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Original Research Article

Comparative evaluation of human papillomavirus DNA and liquid-based cytology in screening of cervical cancer

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ABSTRACT

Background: Cervical cancer ranks as the fourth most prevalent cancer among women worldwide with approximately 604,127 new cases and 341,831 deaths reported in 2020. It takes around 10-20 years for progression from mild dysplasia to invasive cancer, allowing early detection through screening .
Methods: This was a cross-sectional study conducted at the department of obstetrics and gynaecology at GSVM medical college Kanpur between december 2022 to december 2023 involving 241 women aged 30-65 years attending the gynecology OPD. Liquid-based cytology (LBC) and human papillomavirus (HPV) DNA testing was conducted on each woman irrespective of complain of presentation.
Results: The mean age of patients in this study was 39.65 years. HPV DNA had a higher sensitivity (95.24%) than LBC (89.68%). Specificity of HPV DNA (72.16%) was slightly lesser than LBC (79.92%). Positive and negative predictive value for HPV DNA and LBC are (46.1%, 96.22%) and (49.68%, 94.68%) respectively. Area under curve for HPV DNA (0.79) was higher than LBC (0.74) proving its higher diagnosing accuracy. The cluster analysis showed HPV+ clusters covered more area and distribution than LBC proving its better performance.
Conclusions: HPV DNA testing showed superior sensitivity compared to LBC. HPV DNA's higher diagnostic accuracy and broader coverage make it a more reliable screening tool.

Keywords: Cervical cancer, Positive predictive value, HPV DNA, LBC

INTRODUCTION

Cervical cancer ranks as the fourth most prevalent cancer among women worldwide, with approximately 604,127 new cases and 341,831 deaths reported in 2020. In 2020, 94% of deaths attributed to cervical cancer occurred in low- and middle-income countries.[1] In India, there are approximately 511.4 million women aged 15 years and older who are at risk of developing cervical cancer. Each year, an estimated 123,907 women are diagnosed with cervical cancer, resulting in 77,348 deaths from the disease. Cervical cancer is the second most common cancer among women aged 15 to 44 years in India, and it is also the second leading cause of cancer-related deaths among females in the country.[2]

Persistent infection with HPV is necessary for the development of cervical cancer and those with associated risk factors like cigarette smoking, immunodeficiency (HIV, following transplant surgery), early onset of sexual activity, multiple sexual partners, lower socio economic status, poor personal hygiene and tobacco use are at high risk for progression of lesions to invasive carcinoma.[3] Approximately 5% of women in the general population are estimated to have an ongoing infection with HPV 16 or 18 at any given time. High risk types, especially HPV 16 are found be highly prevalent in human population.[4] HPV 16

and 18 were the most carcinogenic which would cause more than 84.5% of cervical cancer.[5] Individuals with multiple HPV types co infection had lower likelihood of spontaneous clearance, increasing their risk of progressing to cancer.[6] Cervical cancer is considered preventable due to its long pre-invasive stage. The natural history typically spans 10-20 years from mild dysplasia to cervical carcinoma, allowing for early detection through screening.[7] Various screening techniques have been employed, including the Pap smear, LBC, and HPV testing. Pap test has proven to be a greatly effective tool for screening in countries with low resources if it is implemented to the majority of the population with drawback of high false positive rate.[8]

LBC offers advantages such as quicker reporting, reduced likelihood of missing cases, and the ability to perform HPV testing using residual fluid. However, it can still produce false negative results in a small number of cases.[9] HPV DNA testing is based on molecular studies, has demonstrated superior sensitivity in detecting cervical cancer risk. While some developing countries have explored using HPV DNA testing as a primary screening method, it remains experimental due to its higher cost. Potential benefits include higher sensitivity compared to cytology, automated high-throughput testing, and longer intervals between screenings. However, its major drawback is lower specificity compared to cytology, largely due to the prevalence of HPV infections, which vary significantly by age (approximately 40% in 20-year-olds to 20% in 30-year-olds). The study compares HPV DNA testing and LBC for cervical cancer screening. HPV DNA testing detects high-risk human papillomavirus strains, the primary cause of cervical cancer, and can identify women at risk before cytological abnormalities appear. It also helps identify persistent infections, a key factor in cancer progression. LBC on the other hand, examines cervical cells microscopically, detecting abnormal cell changes but may miss early-stage infections or subtle cellular changes. This study aims to evaluate and compare diagnostic accuracy of LBC and HPV DNA for screening of precancerous and cancerous lesions of cervix.

