Epidemiological Analysis of Human Papilloma Virus Infection in Inflammatory Smears

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Summary

Infections of the lower reproductive tract (RTI) are common in Indian women of reproductive age. This communication focuses on genital HPV infections in inflammatory smears diagnosed cytologically and to highlight its association with sociodemographic and clinical factors. Of the total 1,19,935 women screened, 89,314 (74.5%) women had inflammatory smears, of which 587 (6.6 per 1000) had HPV infection. Frequency of HPV infection (per 1000 women) was observed to be higher in the age group of 20-29 years as compared to other age groups (6.8 vs. 6.4), in illiterate and less educated women as compared to that in high school and above educated (7.1 vs. 5.0) (p<0.05), women with bleeding symptoms as compared to all other symptoms (12.9 vs. 6.2). A similar trend was observed with the increase in parity (p<0.05). The women with clinical diagnosis of unhealthy cervix and cervical erosion bleeding on touch had higher infections as compared to that in women with normal looking cervix (9.5 vs. 6.2) (P<0.05). The women revealing HPV infection need counselling for genital hygiene, guidelines on safe sex practices and use of barrier contraception.

Introduction

Human papillomavirus (HPV) infection of the lower female genital tract is recognised as the most frequent sexually transmitted diseases (STD) of today (Reid et al, 1987). It is most often found in sexually active women. The prevalence of this infection has been reported to have dramatically increased during past two decades. The subclinical HPV infection can be diagnosed through the cytology in the cervical smears, which are being collected for detection of precancerous/ early cancerous lesions of uterine cervical cancer. Convincing evidence has been published during past few years implicating the etiologic role of human papillomavirus in cervical intra epithelial neoplasia (CIN) and in cervical cancer (Das et al, 1992).

Assessing the factors influencing the risk of a woman to acquire HPV infection in various groups is of prime importance because of the suggested etiologic role of atleast certain HPV types in cervical carcinogenesis and for devising prevention and treatment strategies. The present communication reports the presence of Human papillomavirus infection in the inflammatory cervical smears determined by cytology and attempts to highlight its association with various sociodemographic and clinical factors.

Material and Methods

The cervical smears were collected from squamo-columnar junction of cervix from 1,19,935 married women in the age range of 20-60 years attending the gynae OPDs of 8 collaborating hospitals of Delhi during 1988-96. Clinical history and a brief information on socio-demographic particulars were also collected on a pretested proforma at this visit. The findings of the clinical symptoms and per speculum examination were recorded by the gynaecologists. Women having symptoms with similar frequency of HPV were grouped into 4 categories: a) bleeding symptoms consisted of blood stained discharge, contact bleeding and intermenstrual bleeding, b) vaginal discharge with curdy and foul smelling, c) dyspareunia and d) other symptoms consisted of pain in abdomen, backache, scanty

periods and menstrual irregularities. All the cytological smears were read at the Institute and diagnosis were made according to the WHO criterial (Riotton et al, 1973). As a part of screening, the HPV induced cytopathological changes are being recorded routinely and these changes (Koilocytotic) diagnosed by primary screeners were confirmed by the cytopathologists. Although molecular hybridization especially by PCR is a better technique for diagnosis of latent and subclinical HPV infections, it could not be done in the present study on all the inflammatory smears due to large number of women being screened and was limited to precancerous and cancerous cases along with matched controls.

Statistical Analysis

The data was analysed on PC/AT and the results were expressed as frequency of HPV infection per 1000 women screened. The data was further analysed according to age, parity, educational status of women, symptoms and clinical diagnosis. In the absence of any accepted single parameter for representing the socioeconomic status, educational status of women was considered to represent the same. The Chi square test of significance was used to test for differences in the HPV infection among various groups. Trend chi square was also estimated where applicable. Odds ratios and 95% confidence intervals were computed.

Results

The cytological diagnosis of 1,19,935 women with adequate smears revealed that 29414 (24.5%), 89,314 (74.5%), 815 (0.7%) and 392 (0.3%) were normal, inflammation, dysplasia of various grades and malignancy respectively. It was observed that of the 89, 314 women with inflammatory smears 587 (0.66%) revealed HPV infection. The other infections which were detected in the inflammatory smears were candida (0.58%), herpes simplex virus (0.05%), trichomonas vaginalis (4.68%), chlamydia (0.04%), tuberculosis (0.03%) and miscellaneous infections (0.87%) such as garderenella vaginalis, parasitic, actinomycosis etc.

