



EVALUATION OF CONVENTIONAL PAPANICOLAOU SMEAR AND MANUAL LIQUID-BASED CYTOLOGY FOR DETECTION OF CERVICAL EPITHELIAL ABNORMALITIES

Dr. Vartika Singh

Dr. Shivali Singh

Dr. Aparajita Goel

Dr. Meenal Jain

Dr. Deepa Rani

ABSTRACT **Background:** Cervical cancer is largely preventable when screening and precancerous lesions are detected early. Conventional Papanicolaou (Pap) smear remains widely used, whereas liquid-based cytology (LBC) has been developed to improve specimen preparation and adequacy. **Aim:** To assess specimen adequacy and cytomorphological findings in conventional Pap smear and manual liquid-based cytology (LBC) among women undergoing cervical cytological evaluation. **Methods:** This cross-sectional study was conducted at S. N. Medical College, Agra, using 200 cervical cytology records. Conventional Pap smear findings were available for all 200 cases, while manual LBC findings were available for 41 cases in the master chart. Cytological abnormalities were classified according to the 2014 Bethesda System. Data were analysed using IBM SPSS Statistics version 26.0. Diagnostic-performance measures were not calculated because a complete reference-standard dataset was not available in the master chart. **Results:** Of the 200 conventional Pap smears, 196 (98.0%) were satisfactory and 4 (2.0%) were unsatisfactory. All 41 recorded manual LBC preparations were satisfactory. ASC-US was the most frequent abnormality with both conventional Pap smear (129/196, 65.8%) and manual LBC (24/41, 58.5%). Among satisfactory conventional Pap smears, 169 (86.2%) showed non-malignant epithelial abnormalities and 27 (13.8%) showed malignant diagnoses. Among the manual LBC records, 31 (75.6%) showed non-malignant epithelial abnormalities and 10 (24.4%) showed malignant diagnoses. Endocervical cells were documented in 183 (91.5%) conventional Pap smears. Among the 41 LBC records, they were documented as seen in 31 (75.6%), not seen in 5 (12.2%), and not documented in 5 (12.2%). **Conclusion:** Both conventional Pap smear and manual LBC produced satisfactory cytological preparations in the recorded cases and demonstrated a range of cervical epithelial abnormalities, including malignant lesions. However, manual LBC was available for only 41 cases and a complete reference-standard dataset was unavailable. Therefore, the available master-chart data do not allow a valid comparison of diagnostic accuracy or establish superiority of either technique.

KEYWORDS : Cervical Cancer; Cervical Cytology; Liquid-Based Cytology; Manual Liquid-Based Cytology; Papanicolaou Smear; Screening.

INTRODUCTION

Cervical cancer continues to be an important global public health concern, although most cases are potentially preventable through human papillomavirus (HPV) vaccination, screening, early detection and treatment of precancerous lesions. Persistent infection with high-risk HPV types is the established cause of almost all cervical cancers. In 2022, approximately 660,000 new cases and 350,000 deaths from cervical cancer were estimated worldwide, with the greatest burden occurring in low- and middle-income countries.^{1,2}

The continuing burden of cervical cancer in developing countries highlights the need for effective and accessible screening strategies. Because cervical cancer usually develops through a prolonged precancerous phase, epithelial abnormalities can be detected and treated before invasive disease develops. Conventional cytology and liquid-based cytology are both established approaches to cervical screening.³

Conventional Papanicolaou smear cytology has remained widely used because it is relatively inexpensive, technically simple and feasible in resource-limited settings. Its quality, however, can be affected by blood, mucus, inflammatory material, overlapping cells and uneven distribution of cellular material, all of which may make assessment more difficult.⁴⁻⁶

Liquid-based cytology was introduced to overcome some of these preparation-related problems by providing a more uniform cellular preparation, a cleaner background and more even distribution of cells.^{5,7} Comparative studies and systematic reviews have reported lower rates of unsatisfactory samples with LBC, although its advantage in diagnostic accuracy has not been consistently demonstrated.⁷⁻⁹

