

Association of High-Risk Cervical Human Papillomavirus with Demographic and Clinico-Pathological Features in Asymptomatic and Symptomatic Women

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➤ Background

Human Papillomavirus (HPV) infection is a common cause of cervical cancer. Based on its oncogenic potential, genotypes with high, low and intermediate risks have been classified. The present study was conducted to detect cervical HPV infection in women with and without symptoms and to study high-risk genotypic distribution of cervical HPV infection among them.

➤ Methodology

A hospital-based cross-sectional study was conducted from August 2022-September 2023 wherein 81 women in the age group 20-70 years, attending STI/RTI clinic at Government Medical College and Hospital, Nagpur were screened by cervical cytology for cellular changes and histopathology or colposcopy wherever required. RT-PCR was performed to detect high-risk HPV genotypes.

➤ Results

Out of 81 patients 35 (43.2%) were symptomatic and 46 (56.8%) were asymptomatic. Most of the asymptomatic patients belonged to < 40 years accounting for 35 (76.09%) patients while most of the symptomatic patients belonged to > 40 years accounting for 18 (51.42%). Age of marriage of 20-30 years and a parity of two was found in 29 (63.04%) and 16 (34.78%) of patients respectively in asymptomatic group while the above parameters were found in 20 (57.14%) and 16 (45.71%) of symptomatic patients respectively. Patients attending the clinic were mainly from lower socioeconomic status in both asymptomatic and symptomatic group. Out of 60 adequate smears examined for cervical cytology 8, 46, 4, and 2 showed normal, inflammatory, precancerous and cancerous changes respectively. HPV DNA was detected in 5 (6.17%) patients. Amongst HPV positive patients only HPV-18 and HPV-16 genotypes were detected in 2 (40%) and 3 (60%) patients respectively.

➤ Conclusion

High-risk HPV-16 and HPV-18 genotypes were detected in 6.17% patients. This projects a baseline data for treatment, prophylaxis, post vaccination surveillance and future research.

Keywords:- HPV Genotype, RT-PCR, Cytology.

I. INTRODUCTION

Human papillomavirus (HPV) is the most prevalent sexually transmitted infection with wide variety of genotypic distribution worldwide.^{1,2} Majority of the lesions are benign and are cleared by host immune system however, persistent HPV infection is worrisome as it is associated with more than 5% of all cancers globally.³ According to GLOBOCAN data (2020), cervical cancer is the fourth most frequently diagnosed cancer and fourth leading cause of cancer deaths in women with estimated 6,04,000 new cases and 3,42,000 deaths worldwide.¹

Early detection of cervical HPV infection is important to prevent progression of premalignant lesions to cancer.⁴ Despite of high incidence of cervical cancer in India, studies on HPV prevalence and genotype distribution are scanty. The present study was undertaken to know the prevalence and genotype distribution of cervical HR-HPV infection in symptomatic and asymptomatic women attending a Regional Centre for sexually transmitted infections in Vidarbha region of Maharashtra.

II. MATERIALS AND METHODS

➤ Study Design

A hospital-based cross-sectional study was conducted from August 2022 to September 2023 wherein 81 women in the age group 20-70 years, attending STI/RTI clinic at Government Medical College and Hospital, Nagpur were screened by cervical cytology for cellular changes and histopathology or colposcopy wherever required and RT-PCR for high-risk HPV genotypes.

➤ Sample Collection

Cervical samples of exfoliated cells were collected for HPV DNA detection using HIMEDIA cyto-brush. This was inserted 2 to 3 cm in cervix and gently the mucosal scraping was taken from the wall by rotating it for 5 to 10 seconds. Cyto-brush was removed and stored in Viral Transport Medium at -20° C for genotyping. With a spatula, scraping

from the ectocervix was taken and pap smear prepared on a slide.

