included in the psoriasis group. Cases lacking such clinical features or history and confirmed histopathologically as NPPD were included in the NPPD group.

All samples were appropriately anonymized to prevent bias. Then skin biopsies were graded using a three-tier histological grading [Tables 1 and 2], and Ki-67 immunohistochemistry was performed for each case and graded with a broad category with interobserver agreement.

Histopathological findings for grading used in Table 1 were categorized as: regular acanthosis, confluent parakeratosis, and hypogranulosis or near agranulosis. These were assessed using a three-tier histological grading, with grading based on semi-quantitative criteria to enhance consistency.[7]

For example, in the case of regular acanthosis, a mild change (Grade 1) is referred to as mildly elongated rete ridges with subtle suprapapillary thinning in a single high-power field (HPF), whereas a moderate change (Grade 2) is indicated by pronounced elongation and widening of rete ridges with

Table 1: Three-tier histological grading of histopathological findings.

Grade	Histopathological findings
0	No changes
1	Mild isolated changes focally in one high-power field.
2	Moderate significant changes in one high-power field
3	Moderate to significant changes in nearly all microscopic fields/entire biopsy

Table 2: Three-tier histological grading for proportion of inflammatory cells.

Grade	Proportion of inflammatory cells
Grade 0	No infiltrate when the median count is<10 in all hotspot areas
Grade 1	Mild infiltrate when the median count is from 10 to 30 in all hotspot areas
Grade 2	Moderate infiltrate when the median count is from 31 to 50 in all hotspot areas
Grade 3	Marked infiltrate (severe inflammation) when the median count is >50 in all hotspot areas

prominent suprapapillary thinning in a single HPF. Marked changes (Grade 3) reflected similar Grade 2 findings present throughout most or all fields of the biopsy.

In the case of parakeratosis, a mild change (Grade 1) referred to 1-2 layers of retained nuclei in stratum corneum seen in a single HPF, whereas a moderate change (Grade 2) indicated by 3-5 layers of retained nuclei in stratum corneum seen in single HPF. Marked changes (Grade 3) reflected similar Grade 2 findings present throughout most or all fields of the biopsy.

In the case of near agranulosis, a mild change (Grade 1) is referred to as thinning of the granular layer seen in a single HPF, whereas a moderate change (Grade 2) is indicated by the complete absence of the granular layer seen in a single HPF. Marked changes (Grade 3) reflected similar Grade 2 findings present throughout most or all fields of the biopsy.

The histopathological features were independently reviewed and graded by four dermatopathologists, using the predefined three-tier histological grading. The results were then compared to assess diagnostic utility.

Statistical analysis

The results were statistically analyzed using the Statistical Package for Social Sciences. Variables assessed for their association with Ki-67 expression included histopathological features such as hyperkeratosis, parakeratosis, regular acanthosis, hypo- or agranulosis, and the grade of inflammatory cell infiltrate. Chi-square, *t*-test, and analysis of variance were used to assess the association of Ki-67 with clinical and histopathological parameters. A $P < 0.05$ was considered statistically significant.

RESULTS

The present study analyzed 32 skin biopsy samples, comprising 20 cases of psoriasis and 12 cases of NPPD. The psoriasis cases were diagnosed based on concordant clinical and histopathological features, while the NPPD group included cases with clinical suspicion of psoriasis that were histopathologically confirmed to be other conditions. All samples underwent evaluation using a standardized three-tier histological grading for key histological features, and Ki-67 immunohistochemistry was performed to assess cellular proliferation.

In the psoriasis group ($n = 20$), the most common clinical and histopathological subtype was chronic plaque psoriasis (75%, $n = 15$), followed by guttate psoriasis (15%, $n = 3$) and pustular psoriasis (10%, $n = 2$). In the NPPD group ($n = 12$), the most frequently encountered diagnosis was chronic dermatitis and allergic contact dermatitis (50%, $n = 6$), followed by lichen simplex chronicus (25%, $n = 3$), pityriasis rubra pilaris (16.7%, $n = 2$), and pityriasis lichenoides chronica (8.3%, $n = 1$).

Demographic analysis showed no statistically significant difference in age ($P = 0.9375$) or sex distribution ($P = 0.5011$) between the psoriasis and NPPD groups, indicating a comparable baseline demographic profile.