METHODS

This was a cross-sectional study conducted in the department of obstetrics and gynaecology, Upper India sugar exchange maternity hospital, GSVM medical college Kanpur from December 2022 to December 2023. A total of 241 women between the age group of 30-65 years attending the gynecology OPD, GSVM medical college, Kanpur were included in the study. Pregnant women, patient with history of cervical conization, pelvic irradiation, total hysterectomy was excluded in the study.

Written informed consent was obtained from all women participating in the study. Participants underwent a thorough medical history review and general physical examination and systemic examination. Per speculum examination was done to visually inspect the cervix for any visible abnormalities or lesions. For each women LBC and HPV DNA testing was done

LBC reporting was done according to modified Bethesda system. A positive LBC result typically includes findings such as ASCUS, ASCH (atypical squamous cells high grade squamous intraepithelial lesion), LSIL, HSIL, AGC (atypical glandular cell), Squamous cell carcinoma and adenocarcinoma and negative LBC result includes NILM (negative for intra epithelial lesion and malignancy) and inflammatory smears. After LBC, HPV DNA testing was conducted on the remaining samples using the sure path hybrid capture 2 technique. This method specifically detects high-risk HPV types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59 and 68). Samples showing presence of high-risk HPV types were classified as HPV DNA (+). Patients who tested positive for either LBC or HPV DNA or both positive were referred for colposcopy and cervical biopsy. Cervical biopsy was considered the gold standard for comparison of findings between LBC and HPV DNA testing. This means that biopsy results were used as the reference point to evaluate the accuracy of LBC and HPV DNA testing in detecting cervical abnormalities.

Statistical analysis

Data was collected entered and compiled in Microsoft excel sheet. The collected data was classified, tabulated, and analyzed using origin software and conclusions was drawn accordingly. Descriptive statistics for categorical variables were reported as frequency and percentage, while continuous variables were presented as mean, median, standard deviation, and quartiles. To determine associations between variables, appropriate tests of significance were applied, such as the chi-square test for categorical variables. For diagnostic performance, ROC analysis was done. In ROC analysis area under the curve (AUC) was assessed. A $p<0.05$ was considered significant.

RESULTS

The mean±SD of age in years was 39.65±8.62. The median of age in years was 38 with interquartile value (13). The age in Years ranged from 30-60. It was observed that majority of the participants were in the age group between 30-35 (44.39%) followed by 36-40 years (16.59%) and 41-45 years (17.01%). Participants aged between 46-50 years of age contributed 12.03% and participants with >50 years of age contributed 9.98%. 60.58% of the study participants were between the age of 21-25 years at the time of marriage followed by 15-20 years (32.78%) and 26-30 years (6.64%). Majority of the study participants belongs to class 4 of socioeconomic (66.83%) followed by 5 (18.67%), 3 (9.95%) 2 (3.73%) and 1 (0.82%). Majority of the participant were found to be asymptomatic at presentation (30.76%) followed by 26.13% with discharge per vaginum, 22.81% with irregular vaginal bleeding, 11.60% heavy menstrual bleeding, 6.64% presented with postmenopausal bleeding. On clinical examination most of the study participant had normal cervix (37.75%). Cervical

erosion was the most common abnormality detected (35.26%) followed by cervix hypertrophy (21.58%), Nabothian follicle (4.14%), polyp (0.88%) and growth on cervix (0.41%) (Table 1).

Table 1: Distribution of the participants in terms of demographic profile, symptoms and clinical profile.

Characteristics	N	Percentages (%)
Age (in years)		
30-35	107	44.39
36-40	40	16.59
41-45	41	17.01
46-50	29	12.03
>50	24	9.98
Mean±SD	39.65±8.62	
Age at marriage (in years)		
15-20	79	32.78
21-25	146	60.58
26-30	16	6.64
Socioeconomic status		
1	2	0.82
2	9	3.73
3	24	9.95
4	161	66.83
5	45	18.67
Symptoms		
Heavy menstrual bleeding	28	11.60
Irregular vaginal bleeding	55	22.81
Asymptomatic	74	30.76
Post coital bleeding	5	2.06
Postmenopausal bleeding	16	6.64
Discharge P/V	63	26.13
Clinical sign		
Cervix erosion	85	35.26
Cervix hypertrophy	52	21.58
Growth on cervix	1	0.41
Nabothian follicle	10	4.14
Normal	91	37.75
Polyp	2	0.88

