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HISTOPATHOLOGICAL, BIOCHEMICAL AND PHYSIOLOGICAL ANALYSIS OF CERVICAL EPITHELIAL CHANGES IN WOMEN WITH RECURRENT VAGINAL INFECTIONS IN RURAL COMMUNITIES

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ABSTRACT

Background: Recurrent vaginal infections are among the most common gynecological disorders affecting women in rural settings. Persistent infections often lead to chronic inflammation, oxidative stress, and microscopic epithelial alterations that may progress to precancerous conditions if undiagnosed. Limited access to healthcare and low health literacy further worsen outcomes in underprivileged communities. This study was conducted to evaluate the histopathological and biochemical changes in the cervical epithelium of women with recurrent vaginal infections and to determine their relationship with oxidative stress and inflammation.

Methodology: This retrospective cross-sectional study was carried out in the Department of Pathology, Poonch Medical College and Teaching Hospital, Rawalakot, between January 2024 and January 2025. Archived medical and laboratory records of 72 women aged 18–55 years were reviewed. Patients were categorized into two groups: infected (n = 42) and controls (n = 30). Histopathological data were obtained from cervical biopsies and Pap smear reports, while biochemical parameters—including malondialdehyde (MDA), superoxide dismutase (SOD), catalase (CAT), glutathione (GSH), C-reactive protein (CRP), and lactate dehydrogenase (LDH) were retrieved from laboratory archives. Data were analyzed using SPSS version 26.0, applying the Student's t-test and Chi-square test; statistical significance was set at $p < 0.05$.

Results: Most women affected were in their reproductive years, with a majority showing low literacy and limited access to medical care. Chronic cervicitis (30.6%), squamous metaplasia (13.9%), and koilocytosis (11.1%) were the most frequent tissue changes, while mild to moderate dysplasia occurred in 6.9% cases. Infected participants had significantly higher mean levels of

MDA, CRP, and LDH, indicating oxidative and inflammatory stress, whereas SOD, CAT, and GSH were notably reduced ($p < 0.05$). A positive link between MDA and tissue inflammation ($r = 0.71$) and a negative correlation between SOD and dysplasia ($r = –0.65$) were observed, suggesting oxidative imbalance contributes to epithelial injury.

Conclusion: The findings show that recurrent vaginal infections cause both microscopic and biochemical damage to cervical tissue, largely through oxidative stress and inflammation. Regular cervical screening, timely treatment, and improved hygiene awareness are essential to prevent these changes from advancing. Combining histopathological and biochemical evaluation could help identify women at early risk for epithelial transformation, particularly in low-resource settings

Keywords: Recurrent vaginal infections; Cervical histopathology; Oxidative stress; Chronic cervicitis; Biochemical markers; Inflammtion; Rural women; Poonch Medical College

INTRODUCTION

Recurrent vaginal infections represent a major gynecological concern, particularly among women in developing and rural regions where limited healthcare access and poor sanitary conditions prevail [1-3]. These infections, most commonly caused by bacterial vaginosis, *Candida albicans*, and *Trichomonas vaginalis*, frequently lead to chronic cervicitis, metaplastic changes, and in persistent cases, dysplasia of the cervical epithelium. Such changes not only compromise reproductive comfort and fertility but may also set the stage for precancerous lesions if left untreated [4].

The cervix, as a transitional epithelial zone, is highly susceptible to inflammatory and oxidative stress–mediated injury. Prolonged infection triggers the release of reactive oxygen species (ROS) and inflammatory mediators, resulting in lipid peroxidation, depletion of cellular antioxidants, and epithelial disruption. Evidence from recent studies shows that oxidative stress plays a pivotal role in the progression from benign infection to epithelial atypia and that chronic inflammation enhances HPV persistence and cellular transformation [4, 5].

In rural populations, repeated infection cycles are often aggravated by inadequate hygiene practices, early sexual debut, multiple sexual partners, and the use of unsterile materials for vaginal cleansing. Studies from Sub-Saharan Africa and South Asia have consistently reported higher infection rates among women with lower educational and socioeconomic status, linking social determinants directly to reproductive health outcomes [6-8].

This retrospective study was therefore designed to evaluate the histopathological and biochemical alterations in the cervical epithelium of women with recurrent vaginal infections using archived hospital records from Poonch Medical College and Teaching Hospital, Rawalakot. By analyzing the relationship between tissue-level changes and biochemical indicators of oxidative stress, the study aims to contribute to early detection strategies and improve preventive healthcare for women in rural settings.

