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Age-stratified high risk HPV DNA Testing: A retrospective study in jagjivan ram railway hospital a tertiary center, Mumbai, India

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Abstract

Background: Human Papilloma Virus (HPV) infection is a key etiological factor in cervical cancer. While screening strategies vary globally, the role of HPV DNA testing as a primary screening tool continues to be refined, particularly across different age groups. Age stratified screening for high risk HPV Type 16 and 18 is a paramount importance as persistent infection of these two strains are responsible for nearly 80 to 85% of cervical cancers worldwide.

Objective: The main objective of this study is to compare the necessity of age stratified high risk HPV DNA testing in women of different age group. To analyse the data and to get an overall insight to proper utilization of the resources while screening for cervical cancer especially in the third world countries where availability of resources is a main obstacle in providing the proper preventive health care services.

Methods: This observational study analysis was conducted using equipment PathoDetectTM HPV from Mylab Company for RTPCR amplification of specific target sequence E6/7 region of HPV 16 and 18 and detected by target specific probe data from 1825women of different age group who attended the Gynaec OPD of the hospital for screening. Patients were stratified in to six age groups. High risk HPV DNA testing for type 16 and 18 was done and results were compared between the groups. SPSS version 24 was used for statistical analysis, descriptive statistics was used to calculate frequency, percentage, mean, and standard deviation and HPV positivity rates. Chi–Square used as inferential Statistics for significance.

Results: Among the 1825 women tested, 403were aged 18 to 30 years out of which 14 were tested positive, 514 were between 31 to 40 years and 12 tested positive, 498 were between 41 to 50 years and16 tested positive, 253 were between 51 to 60 years and 7 tested positive, 95 were between 61 to 70 years 2 tested positive and 62 were aged > 71 years and one tested positive. Over all positivity rate among 1825 is 52 with over all positivity rate of 2.85% and p-value across all group is < 0.0001.

Conclusion: High risk HPV DNA positivity is significantly higher in women aged younger (18-30 yrs.) than the older age group, highlighting the increased necessity for the routine high risk HPV DNA testing in this age group. Tailored screening protocols based on age may optimize detection and resource allocation.

Keywords: HPVDNA Testing, HPV Positivity, age stratification, cervical cancer, screening

Introduction

Cervical cancer remains one of the most preventable malignancies in women all over the globe, predominantly caused by Human Papilloma Virus (DNA virus). Recurrent or persistent infection with high-risk Human PapillomaVirus (HPV) types16 and 18 causes nearly 80-85% of cervical cancer and other intermediate to low risk Human Pappilloma Virus types 31, 33, 45, 52,56 and, 58 are responsible for 10-15% of cervical cancer [1]. Nearly 70% of death due to cervical cancer occurs in low and middle income countries all over the world [2, 3]. In May 2018, the WHO Director-General announced a global call for action to eliminate cervical cancer [4], WHO anticipates for a global elimination of cervical cancer, which can be defined as an incidence of cases < 4 per 1,00,000 women per year globally. In that direction World Health Organization plans a 90-70-90 Global strategy to achieve its goal for 2030to eliminate cervical cancer, that is 90% of girls aged 15 years should receive HPV vaccine, 70% of all women should be screened for cervical cancer at least by 35years of age and again at the age of 45 years and 90% women identified with cervical disease (precancerous and invasive cancer) cases should receive treatment [5].

The introduction of HPV DNA testing has revolutionized the cervical screening, offering high sensitivity for detecting precancerous lesions at early stage. According to the World Health Organization, the current recommended screening strategy for HPV DNA testing starting at least at the age 30 and repeated every 5 to 10 years depending on the country's resource setting [6], some regions and in HIV infected population HPV testing is recommended from age of 25years. Cytology (Papsmear) or VIA (visual inspection with acetic acid) may be used where HPV testing is not available. The goal for 2030 is to screen 70% of women at least once by age 35 years and again at the age of 45 years [6]. However, the cost-effectiveness and utility of universal HPV DNA testing are debated, especially for vounger women where transient infections are more common and in third world countries where resources setting is an obstacle^[7] This study aims to explore the age-related necessity of high risk HPV DNA testing by comparing positivity rates by stratifying all the women tested into six age groups aged between 18 to 30 years, 31 to 40 years, 41 to 50 years, 51to 60 years, 61 to 70 years and 71 years and above, in a tertiary care hospital in a closed group of Railway beneficiaries in Mumbai.