➤ DNA Extraction and HPV Genotype Detection

DNA extraction of 81 samples was done using GSure DNA extraction kit (GCC Biotech India Pvt Ltd) as per manufacturer's instructions. DiAGSure HPV 16 high-risk genotypes detection TaqMan PCR kit was used for HPV detection and genotyping by real-time polymerase chain reaction (RT-PCR).

III. RESULTS

In the present study, out of 81 patients 46 were asymptomatic and 35 were symptomatic. Symptomatic patients presented with symptoms like intermenstrual bleeding, white discharge, pain in abdomen, postmenopausal bleeding, postcoital bleeding, dyspareunia, weight loss etc. Most of the asymptomatic patients belonged to < 40 years accounting for 35 (76.09%) patients while most of the symptomatic patients belonged to > 40 years accounting for 18 (51.42%). Age of marriage of 20-30 years and a parity of two was found in 29 (63.04%) and 16 (34.78%) of patients respectively in asymptomatic group while the above parameters were found in 20 (57.14%) and 16 (45.71%) of symptomatic patients respectively. Patients attending the clinic were mainly from lower socioeconomic status in both asymptomatic and symptomatic group as shown in Table 1.

Detection of HPV infection by RT-PCR showed that 2 out of 46 asymptomatic and 3 out of 35 symptomatic patients were positive for HPV DNA. The most common genotype prevalent was HPV-16 present in 3 patients followed by HPV-18 in 2 patients. Other HR-HPV genotypes (HPV-31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 67, 68, 70) were not detected in our study (Table 2 and 3).

Correlating HR-HPV genotypes with demographic characters it was found that 4 (80%) out of 5 positive patients belonged to age group 25-40 years, 4 (80%) had early age of marriage of < 20 years, 3 (60%) belonged to para \geq three and 4 (80%) were of lower socioeconomic status as shown in Table 4.

We found that out of 5 patients positive for HR-HPV genotypes, 3 were symptomatic and 2 were asymptomatic. Comparison of HR-HPV genotypes with clinical presentation and cytological or histopathological findings showed that 2 out of 3 symptomatic patients had erosion clinically, high-grade squamous intraepithelial lesion (HSIL) and atypical squamous cells of undetermined significance (ASCUS) on cytology and dysplasia on histopathology respectively and the remaining one had frank growth clinically, with ASCUS on cytology and squamous cell carcinoma on histopathology. The 2 asymptomatic HR-HPV positive patients showed inflammation on cytology as shown in Table 5.

IV. DISCUSSION

In the present study 81 outpatient women were tested for HR-HPV and among these 5 patients were positive for HPV infection. Overall HR-HPV prevalence was 6.17% (5/81). The prevalence in symptomatic patients was 8.58% which was slightly higher than 4.34% found in the asymptomatic patients. These findings were comparable with the study of Mishra et al that showed prevalence of HR-HPV infection to be 5.5%. They reported prevalence in symptomatic patients and asymptomatic patients as 6.6% and 3.03% respectively.⁴ Other population-based studies from Osmanabad, Mumbai, Trivandrum, and from two different cities in Kolkata showed prevalence of 10.3%, 6.3%, 4.8%, 7.8% and 5.2% respectively.^{5,6} The most common HR-HPV genotype in our study was HPV-16 (60%) followed by HPV-18 (40%). Other HR-HPV types (HPV-31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 67, 68, 70) and infection with multiple types was not detected in our study. A study by Sowjanya et al and Senapati et al from Andhra Pradesh and Odisha respectively showed similar findings of HPV-16 and HPV-18 being the common genotypes detected. However, unlike our study other HR-HPV genotypes along with infection with multiple genotypes was also seen in these studies.^{7,8} Worldwide HPV-16 and 18 constitute approximately 75% of all cervical cancer cases. In our study HR-HPV positivity was associated with risk factors like early age of marriage, high parity and low socioeconomic status. Kennedy et al and Molina Pineda et al also showed positive association of HPV infection with high parity.^{9,10} Lower socioeconomic status can be attributed as risk factor for cervical cancer as there is lack of screening and awareness of HPV infection and lack of hygiene in this population group. A study conducted in South India evaluated cofactors and their association with cancer and found that low socioeconomic status is a major risk factor associated with cervical cancer.¹¹