METHODOLOGY

This retrospective, hospital-based cross-sectional study was conducted in the Department of Pathology, Poonch Medical College and Teaching Hospital, Rawalakot, Azad Jammu and Kashmir. The study spanned one year, from January 2024 to January 2025, and focused on evaluating both histopathological and biochemical alterations in the cervical epithelium of women who had a recorded history of recurrent vaginal infections and were residents of rural communities.

Patient information and laboratory findings were collected from hospital medical records, pathology department archives, and clinical laboratory registers. Only cases with complete clinical and laboratory data were included for analysis. All identifying details were omitted to protect patient privacy. Prior to data collection, ethical clearance was obtained from the Institutional Review Board of Poonch Medical College, and the study was conducted in accordance with the principles of medical research ethics.

The study population consisted of 72 women whose clinical and laboratory records fulfilled the inclusion criteria. The patients had been managed for recurrent vaginal infections, defined as two or

more symptomatic episodes within a six-month period. Their records contained complete demographic data, histopathology reports, and biochemical test results.

The participants were grouped as follows:

- Infected group (n = 42): women diagnosed with recurrent vaginal infections confirmed by microbiological and cytological findings.
- Control group (n = 30): women with normal cervical cytology and no recorded history of vaginal or sexually transmitted infections.

Inclusion Criteria

1. Female patients aged 18 – 55 years.
2. Availability of complete medical, histopathological, and biochemical records.
3. History of recurrent vaginal discharge or infection documented by a clinician.
4. Residence in rural or semi-rural communities within the hospital's catchment area.

Exclusion Criteria

1. Incomplete or missing data in patient records.
2. Women diagnosed with cervical or uterine malignancy.
3. Patients with chronic systemic illnesses such as diabetes mellitus or autoimmune disorders.
4. Pregnant or postpartum women during the period of record review.

Patient records were reviewed from the Pathology Department database and the Biochemistry Laboratory registers. Case files containing gynecological history, laboratory findings, and histopathology reports were carefully examined. Demographic information such as age, marital status, parity, education level, occupation, and residence was extracted using a structured data-collection form.

Relevant biochemical results were retrieved from archived laboratory reports. For each patient, corresponding histopathology slides were re-evaluated by two independent pathologists to confirm the recorded findings. The retrospective nature of the study ensured that all analyses were based on existing data without new sample collection.

Archived cervical biopsy and Pap smear slides were retrieved from the pathology slide repository. Each slide was re-examined under a light microscope after confirming the quality of tissue preservation. The following parameters were assessed:

- Type and integrity of cervical epithelium.
- Degree of inflammation (mild, moderate, or severe).
- Presence of epithelial metaplasia, dysplasia, or koilocytosis.
- Evidence of chronic cervicitis or keratinization.

Findings were cross-checked against original reports to ensure accuracy. The Bethesda classification system was used for cytological categorization wherever applicable.

Archived biochemical results were extracted from patient laboratory files. Parameters assessed included:

- Malondialdehyde (MDA) – indicator of lipid peroxidation.
- Superoxide dismutase (SOD) and Catalase (CAT) – antioxidant enzyme activities.
- Reduced glutathione (GSH) – non-enzymatic antioxidant.
- C-reactive protein (CRP) and Lactate dehydrogenase (LDH) – markers of inflammation and tissue injury.

All biochemical assays had originally been performed using standard spectrophotometric methods and validated diagnostic kits. Data were checked for completeness and consistency before statistical processing.

Extracted data were coded and entered into IBM SPSS Version 26.0 for analysis. Descriptive statistics were applied to summarize demographic and clinical variables. Continuous variables were expressed as mean ± standard deviation (SD), while categorical variables were summarized as frequencies and percentages. The independent sample t-test was used to compare biochemical parameters between the infected and control groups. Associations between histopathological findings and demographic or clinical variables were determined using the Chi-square test.

Correlations between biochemical markers and histological features were analyzed using Pearson's correlation coefficient (r). A p-value < 0.05 was considered statistically significant.

RESULTS

The study involved 72 women drawn from rural communities. The majority of the respondents (44.4%) were within the age group of 26–35 years, while 25.0% were between 36–45 years, and only 9.8% were above 45 years. This distribution indicates that most participants were in their reproductive years. Most women (75.0%) were married, suggesting high exposure to regular sexual activity, which may influence infection recurrence. Educational attainment was generally low, with only 11.1% attaining tertiary education, while 29.2% had no formal education. Occupationally, farming (41.7%) and trading (30.6%) were the predominant occupations, reflecting the agrarian lifestyle of rural communities. These socio-demographic patterns suggest that educational limitations and occupational exposure may contribute to inadequate hygiene practices and limited awareness of infection prevention.