Materials and Methods

This hospital based retrospective observational study analyzed the HPV-DNA test findings of 1,825 women aged between youngest being 18 years to oldest being 84 years. All the women were tested for high risk HPV DNA testing for type 16 and 18 strains at the central molecular lab of the pathology department of Jagjivan Ram Railway Hospital of Western Railways a tertiary center in Mumbai between December 2022 to Aug 2025.High risk HPV DNA testing was conducted by using the equipment PathoDetectTM HPV from Mylab Company for RTPCR amplification of specific target sequence E6/7 region of HPV 16 and 18 and detected by target specific probe. Data from 1,825 women of different age group who attended the Gynaec OPD of the hospital for screening, were stratified in to six age groups.

Inclusion criteria

- All the married women aged 18 years and above up to 84 years of age who attended Gynaec OPD for cervical cancer screening.
- Women with of chronic cervicitis on per speculum examination and willing for testing.
- Those without history any major STD infections like HIV.

Exclusion criteria

- Women aged below 18 years.
- Diagnosed cases of Carcinoma cervix.
- Women with history any active major STD like HIV infection etc.
- Women who are not willing for the procedure.

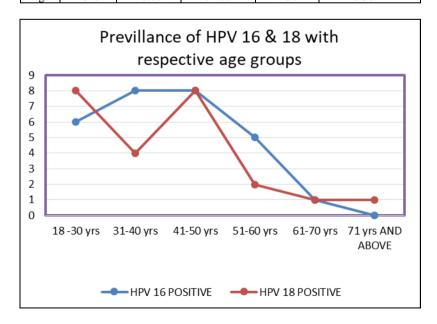
Data of the all eligible women were age stratified and analysed with respect to age and HPV DNA test results. These women were divided into six age groups between 18 to 30 years, 31 to 40 years, 41 to 50 years, 51to 60 years, 61 to 70 years and 71 years and above. The results were compared between the groups. SPSS version 24 was used for statistical analysis, descriptive statistics was used to calculate frequency, percentage, mean, standard deviation and HPV positivity rates, Chi-Square used as inferential Statistics for significance. Necessary institutional ethical committee approval was taken before the study.

Results

The present study involved total of 1825 women who were tested for high risk HPV DNA, age distribution of this study youngest being 18 years and oldest is 84 years, mean age for the overall women who have been tested is 41.45 years with Standard deviation of 16.50. All the women tested were stratified in to six age groups. Data was analysed by descriptive statistics using SPSS version 24 and Chi-Square used as inferential Statistics for significance.

Table 1: Over all age and mean age distribution of the study

	Number	Minimum	Maximum	Mean	Std. Deviation
Age	1825	18.00	84.00	41.45041	16.50411



Age distribution of women screened are 18-30 years group is N=403 (22.0 8%) and 14 tested positive for high risk HPV DNA, yielding a positivity rate of 3.47% highest among all study group. In the 31 to 40 years age group N=514 (28.16%) and 12 tested positive, with a positivity rate of 2.33%. Among 41 to 50 years age group N=498 (27.92%) and 16 tested positive with the positivity rate of 3.21%, where as those age 51 to 60 years age group N=253(13.86%) and 07 tested positive, (2.77%). Among 61 to 70 years age group N=95 (5.21%) 02 tested positive (2.11%), where as in those aged 71 years and above there were only N=62 (3.34%) but only one tested positive (1.61%) with the overall positivity rate of 2.85%.

Table 2: Describe the distribution of Age, Mean and SD of the study groups

Age Yrs.	No	Mean	Std. Deviation	Std. Error	Mini	Maxi
18-30	403	24.0000	2.76864	10413	18.00	30.00
31-40	514	35.5000	2.82513	10670	31.00	40.00
41-50	498	45.5000	2.75524	10535	41.00	50.00
51-60	253	55.5000	2.53361	12182	51.00	60.00
61-70	95	65.5000	2.75662	10561	61.00	70.00
>71	62	77.5000	2.85362	10570	71.00	84.00
Total	1825	41.45041	16.50411	10822	18.00	84.00

The mean age distribution among 18 to 30 years is 24.00 suggestive of early marriage and sexual exposure with positivity rate for HPV DNA at 3.47% highest among all groups. The mean age distribution in 71 years and above is 77.50 with positivity rate of 1.61% suggesting majority of them are sexually inactive with least exposure. This group has the lowest HPV DNA positive rate of 1.61%.

Over all positivity rate among N=1825 (100%) is 52 tested positive with over all positivity rate of 2.85% and p-value across all groups is < 0.0001.

This observation suggests that in our study HPV infection is common among younger age group compare to the older age group.