Manual LBC has received particular attention as a lower-cost alternative in settings where fully automated commercial systems may be difficult to implement. Studies from India have reported differences in specimen adequacy, smear quality and detection of cervical abnormalities when conventional cytology is compared with manual or other low-cost LBC techniques.¹¹⁻¹⁴

The present study was undertaken to describe conventional Pap smear and manual LBC findings in terms of specimen adequacy and cytological abnormalities in a tertiary-care setting. The analysis was based strictly on the available master-chart data; consequently, diagnostic-performance measures requiring a complete reference standard were not estimated.

AIM

To evaluate the adequacy and cytomorphological findings of conventional Papanicolaou smear and manual liquid-based cytology in cervical cytological evaluation.

OBJECTIVES

1. To describe and compare specimen adequacy and cytomorphological quality in cervical smears prepared by conventional Papanicolaou smear and manual liquid-based cytology.
2. To describe the distribution of cervical epithelial abnormalities and malignant lesions identified by conventional Papanicolaou smear and manual liquid-based cytology.
3. To describe the representation of endocervical cells in the available conventional Pap smear and manual LBC records.

MATERIALS AND METHODS

Study Design and Setting

A cross-sectional study was conducted at S. N. Medical College, Agra. Cervical cytology specimens received at the institution were processed and evaluated for epithelial abnormalities and malignant lesions.

Study Population

Women aged 35 years and above who had cervical cytology records available in the study dataset were included.

Inclusion Criteria

1. Females aged 35 years and above.
2. Women with cervical cytology records available in the study master chart.

Exclusion Criteria

1. Patients receiving radiotherapy or chemotherapy.
2. Vault smears from patients who had previously undergone surgery for carcinoma of the cervix.

Sample Size and Data Source

The master chart contained 200 conventional Pap smear records, while manual LBC findings were available for 41 cases. Only the recorded observations were used in the present analysis; no missing observations were imputed.

Sample Collection and Processing

For conventional Papanicolaou smear, cervical material was collected with a sterile wooden Ayre's spatula. The spatula was rotated through 360° over the ectocervical region, and the collected material was immediately spread onto a labelled glass slide and fixed in 95% ethyl alcohol. For manual LBC, cervical cells were collected using a Cervex brush. The brush head was detached and placed in a preservative-containing collection vial. The vial was shaken thoroughly, after which the specimen was transferred to a centrifuge tube containing 4 mL of cleaning solution and centrifuged at 3000 rpm for 10 minutes. The supernatant was discarded and the cellular pellet was retained. Approximately 500 µL of the resulting cellular suspension was pipetted onto a glass slide, spread and allowed to dry before staining.

Papanicolaou Staining and Cytological Evaluation

The prepared smears were stained by the Papanicolaou technique, using hematoxylin followed by OG-6 and EA counterstaining. The smears were then dehydrated through graded alcohol, cleared in xylene and mounted with DPX. Cytological findings were classified according to the 2014 Bethesda System for Reporting Cervical Cytology. The epithelial abnormalities assessed included ASC-US, ASC-H, LSIL, HSIL, squamous cell carcinoma (SCC) and adenocarcinoma.

Statistical Analysis

Data were entered and analysed using IBM SPSS Statistics version 26.0. Frequencies and percentages were used to summarise age, clinical presentation, specimen adequacy, endocervical-cell representation and cytological diagnoses. Sensitivity, specificity, predictive values and diagnostic accuracy were not calculated because a complete reference-standard dataset was not available in the master chart.

Ethical Considerations

The study was approved by the Institutional Ethics Committee, S. N. Medical College, Agra (Approval No. SNMC/IEC/DHR/2025/58). Written informed consent was obtained from participants before sample collection.