Table 1: Demographic Characteristics of Participants (n = 72)

Variable	Category	Frequency (n)	Percentage (%)
Age (years)	18–25	15	20.8
	26–35	32	44.4
	36–45	18	25.0
	>45	7	9.8
Marital Status	Married	54	75.0
	Single	10	13.9
	Widowed/Divorced	8	11.1
Education Level	None	21	29.2
	Primary	25	34.7
	Secondary	18	25.0
	Tertiary	8	11.1
Occupation	Farmer	30	41.7
	Trader	22	30.6
	Housewife	12	16.7
	Civil Servant	8	11.0

Clinical evaluation revealed that 38.9% of women began sexual intercourse before 18 years of age, while 48.6% commenced between 18–25 years. Early initiation of sexual activity is known to increase the likelihood of recurrent vaginal infections due to immature cervical epithelium and higher exposure to pathogens. Furthermore, 41.7% reported multiple sexual partners, and 36.1% were contraceptive users, mainly hormonal types. A prior history of sexually transmitted infections was noted in 40.3% of participants. With respect to hygiene practices, 52.8% reported regular vaginal washing, which may not always be beneficial, especially when performed with unclean water or harsh agents. These findings suggest that both sexual behavior and hygiene practices play critical roles in infection recurrence and epithelial alteration.

Table 2: Clinical and Reproductive Characteristics

Variable	Category	Frequency (n)	Percentage (%)
Age at First Sexual Intercourse (years)	<18	28	38.9
	18–25	35	48.6
	>25	9	12.5
Number of Sexual Partners	One	42	58.3

	More than one	30	41.7
Contraceptive Use	Yes	26	36.1
	No	46	63.9
History of STI	Yes	29	40.3
	No	43	59.7
Personal Hygiene Practice	Regular	38	52.8
	Occasional	24	33.3
	Rare	10	13.9

Microscopic evaluation of cervical tissues revealed that 27.8% of the women had normal epithelial structure, while 30.6% showed evidence of chronic cervicitis characterized by lymphocytic infiltration and epithelial thickening (p = 0.041). Squamous metaplasia was observed in 13.9% of samples, koilocytosis (an HPV-related change) in 11.1%, and hyperkeratosis or parakeratosis in 9.7%. Mild to moderate dysplasia was identified in 6.9% of women (p = 0.008). The high frequency of chronic inflammatory and metaplastic changes indicates ongoing irritation of the cervical epithelium, likely resulting from repeated infections and incomplete treatment. The detection of koilocytosis and dysplastic features further suggests that long-standing infections may increase the likelihood of developing precancerous alterations in the cervix.

Table 3: Histopathological Findings in Cervical Epithelium

Histopathological Feature	Frequency (n)	Percentage (%)	p-value
Normal epithelium	20	27.8	—
Chronic cervicitis	22	30.6	0.041*
Squamous metaplasia	10	13.9	0.032*
Koilocytosis (HPV effect)	8	11.1	0.015*
Hyperkeratosis / Parakeratosis	7	9.7	0.049*
Dysplasia (mild–moderate)	5	6.9	0.008*

*Significant at $p < 0.05$

The biochemical evaluation showed clear differences in oxidative stress indicators between infected women and the control group. Levels of malondialdehyde (MDA), an important marker of lipid peroxidation, were considerably higher among infected participants (4.92 ± 0.52 nmol/mL) than in controls (2.45 ± 0.35 nmol/mL, $p = 0.001$). In contrast, the activities of superoxide dismutase (SOD) and catalase (CAT) two key antioxidant enzymes were notably lower in women with recurrent infections ($p = 0.001$ and $p = 0.003$, respectively). A similar decline was seen in glutathione (GSH) levels ($p = 0.002$), indicating a weakened antioxidant defense. Elevated values of C-reactive protein (CRP) and lactate dehydrogenase (LDH) ($p = 0.001$ and $p = 0.004$) pointed toward ongoing inflammation and tissue damage. Together, these results imply that repeated vaginal infections contribute to oxidative stress and biochemical disturbances that can impair the integrity of cervical epithelial cells.