Table 3: Cross tabulation of age distribution and positivity of HPV DNA test

			Positive	Negative	Total	
	< 30	Count	14	389	403	
	< 30	% within ages	3.47%	96.53%	100.0%	
	31-40	Count	12	502	514	
		% within ages	2.33%	97.67%	100.0%	
	41-50	Count	16	482	498	
1 00		% within ages	3.21%	96.79%	100.0%	
Age	51-60	Count	07	246	253	
		% within ages	2.77%	97.23%	100.0%	
	61-70	Count	02	93	95	
	01-70	% within ages	2.11%	97.89%	100%	
	> 71	Count	01	61	62	
		% within ages	2.85%	98.39%	100%	
		Count	52	1773	1825	
T	otal	% within ages	2.85%	97.15%	100%	

Discussion

The results of the present study indicates a substantially higher rate of HPV DNA positivity observed 3.47% among women aged between 18-30 years group. Next highest positivity is among 41 to 50 years group with 3.21% and among 51 to 60 years group with 2.77%. Our idea of stratifying the study groups in to six group yielded a better clarity that all the available resources should be concentrated to screen all the women who

are not immunized (vaccinated) for HPV and likely to have more of persistent or recurrent High risk HPV infection leading to CIN 3 and invasive cervical cancer, thereby it is possible to detect early and treat them effectively. HPV prevalence was highest in women younger than 35 years of age, declining in middle age. In Africa, the Americas, and Europe, a clear second peak of HPV prevalence was observed in women aged 45 years or older [8, 9]. In our study highest infection rate observed in 18-30 years age group with 3.37% positivity, which is comparable to other studies where they found higher rate of infection among vounger age group aged below 30 years with the comment that they are more exposed to sexual activities more than the older. Our study group involved the closed community of Railway employees and their beneficiaries and they are well aware of the HPV vaccination age group. Indeed, in women of Europe and North America, the peak of HPV infection happens in women in their 20s, with a steady decrease in prevalence as age increases [10, 11]. However in this study age group between 41-50 years shows a spike in infection with positivity rate of 3.21% which is contrary to other study where the rate of infection decreases as the age advances. These findings challenge the existing recommendations advocating for age-targeted HPV screening strategies and also that younger women are more likely to clear HPV infections spontaneously [12], suggesting that primary HPV testing may not be beneficial in this group. On the contrary, persistent infections in older women are more likely to progress, making HPV DNA testing more crucial. However, our study found that the peak of high risk HPV infection in women is in the age group of those aged less than 30 years. This peak was also noted in previous studies in China [13, 14], which might be related with the onset of sexual activity among this population. Even though women in this age group have a higher rate of infection in China and about 80% of HPV infections are cleared within a year, [15] this group could be testing positive more often for infections that will not lead to cancerous lesions, causing the specificity in this age group to decrease. Conversely, the lower rate of HPV infection in older women in our population might explain the high specificity in this age group. Considering our small percentage of women tested positive in age group of 71 years and above (1.61%), our public health care system must concentrate more on vaccination of the younger group along with primary HPV testing, on the other hand we can concentrate and divert the available resources towards high risk HPV DNA testing for women less than 30 years also, in doing so we can identify the women with risk of recurrent or persistent infection which leads to dysplasia and CIN lesions. Timely vaccination and proper health education about HPV infection and its prevention to younger women will go a long way in achieving the WHO goal of cervical cancer eradication.

Our study has limitations Firstly that this study is conducted in an urban population where late marriage is common compare to rural population. Secondly study population is a closed community (Railway employees and their beneficiaries) with good literacy rate, they are well aware of preventive aspect like HPV vaccination among younger generations that may be the one of the reason for comparatively low positivity rate across all age group compare to other studies.

Conclusion

This present study of age stratified high risk HPV testing reveals that Younger age group < 30 years has the highest positivity rate (3.37%) and age group 41-50 years group has the second highest positivity (3.21%), and moderate positivity between 31 to 40 years group (2.33%) and 51-60 years group with (2.77) Age is a

significant determinant in the utility of high risk HPV DNA testing. This study guide proper utilization of resources and the necessity of routine HPV DNA screening in women aged not only above 30 years, but also in less than 30 years, to detect carcinoma cervix early in dysplasia /CIN stage only to treat the condition effectively. For the younger group < 30 years primary HPV DNA testing has a vital role in early detection of vulnerable group along with HPV vaccination should be the first priority to prevent HPV infection thereby aiming at elimination of cervical cancer as per the WHO global Strategy.

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Conflicts of Interest: None declared.

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Ethical Approval: This study was conducted as an observational cross section study and approved by the institutional ethical committee (Review Board).

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