



ORIGINAL RESEARCH PAPER

COMPARING THE EFFECTIVENESS OF LIQUID BASED CYTOLOGY (LBC) WITH CONVENTIONAL PAP SMEAR: A PROSPECTIVE STUDY

Pathology

KEY WORDS: CS-

Conventional Smear, LBC- Liquid Based Cytology, PAP- Papinicolaou

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ABSTRACT

Background: Cervical cancer is unique among human cancers which is mostly attributable to infection. Conventional PAP smear method is most effective for prevention and detection of cervical cancer but the accuracy of this method is low. This PAP smear now evolved to Liquid Based Cytology (LBC). **Method:** All the patients visiting Gynaecology OPD who fulfilled the inclusion Criteria in the duration starting from 1 January 2020 to 30 June 2021 in the Department of Pathology of Gajra raja Medical College, Gwalior (MP) were included in this study. 50 atypical smears by Conventional PAP test were then collected and these cases were subjected to Eziprep Liquid Based Cytology and Conventional PAP smear in private laboratory setup after taking history and clinical examination. The smears were studied by using 7 morphological parameters. **Result:** Smears were analysed for adequate cellularity, clean background, uniform distribution of cells, cellular overlapping, inflammation, distinct cell border, nuclear irregularity and then categorise by Bethesda reporting system. The results were significant only for clean background, uniform distribution of cells, cellular overlapping and inflammation. **Conclusion:** Results of cervical cytology smears by both methods showed that LBC provides more representative sample with reduced obscuring material, improved clarity allowing better morphological evaluation.

1. INTRODUCTION:

According to the Indian council of medical research (ICMR), the incidence of cervical carcinoma in India varies from 20-35/1,00,000 women between age group 35-64 year when compared with developed countries, it is as low as 1 to 8/1,00,000 women.¹

Cervical cancer is a devastating disease with an estimated 20 million cases by the year 2020.² The only gynecological cancer that satisfies the WHO criteria for implementation of screening program is the cervical cancer.³

In 1928 Dr. Papanicolaou first proposed cancer cells on a vaginal smear. It is a convenient, cost-effective and simple test. To overcome the limitation of conventional PAP smear, liquid-based cytology was introduced in 1990's as a much better tool for processing of cervical sample.⁴

LBC is the most approved method for detection of premalignant lesion and enhances the smear sensitivity. The basic principle is to collect cell sample into a liquid fixative solution and then create a monolayer of cells ready for microscopic observation after PAP's staining. The advantage of LBC includes removal of blood, mucus and obscuring cells; reduction of unsatisfactory smears; provision of cells for detection of HPV; presence of residual sample for performing ancillary techniques such as immunocytochemistry.⁵

Bethesda system is a system for reporting cervical or vaginal cytologic diagnosis, used for reporting PAP smear results. It was introduced in 1998 and revised in 1991, 2001 and 2014.

2. MATERIAL AND METHOD:

2.1 Inclusion Criteria:

All females from gynecology OPD visiting to Pathology Department for PAP smear evaluation, showing atypical cells in Conventional PAP Smear.

2.2 Exclusion Criteria:

- Normal PAP smear.

- Unsatisfactory PAP smear.
- Women with post hysterectomy.
- Women previously diagnosed or already treated as carcinoma.

50 atypical smears by Conventional PAP test were then collected and these cases were subjected to Eziprep Liquid Based Cytology and Conventional PAP smear in private laboratory setup after taking history and clinical examination. The smears were studied by using these morphological parameters-

1. Cellularity (adequate/inadequate).
2. Clean background (present/absent).
3. Uniform distribution (present/absent).
4. Cellular overlapping (present/absent).
5. Inflammation (present/absent).
6. Cell borders (distinct/indistinct).
7. Nuclear irregularity (present/absent).
8. Final interpretation (Based on The Bethesda System 2014)

3. RESULTS:

3.1 Age Wise Distribution Of Cases

02 cases (04%) studied belonged to third decade of life, 15 cases (30%) belonged to fourth decade of life, 19 cases (38%) belonged to fifth decade of life, 06 cases (12%) belonged to sixth decade of life, 05 cases (10%) belonged to seventh decade of life, 02 cases (04%) belonged to eighth decade of life and 01 case (02%) studied belonged to ninth decade of life.

3.2 Distribution Of Presenting Complaint

20 cases (40%) present with complaint of white discharge per vagina, 18 cases (36%) present with complaint of lower abdominal pain, 9 cases (18%) present with complaint of difficulty in micturition, 01 case (02%) presents with complaint of bleeding per vagina, 01 case (02%) presents with dysfunctional uterine bleeding, 01 case (02%) presents with complaint of postcoital bleeding.

3.3 STATISTICAL ANALYSIS OF CS & LBC RESULTS

50 cytology smears were compared by CS & LBC. The data

were analysed & interpreted by χ^2 (Chi square) test by the statistical software IBM SPSS statistics 20. The P values <0.05 were treated as statistically significant.