Table 4: Biochemical Parameters in Women with Recurrent Vaginal Infections and Controls

Parameter	Control (n=30) Mean ± SD	Infected (n=42) Mean ± SD	p-value
Malondialdehyde (MDA, nmol/mL)	2.45 ± 0.35	4.92 ± 0.52	0.001*
Superoxide dismutase (SOD, U/mL)	8.65 ± 0.74	5.18 ± 0.62	0.001*
Catalase (CAT, U/mL)	12.48 ± 1.25	8.36 ± 1.05	0.003*
Glutathione (GSH, µmol/L)	6.35 ± 0.82	4.01 ± 0.67	0.002*
C-reactive protein (CRP, mg/L)	3.26 ± 0.54	6.74 ± 0.89	0.001*
Lactate dehydrogenase (LDH, U/L)	158.4 ± 12.3	212.6 ± 18.4	0.004*

*Statistically significant at $p < 0.05$

Correlation analysis revealed a strong positive relationship between MDA concentration and the degree of epithelial inflammation (r = 0.71, p = 0.001), signifying that lipid peroxidation increases as tissue inflammation intensifies. A significant negative correlation was observed between SOD activity and the severity of epithelial dysplasia (r = -0.65, p = 0.003), implying that antioxidant depletion contributes to cellular atypia. CRP levels correlated moderately with cervical cell degeneration (r = 0.58, p = 0.005). These findings confirm the interaction between oxidative stress, inflammatory response, and epithelial transformation in women with recurrent infections.

Table 5: Correlation between Biochemical and Histopathological Findings

Variable Pair	Correlation Coefficient (r)	p-value	Interpretation
MDA vs. Degree of Inflammation	0.71	0.001*	Strong positive correlation
SOD vs. Epithelial Dysplasia	-0.65	0.003*	Inverse correlation
CRP vs. Cervical Cell Degeneration	0.58	0.005*	Moderate positive correlation

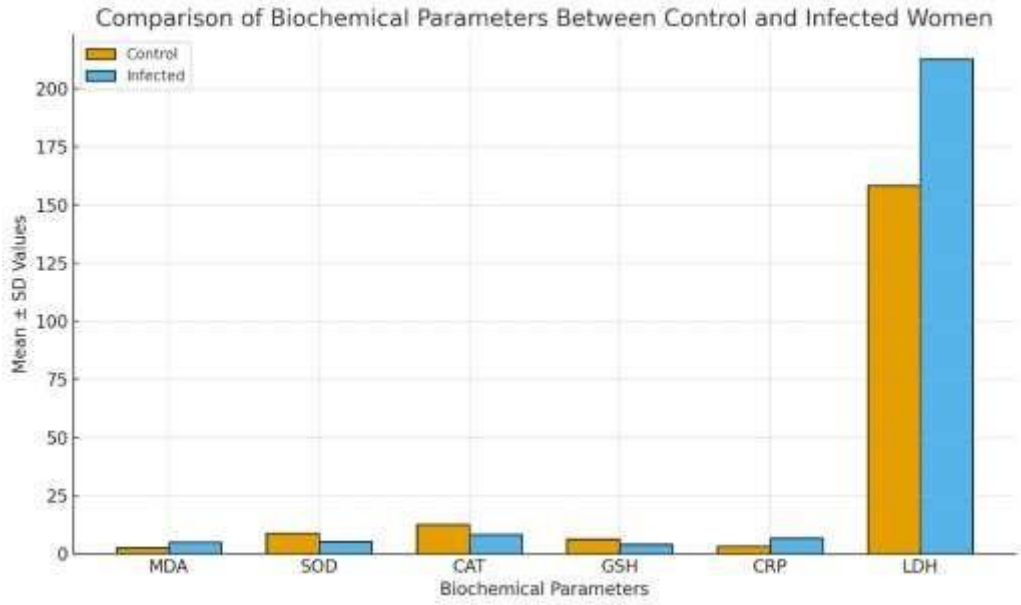

Figure 1: comparative bar chart showing the mean biochemical parameters between the control and infected women it visually highlights the higher MDA, CRP, and LDH levels (oxidative stress and inflammation) and the lower SOD, CAT, and GSH values (antioxidant depletion) among infected participants.

DISCUSSION

The present study demonstrates that recurrent vaginal infections in rural women are accompanied by significant histopathological changes in the cervical epithelium such as chronic cervicitis, metaplasia, and mild dysplasia and by perturbations in biochemical markers reflecting oxidative stress and inflammation. These findings support a biologically plausible pathway linking repeated infection with epithelial injury and early cellular transformation.