On LBC 218 reports were negative with 48.14% [116] were inflammatory and 42.33% (102) were NILM. Among abnormal reports 4.15% were LSIL, 2.48% ASCUS, 2.08% HSIL and 0.82% were found to be Ca cervix. HPV DNA was negative in majority of cases (86.36%) only 13.64% were HPV DNA positive most common Hr HPV DNA detected was HPV 16 (8.29%). Colposcopy guided cervical biopsy was performed among patients who were either LBC or HPDNA or both positive. Most common abnormality detected on colposcopy was atypical vessels (8.29%) followed by acetowhite area in 6.22% and in 1.65% cases colposcopy was normal and unsatisfactory in 0.41%. most common abnormality detected on histopathology was chronic cervicitis in 12.49% followed by CIN 1 in 1.65%, CIN2 in 0.82%, CIN3 in 0.82%, adenocarcinoma in situ in 0.41% and SCC in 0.41%

Table 2: Distribution of study participants based on their LBC, HPV DNA, colposcopy and histopathology (in LBC or HPV positive patients).

Variables	N	Percentages (%)
LBC		
NILM	102	42.33
ASCUS	6	2.48
LSIL	10	4.15
HSIL	5	2.08
Inflammation (mixed infection)	116	48.14
Carcinoma	2	0.82
HPV DNA		
16	20	8.29
18	1	0.41
52	1	0.41
54	1	0.41
56	2	0.82
58	1	0.41
66	3	1.24
68	3	1.24
51,56,66	1	0.41
Negative	208	86.36
Colposcopy		
Acetowhite area	15	6.22
Atypical vessels	20	8.29
Normal	4	1.65
Unsatisfactory	1	0.41
Histopathology		
Adenocarcinoma *in situ*	1	0.41
Chronic cervicitis	30	12.49
CIN 1	4	1.65
CIN 2	2	0.82
CIN 3	2	0.82
SCC	1	0.41

*201 patients considered negative.

Positive detection rates of different schemes increased with the severity of cervical lesions; positive detection rates of the 2 methods for the same grade lesion were significantly different ($p<0.00375$). When subjects were tested by LBC or HPV alone, the positive detection rates of high-grade cervical lesions all differences were statistically significant ($p=0.3$) (Table 3).

Sensitivity of HPV DNA was (95.24%) as compare to LBC (89.68%) and specificity of HPV DNA was 72.16% as compare to LBC 79.92% in detection of precancerous and cancerous lesions of cervix. Positive and negative predictive value of HPV DNA was 46.1% and 96.22% and for LBC was 49.68% and 94.68% respectively. Area under curve for HPV DNA was found to be higher 0.792 compare to 0.746 for LBC it implies HPV DNA was best predictor of preinvasive and invasive lesion of cervix (Table 4).

Table 3: Detection rates of different cervical lesions by different test methods.

Result of histopathology confirmation	Total cases	LBC+	Chi square	P value	HPV+	Chi square	P value
Adenocarcinoma *in situ*	1	1	12.609	0.0619	1	18.025	0.0375*
Chronic cervicitis	30	16			24		
CIN 1	4	2			3		
CIN 2	2	2			2		
CIN 3	2	2			2		
SCC	1	1			1		

*P value statistically significant

Table 4: Detection rates of test methods.

Test methods	Sensitivity (%)	Specificity (%)	Diagnosis accuracy (%)	Positive predictive value (%)	Negative predictive value (%)	Area under curve
LBC+	89.68	79.92	81.64	49.68	94.68	0.746
HPV+	95.24	72.16	71.92	46.1	96.22	0.792

DISCUSSION

In the present study, mean age of the participants was recorded as 39.65 years. The results demonstrated that the maximum participants were in the age group between 30-35 followed by 36-40 years and 41-45 years. The results demonstrated that the majority of the participants were in the age group belonged to 30-35 years of age while the least number of participants belonged to more than 50 years of age. This is because the recommendation of screening starts at early age 21 years but in our country due to lack of awareness and ignorance the initial screening starts at a later stage. The study done by Raj et al demonstrated that majority of the patients were 36-40 years of age, followed by age groups 41-45 and 46-50 years.[10] The study of Shreya et al and Zarchi et al found median age of 43.36 and 42 years respectively in their studies.[10,11]

In the present study majority of patients 66.83% belonged to lower class of socioeconomic status according to modified Kuppuswamy scale. The results of this study co related with the study of Shreya et al in which maximum number patients (44.10%) belonged to lower class of socioeconomic status. Overcrowding and poor nutrition and lower socio-economic status all lead to development of Ca cervix.