Table 1: Comparison of CS and PAP on morphological parameters

Morphological parameter	CS	LBC
Cellularity (Adequate)	43(86%)	47(94%)
Clean background (Present)	00(0%)	37(74%)
Uniform distribution (Present)	03(06%)	30(60%)
Cellular overlapping (Present)	47(94%)	20(40%)
Inflammation (Present)	47(94%)	35(70%)
Cell border (Distinct)	41(82%)	43(86%)
Nuclear irregularity (Present)	09(18%)	05(10%)

A. Comparison Of Cellularity (CS vs LBC)

Adequate cellularity was found in 47cases (94%) of LBC smears & 43 cases (86%) of CS. This result revealed that there was no significant difference between the two procedures ($P>0.05$ and χ^2 is 1.778).

B. Comparison Of Clean Background (CS vs LBC)

Clean background was seen in 37(74%) cases of LBC preparations but none of the CS showed clean background. This result revealed that there was statistically significant difference between the two procedures ($P<0.001$ and χ^2 is 58.73).

C. Comparison Of Uniform Distribution (CS vs LBC)

Uniform distributions of cells were found in 30 (60%) cases of LBC smears whereas it was observed in only 03 (06%) cases of CS. This result revealed that there was significant statistical difference between two methods ($P<0.001$ and χ^2 is 32.972).

D. Comparison Of Cellular Overlapping (CS vs LBC)

Cellular overlapping was seen in 20(40%) cases of LBC preparations and 47(94%) cases of CS. This result revealed that there was a significant statistical difference between two methods ($P<0.001$ and χ^2 is 32.972).

E. Comparison Of Inflammation (CS vs LBC).

Inflammation was seen in 35(70%) cases of LBC preparations and 47(94%) cases of CS. This result revealed that there was a significant statistical difference between two methods. ($P<0.05$ and χ^2 is 9.756).

F. Comparison Of Cell Border (CS vs LBC)

Cell border was distinct in 43(86%) cases of LBC preparations and 41(82%) cases of CS. This result revealed that there was no significant difference between the two procedures ($P>0.05$ and χ^2 is 0.298).

G. Comparison Of Nuclear Irregularity (CS vs LBC)

Nuclear irregularity was present in 05(10%) cases of LBC preparations and 09(18%) cases of CS. The results revealed that nuclear irregularity was present more in CS, which was statistically not significant ($P>0.05$ and χ^2 is 1.329).

Table 2: Comparison Of Bethesda Results (CS vs LBC).

INTERPRETATION	CS	LBC	X^2	DF	PVALUE
Unsatisfactory	07 (14%)	03(06%)			
ASCUS	05(10%)	01(02%)			
AGC NOS	01(02%)	01(02%)			
LSIL	22(44%)	28(56%)	6.303	6	$P>0.05$
HSIL	08(16%)	07(14%)			

SCC	07(14%)	09(18%)			
ENDOCERVIX ADENOCARCINOMA	0(0%)	01(02%)			
TOTAL	50	50			

The results revealed that unsatisfactory smears were reported in 14% and 6% cases of CS and LBC respectively. ASCUS was reported more in CS (10%) than LBC (02%). In CS 5 cases of ASCUS were reported, out of which 4 cases were reported as LSIL and only 1 case was reported as ASCUS in LBC. LSIL was reported more in LBC (56%) than CS (44%). A 68year old female with complaint of lower abdominal pain was reported as LSIL in CS and SCC in LBC. HSIL was reported more in CS (16%) than LBC (14%). A 35year old female with complaint of difficulty in micturition was reported as HSIL in CS and SCC in LBC. SCC was reported more in LBC (18%) than CS (14%). Endocervix Adenocarcinoma was reported in 1 case of LBC. No case was reported as Adenocarcinoma in CS. Although the result is not statistically significant ($P>0.05$).

Microscopy-

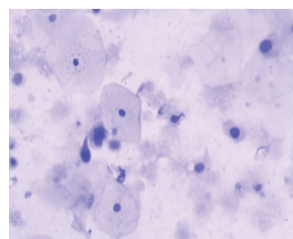


Fig.1. Pap Cellularity At 40x Magnification

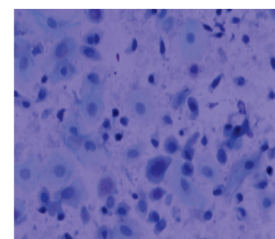


Fig.2. Lbc Cellularity At 40x Magnification

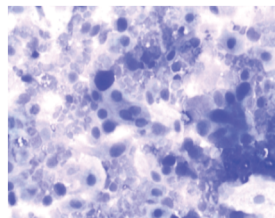


Fig.3. HSIL with Cellular Overlapping and Indistinct

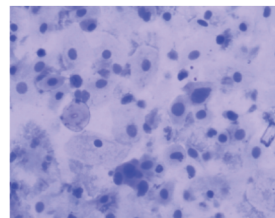


Fig.4. HSIL with Clear Background, Uniform Distribution of Cells and Distinct Cell Border in LBC at 40X magnification.