Histopathological examination revealed significant differences between the two groups. Confluent parakeratosis of Grade 3 was significantly more common in psoriasis cases ($P = 0.022$), whereas in NPPD, it was either absent or present in lower grades (Grade 1 or 2) and was typically focal. Regular acanthosis of Grade 3, characterized by uniform elongation of rete ridges across all HPF, was also significantly more frequent in psoriasis ($P = 0.0173$). Likewise, hypogranulosis or agranulosis was a consistent feature in psoriasis, predominantly observed as Grade 3, with a few cases showing Grade 2, and was significantly more common compared to NPPD ($P = 0.0095$). Minimal dermal inflammation was more frequently seen in psoriasis cases ($P = 0.0325$), whereas NPPD cases exhibited variable or more prominent inflammation depending on the specific diagnosis. In contrast, hyperkeratosis was seen in both groups and did not show a statistically significant difference ($P = 0.3372$), indicating that this feature alone is not helpful in distinguishing between the two conditions.

Ki-67 immunohistochemistry demonstrated notable differences in proliferative activity between the two groups. All 20 psoriasis cases showed positive Ki-67 staining, exhibiting >50-75% positivity in this study, reflecting high epidermal turnover. Among the NPPD group, 33% ($n = 4$) of the cases were negative for Ki-67, 50% ($n = 6$) showed positivity ≤50%, and only 16% ($n = 2$) exhibited >50% positivity, a difference that was statistically significant ($P = 0.0173$). However, the intensity of Ki-67 staining was not significantly different between the groups ($P = 0.5809$), suggesting that the extent of positivity rather than staining strength is diagnostically relevant [Table 3] for a comprehensive summary of the results.

DISCUSSION

The differentiation between psoriasis and NPPD remains a diagnostic challenge due to overlapping clinical and histopathological features such as hyperkeratosis, parakeratosis, agranulosis, regular acanthosis, and dermal inflammation. Features such as Munro's microabscess and suprapapillary thinning are hallmark features of psoriasis, however due to their inconsistent presence in all cases, they were excluded from our grading parameters to maintain objectivity and reproducibility. Our study employed a three-tier histological grading for overlapping histopathological features alongside Ki-67 immunohistochemistry to enhance diagnostic accuracy.

Histopathologically, psoriasis cases exhibited significant features such as confluent parakeratosis, regular acanthosis, and hypogranulosis/agranulosis, predominantly graded as 3. These findings align with previous studies that emphasize these features as histologic hallmarks of psoriasis.[15,16] In contrast, NPPD cases demonstrated focal or absent parakeratosis and irregular acanthosis, with variable grades, underscoring the heterogeneity within this group. Cesinaro *et al.*[12] and Tortola *et al.*[17] have documented similar differences between psoriasis and other psoriasiform disorders.[12,17]

The Ki-67 proliferation index further distinguished the two groups. All psoriasis cases showed Ki-67 positivity, with 60% of the cases exhibiting >60% positivity, indicating heightened epidermal proliferation. Conversely, 33% of NPPD cases were Ki-67 negative, and a few cases had ≤50% positivity, and only 16% of cases showed >50% positivity. This statistically significant difference ($P = 0.0173$) corroborates findings from several earlier studies that reported markedly elevated Ki-67 expression in psoriasis compared to other dermatoses.[15,16,18,19]

In our study, for example, one case of psoriasis vulgaris exhibited parakeratosis (Grade 3), agranulosis (Grade 3), regular acanthosis (Grade 2), and moderate dermal inflammation (Grade 2). Ki-67 immunohistochemistry revealed 60% positivity [Figure 1a-c].

These findings are consistent with the known pathophysiology of psoriasis, which is characterized by rapid epidermal turnover and increased mitotic activity of basal keratinocytes.[15,20] In contrast, many forms of NPPD, such as chronic spongiotic dermatitis and lichen simplex chronicus, exhibit variable or lower proliferative indices.[21]. Ki-67, a nuclear protein expressed in all active phases of the cell cycle except G0, is a widely accepted marker of cellular proliferation and has been used as a differentiator in many dermatopathological studies.[15,20]

Interestingly, in our study, a few NPPD cases with >50% Ki-67 positivity corresponded to lower histological grades (1-2) [Figure 2a-b], and another case of NPPD in our study