The study found that most participants were asymptomatic, with a minority experiencing post-menopausal bleeding. This contrasts with previous studies by Suguna et al, Gopal et al and Shreya et al which mostly reported discharge per vaginum.[10,12,13] In this study these tests were used as screening method i. e., most of patients were asymptomatic on presentation.

We found that the majority of study participants had normal cervix (37.75%) on per speculum examination. Erosions on the cervix was the major clinical sign presented in participants (35.26%) followed by cervix hypertrophy (21.58%), while least participants (0.41%) had clinical sign of growth on cervix. Similar observations were also made by Gupta et al in which unhealthy cervix was reported by majority (40%), followed by cervix erosion (34.55%), polyp in least (3.64%) study participants.[14] In another study by Raj et al reported that hypertrophy was found in the majority (84.38%) followed by cervical ectropion (25.88%), and least presented with growth at cervix (0.39%).[10] Cervical erosions during intercourse were the source for HPV invasion into cervix and its progression in presence of various risk factors.

The study found that 48.1% of participants had non-specific inflammation due to mixed infection in LBC, with Candida, bacterial vaginosis, and trichomoniasis being the most common. Non-specific inflammation was present in 42.23% of participants, with HSIL and carcinoma being the least common. High-grade lesions were present in a minority of participants, but it is crucial to focus on those with the highest chances of conversion into ca cervix to prevent carcinoma-related morbidity and mortality. Previous study by Virta et al found inflammation in 48.61% of participants, followed by ASCUS in 4.94%, endometrial cell in 0.79%, and cancer in 0.39%.[15]

In our study we found that 13.64% of the study participant were found to be high risk HPV DNA positive out of which majority (8.29%) of the study participants were HPV 16 positive. Other high risk HPV types contributed only 5.35%. In the studies done by Virta et al and Shreya et al they also found that 15.45% and 7.70% of the participants respectively were found to be high risk HPV DNA positive which was similar to our study.[15,10] HPV 16 is the most common strain responsible for majority of cases of high-grade lesions and Ca cervix.

More than one colposcopic findings were present in same patient, the most common colposcopic finding in our study

was atypical vessels (51.28%) followed by acetowhite areas (38.46%). Study conducted by Gupta et al and Handa et al found that atypical vessels were present in 81.8% and 75% of patients respectively which correlated with the findings our study.[14,16] This is because neo-angiogenesis or neovascularization is the initiation of carcinogenesis in high grade lesions which is most readily visible on application of green filter during colposcopy.

Colposcopy guided cervical biopsy was done in only 40 participants in whom both LBC and Hr HPV DNA tests or either, one was positive. Majority (12.49%) of the study participants presented with chronic cervicitis, and SCC was present in the least (0.41%) number of the study participants. Similar findings were reported in the study conducted by Shreya et al and Gupta et al where chronic cervicitis was present in 77.02% and (72.72%) participants, invasive cancer in 6.75% and 5.45% of the study participants respectively.[10,14] Since majority of the participants had lower grade lesions i.e. ASCUS and LSIL therefore the most common histopathological finding was chronic cervicitis. Therefore, our study suggests that HSIL and higher-grade lesions have to be managed more appropriately at higher centers.

In the present study we observed that the area under curve for HPV DNA was found to be higher as compared to LBC which was significant and hence HPV DNA possess higher sensitivity (95.24%) as compared to LBC (89.68%) proving its diagnosing accuracy as 81.64% ($p<0.05$). In the study conducted by Shreya raj et al it was observed that the sensitivity of LBC (86.7%) in the testing of high-grade (≥CIN2) cervical lesions was slightly lower than that of HPV test (92.3%), but the specificity and accuracy of LBC (74.7% and 76.2%) were higher than that of HPV test (69.8% and 72.6%), and the differences were statistically significant ($p<0.05$) which co related with our results.[10]

Limitations

The present study included only 241 patients, which was a small sample size, which may restrict the generalizability of the results so similar studies with larger sample size are needed. Also, this was a single center study. As the LBC and HPV DNA is an expensive test affordability of the test was an issue because this test was not available at our institute.