4. DISCUSSION:

This study compares a low-cost LBC technique with conventional smear preparation for cervical cytology in precancerous and cancerous lesions. The reduced rates of unsatisfactory samples, clarity of the background, and small area to be screened with resultant improved efficiency have led to a switch to LBC technique in most of the western countries.⁶

The study conducted by Shanmugapriya N et al⁷ showed that the mean age of the study group was 41.6 with range of 23 to 70 years. In our study the mean age was 48.1 with range of 28 to 85 years since we included atypical smears only.

The study conducted by Sunita Rai et. al⁸ showed that the most common presenting complaint of patients was of white discharge per vagina 42.74%. Similarly in our study also, most common presenting complaint of patients was of white discharge per vagina 40%.

The study conducted by Nadereh Behtash et al⁹ showed adequate cellularity in 94.7% and 92.1% cases of LBC and CS preparations respectively. This correlate with our study as it showed adequate cellularity in more no. of LBC (94%) cases compared to CS (86%).

The study conducted by Aboobacker and Shariff et al⁵

showed clean background in 4.2% and 90% cases of CS and LBC respectively. This correlates with our study as it showed clean background in more no. of LBC (74%) cases compared to CS (0%).

The study conducted by Aboobacker and Shariff et al⁵ showed uniform distribution in 5.8% and 69.2% cases of CS and LBC respectively. This correlates with our study as it showed uniform distribution in 6% and 60% cases of CS and LBC respectively.

The study conducted by Aboobacker and Shariff et al⁵ showed cellular overlapping in 86 % and 33.3% cases of CS and LBC respectively. This correlates with our study as it showed cellular overlapping in 94% and 40% cases of CS and LBC respectively.

The study conducted by Aboobacker and Shariff et al⁵ showed inflammatory background in 92.2% and 22.6% cases of CS and LBC respectively. This correlates with our study as it showed inflammation in 94% and 70% cases of CS and LBC respectively.

The study conducted by Ruchika Gupta et al¹⁰ showed distinct cell border in 83.1% and 84.8% cases of CS and LBC respectively. This correlates with our study as it showed distinct cell border in 82% and 86% cases of CS and LBC respectively.

The study conducted by Chandwani et al¹¹ showed nuclear irregularity in 20% and 10% cases of CS and LBC respectively. This correlates with our study as it showed nuclear irregularity in 18% and 10% cases of CS and LBC respectively.

The study conducted by Liu W et al¹², Singh VB et al⁶ and Díaz Rosario LA et al¹³ showed that the most common reason for unsatisfactory was low cellularity in both categories. This correlates with our study as it showed unsatisfactory smears in 07 and 03 cases of CS and LBC respectively. All of which are due to low cellularity in both categories. The study conducted by Tench et al¹⁴ showed that, there is increase in detection of pre invasive lesions and decrease in the number of indeterminate results such as ASC. This correlates with our study as it showed ASCUS in 05 and 01 cases of CS and LBC respectively.

The study conducted by Aboobacker and Shariff et al⁵ showed 0.4% cases of AGC NOS in both CPS and LBC. This correlates with our study as it showed 2% cases of AGC NOS in both CPS and LBC. The study conducted by Stabile et al¹⁵ showed more cases of LSIL in LBC as compared to CPS. This correlates with our study as it showed LSIL in 22 and 28 cases of CS and LBC respectively. The study conducted by Aboobacker and Shariff et al⁵ showed more cases of HSIL in CPS as compared to LBC. This correlates with our study as it showed HSIL in 08 and 07 cases of CS and LBC respectively.

The studies conducted by Budak M et al and Qureshi et al showed more cases of SCC in LBC than in CPS. This correlates with our study as it showed SCC in 07 and 09 cases of CS and LBC respectively.

The study conducted by Aboobacker and Shariff et al⁵ showed 0.4% cases of Adenocarcinoma in both CPS and LBC. This correlates with our study as it showed 0% and 2% cases of Adenocarcinoma in CPS and LBC respectively. Because the smear that was reported as Adenocarcinoma in LBC, was unsatisfactory for evaluation in CPS.

5. CONCLUSION:

This study was conducted to evaluate Liquid Based Cytology (EziPrep) and compare it with Conventional Cervical Smears and categorize according to "THE BETHESDA SYSTEM 2014".

In this study most of the Conventional Smears showed cellular

overlapping, inflammation, blood and mucus that obscure the epithelial cell morphology which was much reduced in Liquid Based Cytology (LBC).

Comparison of morphological details and results of cervical cytology smears by both methods showed that LBC method provides more representative sample with reduced obscuring material which allows better morphological evaluation and thus LBC is better in detection of preinvasive cervical lesions. LBC method also generated higher number of satisfactory smears and provides better cytomorphological features like cell borders, nuclear distortion and uniform distribution of cells compared to Conventional Smears.

The main aim of cervical screening test is detection of precancerous cervical lesions; it is obvious in this study Liquid Based Cytology certainly contributes to the achievement of this aim. It is inferred from our findings that Liquid Based Cytology is a superior screening test compared to Conventional Pap test. Taking into account the higher cost of Liquid Based Cytology, it could not be generalized but wherever it is feasible LBC can be used instead of Conventional Cytology.

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