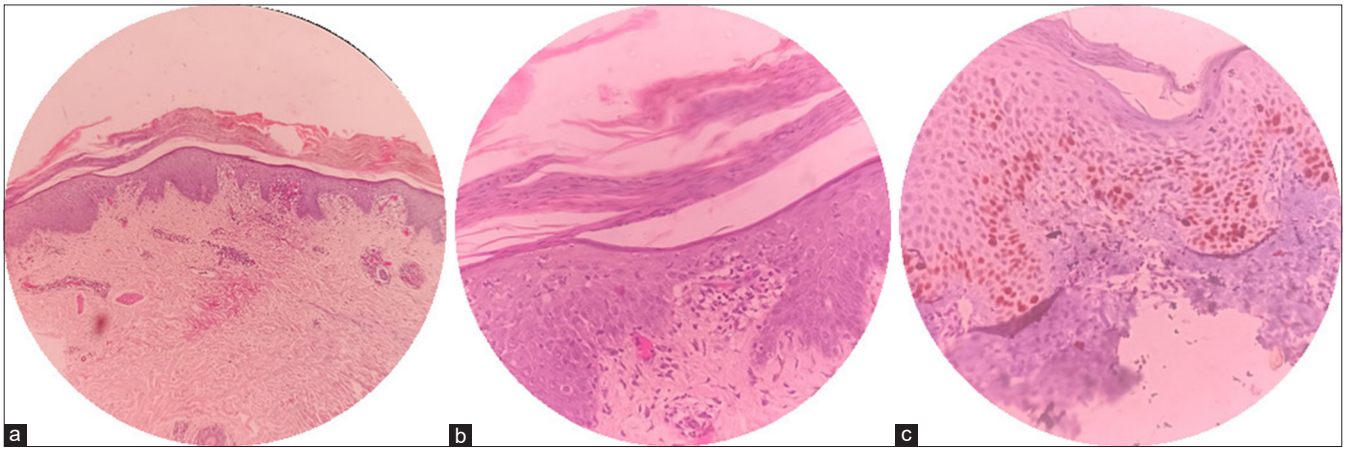

Figure 1: (a) Hematoxylin and Eosin (H&E) at 10x, Low Power Field (LPF) - A case of psoriasis vulgaris. (b) H&E at 40x high-power field (HPF) -in psoriasis vulgaris showing parakeratosis (Grade 3), agranulosis (Grade 3), regular acanthosis (Grade 2), and moderate dermal inflammation (Grade 2) (c) Immunohistochemical at 40x, HPF, in psoriasis vulgaris Ki-67 showing 60% nuclear positivity.

Table 3: Comparison between psoriasis and NPPD cases.

Parameter	Psoriasis (*n*=20)	NPPD (*n*=12)	*P*-value	Remarks
Most common subtype	Chronic plaque psoriasis (75%)	Chronic dermatitis and allergic contact dermatitis (50%)	-	-
Age distribution	No significant difference	No significant difference	0.9375	-
Gender distribution	No significant difference	No significant difference	0.5011	-
Confluent parakeratosis (Grade 3)	Common	Focal or lower grade	0.022	Significant
Regular acanthosis (Grade 3)	Frequent	Less frequent	0.0173	Significant
Hypogranulosis/agranulosis	Predominantly Grade 3	Less frequent	0.0095	Significant
Dermal inflammation	Minimal	Variable/prominent	0.0325	Significant
Hyperkeratosis	Seen	Seen	0.3372	Not significant
Ki-67 positivity	>50–75% in 100% cases	33% of cases negative, 16% of cases >50%	0.0173	Significant

NPPD: Non-psoriatic psoriasiform dermatoses, *P*-values were calculated using the Chi-square test (or Fisher's Exact Test when expected cell counts were <5). For Ki-67 positivity, the Mann–Whitney U test was applied. A *P*-value <0.05 was considered statistically significant.

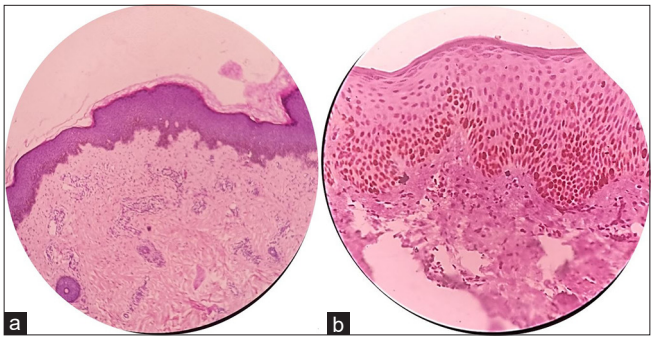

Figure 2: (a) Hematoxylin and eosin (H&E) at 10x, lpf- a case of NPPD showing Grade 1 parakeratosis, agranulosis, irregular acanthosis, hyperkeratosis, and Grade 2 perivascular inflammation. (b) IHC at 40x, HPF Ki-67 - a case of non-psoriatic psoriasiform dermatoses showing 50% nuclear positivity.