V. CONCLUSION

The present study provides information about the genotype distribution among women of the state of Maharashtra which would help in planning an appropriate strategy for disease monitoring and control. An effective vaccination program based on regional HPV epidemiological profile along with screening is necessary to reduce the cervical cancer burden in India. This study offers a baseline data for treatment, prophylaxis, post vaccination surveillance and future research.

VI. LIMITATIONS

HR-HPV types other than HPV-16 and 18 were not detected in our study. This could be because our study included patients attending the STI/RTI clinic and did not focus on cervical cancer patients. Further research including cervical cancer patients can help detect more HR-HPV types other than HPV-16 and 18. Our study included unvaccinated women; however, post vaccination surveillance could also throw light on effectiveness of HPV vaccination in vaccinated women.

Table 1: Distribution of sociodemographic characters

Factors	No of patients n (%)	
	Asymptomatic (n=46)	Symptomatic (n=35)
Age ≤ 40	35 (76.09%)	17 (48.58%)
Age > 40	11 (23.91%)	18 (51.42%)
Age of marriage < 20 years	17 (36.96%)	15 (42.86%)
Age of marriage 20-30 years	29 (63.04%)	20 (57.14%)
Parity < 3	34 (73.91%)	21 (60.00%)
Parity ≥ 3	12 (26.09%)	14 (40.00%)
Low socioeconomic status	35 (76.09%)	33 (94.29%)
High socioeconomic status	11 (23.91%)	2 (5.71%)

Table 2: Detection of HR-HPV infection by RT-PCR

Sr no.	RT-PCR findings	No of patients n (%)	
		Asymptomatic (n=46)	Symptomatic (n=35)
1.	Positive for HPV	2 (4.34%)	3 (8.58%)
2.	Negative for HPV	44 (95.66%)	32 (91.42%)

Table 3: High-risk genotype distribution of HPV in RT-PCR positive patients

Sr no.	High risk HPV genotypes	No of patients n (%)	
		Asymptomatic (n=46)	Symptomatic (n=35)
1.	HPV-16	1 (2.17%)	2 (5.71%)
2.	HPV-18	1 (2.17%)	1 (2.86%)
3.	Other HR-HPV (HPV 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 67, 68, 70)	0	0
Total		2 (4.34%)	3 (8.57%)

Table 4: Comparison of HR-HPV genotypes with demographic characters

Sr no.	Patient Id	HR-HPV genotype	Age	Age of marriage	Parity	Socioeconomic status
1.	4474	HPV-16	32	22	Two	Low
2.	8312	HPV-16	38	19	Four	Low
3.	1732	HPV-16	25	19	Two	Low
4.	7385	HPV-18	27	18	Five	Upper middle
5.	1136	HPV-18	61	19	Three	Low

Table 5: Correlation of HR-HPV genotype with clinical presentation and cytological /histopathological findings

* HSIL – high grade squamous intraepithelial lesion
 ** ASCUS – atypical squamous cells of undetermined significance
 *** SCC – squamous cell carcinoma

Sr no	Patient Id	PCR	Group	Clinical presentation	Cytological findings	Histopathological findings
1.	4474	HPV-16	Symptomatic	Erosion	HSIL*	Moderate dysplasia
2.	8312	HPV-16	Symptomatic	Erosion	ASCUS**	Mild dysplasia
3.	1732	HPV-16	Asymptomatic	-	Inflammation	-
4.	7385	HPV-18	Asymptomatic	-	Inflammation	-
5.	1136	HPV-18	Symptomatic	Visible growth	ASCUS	Moderately differentiated SCC***

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