CONCLUSION

The study involved 66.83% of participants, with a median age of 38 years. Cervical erosion and hypertrophy were common clinical findings, and most had non-specific inflammation in LBC report. The study compares HPV DNA testing and LBC in cervical cancer screening. HPV DNA testing showed superior sensitivity (95.24%) compared to LBC (81.64%). It was positive in 13.64% of participants and the most common strain detected was HPV 16. Histopathological findings indicated chronic cervicitis in majority and CIN1 in a small percentage. HPV DNA's higher diagnostic accuracy and broader coverage make it a more reliable screening tool.

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REFERENCES

1. Stelzle D, Tanaka LF, Lee KK, Khalil AI, Baussano I, Shah AS, et al. Estimates of the global burden of cervical cancer associated with HIV. The Lancet Global Health. 2021;9(2):e161-9.
2. Bruni L, Barrionuevo-Rosas L, Albero G, Serrano B, Mena M, Gómez D. ICO information center on HPV and cancer (HPV Information Center). Human papilloma virus and related diseases in Bangladesh. Summary report. 2015.
3. Franco EL, Villa LL, Sobrinho JP, Prado JM, Rousseau MC, Désy M, et al. Epidemiology of acquisition and clearance of cervical human papillomavirus infection in women from a high-risk area for cervical cancer. The J Infect Dis. 1999;180(5):1415-23.
4. Cohen PA, Jhingran A, Oaknin A, Denny L. Cervical cancer. The Lancet. 2019;393(10167):169-82.
5. Chen W, Zhang X, Molijn A, Jenkins D, Shi JF, Quint W, et al. Human papillomavirus type-distribution in cervical cancer in China: the importance of HPV 16 and 18. Cancer Causes Control. 2009;20:1705-13.
6. Fowler JR, Maani EV, Dunton CJ, Gasalberti DP, Jack BW. Cervical Cancer. StatPearls. Treasure Island (FL): StatPearls Publishing. 2024.
7. Yuan Y, Cai X, Shen F, Ma F. HPV post-infection microenvironment and cervical cancer. Cancer Letters. 2021;497:243-54.
8. Rathore R, Gupta S, Gupta M. To compare liquid based cytology and HPV DNA testing in detection of cervical intraepithelial Neoplasia. Int J Clin Obstet Gynaecol 2024;8(5):43-51.
9. Wang J. Analysis of the application values of different combination schemes of liquid-based cytology and high-risk human papilloma virus test in the screening of high-grade cervical lesions. Braz J Med Biological Res. 2019;52(1):e7517.
10. Raj S, Srivastava M. Comparative analysis of HPV DNA and LBC for screening of cervical cancer in women with unhealthy cervix. Age. 2022;195:38-50.
11. Karimi-Zarchi M, Peighmbari F, Karimi N, Rohi M, Chiti Z. A comparison of 3 ways of conventional pap smear, liquid-based cytology and colposcopy vs cervical biopsy for early diagnosis of premalignant lesions or cervical cancer in women with abnormal conventional pap test. Int J Biomed Sci. 2013;9(4):205.
12. Suguna M, Rajeshwar A, Pasula S. A study on pap smear and colposcopy in unhealthy cervix in women. Int J Med Sci Public Health. 2014;3(7):889-91.

13. Gopal N, Joshi PS, Pukale RS. Colposcopic findings in Unhealthy Cervix and its comparison with Cytology and Histopathology. J Evolution Med Dental Sci. 2013;2:4663-71.
14. Gupta P, Faruqi M, Chandra S, Shah SS, Kulshrestha R. The correlation between colposcopy, cervical cytology and histopathology in the diagnosis and management of cervical lesions: a cross sectional study. Int J Reproduct Contracept Obstetr Gynecol. 2020;9(7):2729-36.
15. Chauhan V, Bijyal S. Comparative Study of Liquid-Based Cytology Versus Hpv Dna for Screening of Cervical Cancer At A Tertiary Hospital. Europ J Molecular Clin Med. 2022;9:2129-35.
16. Handa PR, Dhall GI. Colposcopy as diagnostic aid in cases of suspicious cervix. J Obstet Gynaecol. 1991;41(6):785-92.

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