RESULTS

A total of 200 women were included in the study. Their ages ranged from 35 to 82 years. The largest proportion was in the 41–50-year age group (35.5%), followed by those aged 35–40 years (34.0%), 51–60 years (21.5%), 61–70 years (8.0%) and >70 years (1.0%).

Age group	Frequency	Percentage
35–40 years	68	34.0
41–50 years	71	35.5
51–60 years	43	21.5
61–70 years	16	8.0
>70 years	2	1.0
Total	200	100.0

White vaginal discharge was the most common presenting symptom (119; 59.5%), followed by post-coital bleeding (24; 12.0%), abdominal pain (21; 10.5%), dyspareunia (16; 8.0%), post-menstrual bleeding (7; 3.5%), dysuria (6; 3.0%), foul discharge (3; 1.5%), post-menopausal bleeding (2; 1.0%) and asymptomatic presentation (2; 1.0%).

Clinical presentation	Frequency	Percentage
White discharge	119	59.5
Post-coital bleeding	24	12.0
Abdominal pain	21	10.5
Dyspareunia	16	8.0
Post-menstrual bleeding	7	3.5
Dysuria	6	3.0
Foul discharge	3	1.5
Asymptomatic	2	1.0
Post-menopausal bleeding	2	1.0
Total	200	100.0

Adequacy of Conventional Pap Smears

Among the 200 conventional Pap smears, 196 (98.0%) were satisfactory for evaluation and 4 (2.0%) were unsatisfactory.

Endocervical cells were identified in 183 (91.5%) cases and were not identified in 17 (8.5%).

Parameter	Frequency	Percentage
Satisfactory	196	98.0
Unsatisfactory	4	2.0
Endocervical cells seen	183	91.5
Endocervical cells not seen	17	8.5

Manual Liquid-based Cytology

Manual LBC findings were available for 41 cases, all of which were satisfactory. Endocervical cells were documented as seen in 31 (75.6%) cases and not seen in 5 (12.2%). In the remaining 5 cases (12.2%), the endocervical-cell field was blank in the master chart; these were therefore recorded as not documented rather than not seen.

Parameter	Frequency	Percentage
Satisfactory	41	100.0
Unsatisfactory	0	0.0
Endocervical cells seen	31	75.6
Endocervical cells not seen	5	12.2
Endocervical cells not documented	5	12.2

Distribution of Cervical Epithelial Abnormalities and Malignant Lesions

Among the 196 satisfactory conventional Pap smears, 169 (86.2%) showed non-malignant epithelial abnormalities and 27 (13.8%) showed malignant lesions. Among the 41 manual LBC cases, 31 (75.6%) showed non-malignant epithelial abnormalities and 10 (24.4%) showed malignant lesions.

Diagnostic modality	Non-malignant epithelial abnormalities, n (%)	Malignant, n (%)	Total
Conventional Pap smear	169 (86.2)	27 (13.8)	196
Manual LBC	31 (75.6)	10 (24.4)	41

Cytological Spectrum of Lesions

On conventional Pap smear, ASC-US was the most frequently reported abnormality, with 129 cases (65.8%), followed by LSIL in 22 (11.2%), SCC in 20 (10.2%), HSIL in 10 (5.1%), ASC-H in 8 (4.1%) and adenocarcinoma in 7 (3.6%). Among the 41 manual LBC cases, ASC-US was reported in 24 (58.5%), SCC in 9 (22.0%), LSIL in 6 (14.6%), HSIL in 1 (2.4%) and adenocarcinoma in 1 (2.4%).