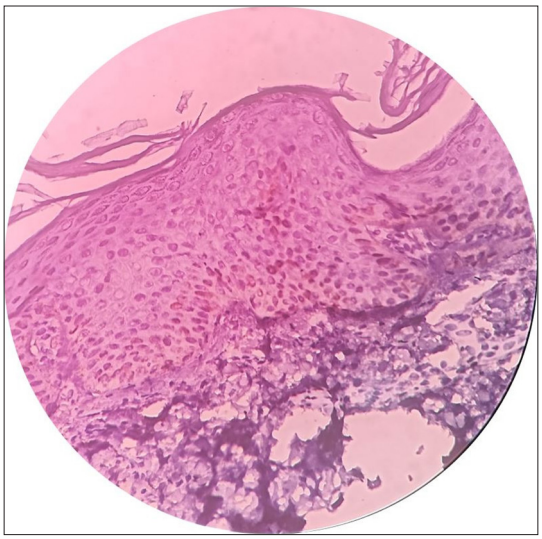

Figure 3: Immunohistochemical, high-power field Ki-67 - another case of non-psoriatic psoriasiform dermatoses showing 20% nuclear positivity at 40x, high-power field.

showing Ki-67 positivity of 20% [Figure 3], suggesting that Ki-67 alone may not suffice for definitive diagnosis. Similar findings were observed by Sezer *et al*.[15] and Abdelsalam *et al*.,[7] who noted overlapping Ki-67 indices in some non-psoriatic dermatoses.[7,15,16] This observation emphasizes the necessity of integrating both histopathological grading and Ki-67 indices for accurate differentiation. Mizrak *et al*.[9] and Kim *et al*.[5] also advocated for the combined use of multiple histological and IHC parameters to improve diagnostic reliability.

In addition to Ki-67, previous research has also explored adjunctive markers such as Cyclin D1, p53, Bcl-2, GLUT-1, and CD34 to aid in distinguishing psoriasis from its mimics.[15,16,19,21] While our study focused on Ki-67, incorporating such markers in future investigations may further improve specificity.

Our demographic analysis revealed no significant differences in age or gender distribution between the two groups, consistent with existing literature that suggests these demographic factors do not markedly affect histopathological patterns or Ki-67 expression.[16,18]

A few NPPD cases with >50% Ki-67 positivity corresponded to lower histological grades (1-2), suggesting that Ki-67 alone may not be sufficient for definitive diagnosis. This overlap underscores the limitations of relying solely on proliferation indices and highlights the necessity of integrating morphological assessment. Our study shows that a Ki-67 positivity threshold of >60%, when interpreted alongside structured histopathological grading, provides greater diagnostic reliability in distinguishing psoriasis from NPPD.

In particular, the combination of specific histological features, such as confluent parakeratosis, regular acanthosis, hypogranulosis, and minimal dermal inflammation, with Ki-67 expression exceeding 60% forms a robust diagnostic framework. While overlaps may still occur, especially in early or mild NPPD cases, the application of a standardized three-tier histological grading in conjunction with Ki-67 immunolabeling significantly enhances diagnostic precision.

Our findings reinforce the diagnostic value of integrating detailed histopathological criteria with Ki-67 immunostaining. This dual approach proves especially valuable in differentiating psoriasis from psoriasiform mimickers, where clinical and microscopic features may converge. The use of a structured grading system in our study ensures objectivity and reproducibility, while Ki-67 with a cutoff range of more than >60% provides an added layer of diagnostic support. Future research involving larger sample sizes and additional biomarkers such as cyclin D1 and GLUT-1 may further refine diagnostic algorithms and aid in optimizing patient management.[15,16,21]

CONCLUSIONS

This study suggests that combining a structured three-tier histological grading with Ki-67 IHC positivity 60% or above may aid in improving the differentiation of psoriasis from NPPD. While Ki-67 supports the diagnosis of psoriasis, it cannot serve as a standalone marker for psoriasis. Integrating it with our structured histopathological grading improves diagnostic accuracy and reliability.

Author contribution: KSES: Concept and design of the study, acquisition of data, or analysis and interpretation of data, drafting the article or revising it critically for important intellectual content; VS: Drafting the article or revising it critically for important intellectual content, final approval of the version to be published; SS: Aptitude to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of of the

work are appropriately investigated and resolved; SK: Aptitude to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of of the work are appropriately investigated and resolved.

Ethical approval: The research/study was approved by the Institutional Review Board at MGMCRI, approval number MGMCRI/FP/01/2024/13, dated 28th April 2024.
Declaration of patient consent: Patient's consent not required as there are no patients in this study.
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Use of artificial intelligence (AI)-assisted technology for manuscript preparation: The authors confirm that there was no use of artificial intelligence (AI)-assisted technology for assisting in the writing or editing of the manuscript, and no images were manipulated using AI.

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