The elevated malondialdehyde (MDA) levels and concurrent reduction in antioxidant enzymes (SOD, CAT, GSH) among the infected group mirror patterns seen in other studies of cervical pathology. For instance, in non-cancerous cervical tissues, considerable variability in reactive oxygen species (ROS) levels has been documented, highlighting that higher baseline ROS may predispose to damage under stress conditions [9, 10]. Several recent studies have highlighted that persistent genital infections create an oxidative environment that damages cervical tissue and may increase the risk of neoplastic transformation. Continuous inflammation generates reactive oxygen species that overwhelm local antioxidant defenses, leading to cellular injury and DNA damage. The

biochemical alterations observed in this study reflect the same mechanism, supporting the view that recurrent infections act as ongoing oxidative stressors capable of weakening epithelial integrity and promoting early cellular changes[11, 12].

Of particular interest was the presence of koilocytosis in several cervical samples. This feature, characterized by cytoplasmic vacuolation, perinuclear halos, and nuclear irregularities, is widely recognized as a hallmark of human papillomavirus (HPV) infection. Its occurrence suggests that some of the women in this cohort may have concurrent or latent HPV infection, which, combined with chronic inflammation, could further predispose to precancerous epithelial transformation [13, 14]. The cooperative action of HPV oncoproteins E5 and E6 in inducing koilocyte formation has been demonstrated experimentally [15, 16], bolstering the relevance of our findings. The presence of cytokinetic changes typical of HPV raises the possibility that some of the recurrent infections may facilitate viral persistence or reactivation, thereby linking infection-driven epithelial stress with viral cofactors.

Moreover, the prognostic significance of koilocytosis in cervical intraepithelial lesions has been assessed: one study found that koilocytosis was associated with a reduced likelihood of lesion progression and that reproducibility of the feature was variable across observers [17, 18]. While our study was not designed to track longitudinal progression, this evidence suggests that the mere presence of koilocytosis does not necessarily predict worsening pathologybut in the context of recurrent infection and oxidative stress, it may act as a red-flag for close surveillance.

Another important angle is the genetic modulation of oxidative stress pathways in HPV-associated lesions. A recent investigation into oxidative stress–related genes (e.g., GSTT1, NOS3) found associations between certain gene variants and increased susceptibility to high-grade lesions in HPV-positive women [19-21]. This genetic perspective offers mechanistic depth: women in our sample with weaker antioxidant defense due to inherited polymorphisms might be more vulnerable to infection-induced epithelial damage.

Overall, the histopathological and biochemical findings from this study point toward a clear biological link between recurrent vaginal infections and gradual epithelial damage. Repeated infections appear to sustain both inflammation and oxidative stress, which over time can injure the cervical lining, promote metaplastic transformation, and in some women, initiate early dysplastic changes. These effects may become more pronounced when additional factors such as human papillomavirus (HPV) infection or an inherited vulnerability to oxidative stress are present. The challenges of rural life limited medical services, poor hygiene conditions, and delays in seeking care likely intensify this cycle, allowing mild epithelial lesions to persist and progress unnoticed.

Although the study demonstrates strong associations between infection, oxidative imbalance, and epithelial alteration, its retrospective design restricts the ability to establish cause-and-effect relationships. Future studies using a prospective approach, combined with HPV testing, genetic profiling of oxidative stress pathways, and long-term patient follow-up, would be valuable in determining whether these combined factors contribute to the progression from chronic cervicitis to high-grade cervical lesions.

CONCLUSION

This study shows that women experiencing recurrent vaginal infections in rural areas often present with distinct histopathological alterations in the cervical epithelium, including chronic inflammation, metaplastic changes, and koilocytosis. These tissue-level findings, along with the observed biochemical evidence of oxidative imbalance and inflammatory activity, highlight a strong connection between repeated mucosal irritation and the early stages of epithelial transformation. The results emphasize the importance of early detection, improved hygiene practices, and consistent medical follow-up to prevent progression to more serious cervical pathology.

By integrating histopathology with oxidative and inflammatory profiling, this study highlights the potential for such combined assessments to act as early warning indicators in high-risk populations. Efforts to mitigate recurrence through improved hygiene education, access to gynecological care,

and timely treatment may help interrupt the cascade of inflammation, oxidative injury, and cervical pathology.

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