Diagnosis	Conventional Pap smear n (%)	Manual LBC n (%)
ASC-US	129 (65.8)	24 (58.5)
ASC-H	8 (4.1)	0 (0.0)
LSIL	22 (11.2)	6 (14.6)
HSIL	10 (5.1)	1 (2.4)
SCC	20 (10.2)	9 (22.0)
Adenocarcinoma	7 (3.6)	1 (2.4)
Total	196 (100.0)	41 (100.0)

DISCUSSION

In this study, conventional Pap smear findings were available for 200 women, whereas manual LBC findings were available for only 41 cases. The largest age group was 41–50 years (35.5%), followed by 35–40 years (34.0%). White vaginal discharge was the most common presenting symptom (59.5%), followed by post-coital bleeding (12.0%) and abdominal pain (10.5%). These symptoms are nonspecific and, by themselves, cannot establish the presence or severity of cervical epithelial abnormalities.

The conventional Pap smear had a satisfactory rate of 98.0%, while all 41 recorded manual LBC preparations were satisfactory. This LBC finding should be interpreted descriptively because LBC was available for only a small subset of the study records and was not documented for the entire conventional Pap smear cohort. Thus, the available data do not constitute a balanced paired comparison of specimen adequacy between the two techniques.

ASC-US was the most frequently reported cytological category in both modalities. It accounted for 129 cases (65.8%) among satisfactory conventional Pap smears and 24 cases (58.5%) among the 41 manual LBC records. The distribution of individual Bethesda categories may reflect differences in the clinical characteristics and referral pattern of the study population, as well as the smaller number of cases for which LBC was available.

Previous comparative studies have reported lower rates of unsatisfactory preparations with LBC, while systematic reviews have indicated that better specimen preparation does not necessarily result in a consistent improvement in the detection of high-grade cervical

lesions.⁷⁻⁹ Indian studies have likewise examined manual and low-cost LBC approaches as potentially useful options for improving specimen preparation in resource-constrained settings.¹¹⁻¹⁴ The findings of the present study are consistent with the observation that manual LBC can produce satisfactory preparations; however, the available master-chart data are insufficient to estimate comparative diagnostic accuracy.

A major methodological limitation of the present study is the absence of a complete reference-standard dataset in the master chart. Although columns related to histopathology were present, no HPE diagnoses were recorded in the available data sheet. Consequently, sensitivity, specificity, positive predictive value, negative predictive value and diagnostic accuracy were not calculated in this revised analysis. Reporting these measures without a verifiable 2×2 table could lead to an inaccurate representation of the study findings.

There was also a marked difference in the number of observations available for the two modalities: conventional Pap smear was recorded for 200 cases, whereas manual LBC was recorded for 41 cases. Therefore, differences in the proportions of epithelial abnormalities and malignant diagnoses between the two groups should not be interpreted as evidence that one technique is diagnostically superior.

Strengths and Limitations

Strengths: The study provides a description of conventional Pap smear and manual LBC findings from a tertiary-care setting, including specimen adequacy and the spectrum of cytological abnormalities using Bethesda terminology. Importantly, the revised analysis is limited to findings that can be directly verified from the available master chart.

Limitations: Manual LBC was recorded for only 41 of the 200 cases, resulting in unequal sample sizes and preventing a fully paired comparison of the two techniques. Five LBC records did not contain documented information on endocervical-cell representation. In addition, a complete histopathological/reference-standard dataset was unavailable, so diagnostic-performance measures and cyto-histopathological correlation could not be validly estimated. Finally, the study was conducted at a single institution, which limits the generalisability of the findings.

CONCLUSION

Both conventional Pap smear and manual LBC produced satisfactory preparations in the recorded cases and identified a range of cervical epithelial abnormalities, including malignant lesions. The satisfactory rate was 98.0% for the 200 conventional Pap smear records, while all 41 recorded manual LBC preparations were satisfactory. However, because manual LBC was available for only 41 cases and a complete reference-standard dataset was not available, these data do not establish comparative diagnostic accuracy or superiority of either technique. Larger, appropriately paired studies with systematic histopathological or colposcopic verification are needed to provide a reliable comparison of diagnostic performance.

REFERENCES

1. World Health Organization. Human papillomavirus and cancer. Geneva: World Health Organization; 2024.
2. World Health Organization. Cervical cancer. Geneva: World Health Organization; 2024.
3. IARC Working Group on the Evaluation of Cancer-Preventive Interventions. Cervical cancer screening. Lyon (FR): International Agency for Research on Cancer; 2022. PMID: 38507539.
4. Sreedevi A, Javed R, Dinesh A. Epidemiology of cervical cancer with special focus on India. *Int J Womens Health*. 2015;7:405-14. doi:10.2147/IJWH.S50001.
5. Honarvar Z, Zarisfi Z, Salari Sedigh S, Masoumi Shahrbabak M. Comparison of conventional and liquid-based Pap smear methods in the diagnosis of precancerous cervical lesions. *J Obstet Gynaecol*. 2022;42(6):2320-4. doi:10.1080/01443615.2022.2049721.
6. Kaban I, Bacanakgil BH, Koca S. The comparison of two methods in cervical smear screening - which method is better for smear adequacy rates? *Ginekol Pol*. 2021;92(5):335-8. doi:10.5603/GPa.2020.0185.
7. Arbyn M, Bergeron C, Klinkhamer P, Martin-Hirsch P, Siebers AG, Bulten J. Liquid compared with conventional cervical cytology: a systematic review and meta-analysis. *Obstet Gynecol*. 2008;111(1):167-77. doi:10.1097/01.AOG.0000296488.85807.b3.
8. Davey E, Barratt A, Irwig L, Chan SF, Macaskill P, Mannes P, et al. Effect of study design and quality on unsatisfactory rates, cytology classifications, and accuracy in liquid-based versus conventional cervical cytology: a systematic review. *Lancet*. 2006;367(9505):122-32. doi:10.1016/S0140-6736(06)67961-0.
9. Siebers AG, Klinkhamer PJ, Grefte JM, Massuger LF, Vedder JE, Beijers-Broos A, et al. Comparison of liquid-based cytology with conventional cytology for detection of cervical cancer precursors: a randomized controlled trial. *JAMA*. 2009;302(16):1757-64. doi:10.1001/jama.2009.1569.
10. Böhmer G, Stolte C, Vogelmann T, Schubert T. Liquid-Based Cytology in Cervical Cancer Screening: A Systematic Review and Meta-Analysis. *Acta Cytol*. 2026;1-11. doi:10.1159/000551747.
11. Nandini NM, Nandish SM, Pallavi P, Akshatha SK, Chandrashekhara AP, Anjali S, et al. Manual liquid based cytology in primary screening for cervical cancer—a cost effective proposition for scarce resource settings. *Asian Pac J Cancer Prev*. 2012;13(8):3645-51. doi:10.7314/APJCP.2012.13.8.3645.

12. Kavatkar AN, Nagwanshi CA, Dabak SM. Study of a manual method of liquid-based cervical cytology. *Indian J Pathol Microbiol*. 2008;51(2):190-4. doi:10.4103/0377-4929.41678.
13. Gupta R, Yadav R, Sharda A, Kumar D, Sandeep, Mehrotra R, et al. Comparative evaluation of conventional cytology and a low-cost liquid-based cytology technique, EziPREP™, for cervicovaginal smear reporting: a split sample study. *Cytojournal*. 2019;16:22. doi:10.4103/cytojournal.cytojournal_11_19.
14. Pankaj S, Nazneen S, Kumari S, Kumari A, Kumari A, Kumari J, et al. Comparison of conventional Pap smear and liquid-based cytology: a study of cervical cancer screening at a tertiary care center in Bihar. *Indian J Cancer*. 2018;55(1):80-3.
15. Bavikar R, Patel N, Buch A, Kulkarni M, Dharwadkar A, Viswanathan V. A comparison of conventional Pap smear and liquid-based cytology for cervical cancer screening. *Gynecol Minim Invasive Ther*. 2023;12(2):77-82. doi:10.4103/gmit.gmit_118_22.