The frequency of HPV infection was found to be highest in 20-29 years (6.8 per thousand women) as compared to other age groups (Table I). The frequency and the risk of infection further declined with the increasing age. However, the differences in the frequency of infection amongst different age groups was not statistically significant (p>0.05). The mean age in years of all women examined with inflammatory smears and of women with HPV infection were found to be almost same being 34.6 (SD=9.5) and 34.7 (SD=9.4) respectively. HPV infection revealed an increasing trend with increase in parity status of women (P=0.002, X^2_{trend}=9.4). Frequency of infection with 1-2 parity was 6.0 per thousand women screened while for women with more than 5 parity, it was found to be 8.7 per thousand women. The differences in the frequency of infection among various parity groups were statistically significant (p=0.008). The prevalence odds ratio (OR) was found to be 1.6 (95% CI: 1, 0-2.5) for women with 5 or more parity as compared to nulliparous women. Women with education level of high school and above had lower frequency of infections (5.0 per thousand) compared to women who were illiterates and less educated (7.1 per thousand). The differences in the frequency of infection with regard to literacy was statistically significant (p=0.002). Illiterate women had a risk of 1.4 fold higher than women with education upto high school & above. It was observed that infections were high among women with bleeding symptoms (12.9 per thousand) as compared to women with other symptoms (6.0 per thousand). The corresponding odds ratio was 2.2 (95% CI:1.8-2.6) (Table II). Analysis of data with regard to various clinical diagnosis (Table III) revealed that the frequency of infection amongst women of category II (9.5 per thousand, OR = 1.54) was observed to be significantly higher (p=0.002), compared to women with normal cervix (6.2 per thousand, OR=1.0). The risk of infection showed an increasing trend with the various clinical diagnosis (X^2_{trend}=6.45, p=0.01).

Table I: Frequency of Human Papilloma Virus (HPV) infection by Socio-Demographic factors

Factors	No. of women Screened	Women with HPV	Freq. Of HPV Iinfection (per 1000)	O.R	95% C.I.
Age (Yrs)					
20-29	33178	226	6.8	1.1	0.8-1.5
30-39	32556	213	6.5	1.2	0.8-1.5
40-49	15981	101	6.3	1.1	0.7-1.5
50~	7599	47	6.2	1.0	
			X^2_{trend} 0.62	X 0.63	
				P=0.89	
			P=0.43		
Parity					
Nulliparous	5164	28	5.4	1.0	
1-2	34566	208	6.0	1.1	0.7-1.7
3-4	36093	234	6.5	1.2	0.8-1.8
5+	13491	117	8.7	1.6	1.0-2.5
			X^2_{trend} = 9.3	X - 11.83	
				P=0.008*	
			P=0.002*		
Education					
Low nil	67512	477	7.1	1.4	1.1-1.7
High school & Above	21802	110	5.0	1.0	
				X^2 9.99	
				P 0.002*	
Total	89314	587	6.6		

* Significant

Table II: Frequency of HPV infection per 1000 women screened with different clinical symptoms

Clinical symptoms	No. of women Screened	Freq. Of HPV I infection with HPV	O.R.		95%CI
Bleeding conditions«	5041	65	12.9	2.2	1.8-2.6
Curdy Foul smelling Discharge	3041	26	8.6	1.4	0.9-2.2
Dysparunia	3457	28	8.1	1.4	0.9-2.0
Other symptoms¥	77775	468	6.0	1.0	
		X^2_{trend} = 36.04		X 37.58	
				P 0.001*	
		P 0.001*			
Total	89314	587	6.6		

« Includes blood stained discharge, contact bleeding and irregular vaginal bleeding.

¥ Includes whitish discharge, pain in lower abdomen and other nonspecific symptoms.

+ Significant

Table III: Frequency of HPV infection per 1000 women screeened by clinical diagnosis

Clinical diagnosis	No. of women with screened	No. of women with HPV	Freq. Of HPV Infection	O.R	95%CI
Normal Looking Cervix	25671	158	6.2	1.0	
Category I	54764	345	6.3	1.03	0.9-1.3
Category II	8879	84	9.5	1.54	1.2-2.0
		$X^2_{trend} = 6.45$		$X^2 = 12.65$	
			P=0.002*		
		P=0.01*			
Total	89314	587	6.6		

Category I includes cervical erosion, cervical polyp and cervicitis

Category II includes cervical erosion which bleeds on touch, hypertrophy of cervix, unhealthy cervix and dysfunctional

* Significant

Discussion

Human papillomavirus, a sexually transmitted agent is increasingly implicated in the pathogenesis of cervical cancer (Das et al, 1992). Limited information is available on the incidence and prevalence of condyloma accuminatum. The prevalence rate is dependent on the detection method applied i.e. whether by Papanicolaou smear, colposcopy, punch biopsy, molecular hybridization or polymerase chain reaction. In the studies based on routinely screened smears of women attending a gynaecological department or out-patient clinic of De Villiers et al (1987), the frequency of HPV infection has been reported to be in the range from 4.1 to 22.4 per thousand women screened (Chakrabarti et al, 1987). The results from mass screening investigations reveal a variation of nearly the same magnitude, the prevalence of infection detected cytologically has been reported to range from 7.0 (de Brux & Orth, 1983) to 27.0 per thousand women screened (Meisels & Morin, 1986). The variability may be due to true differences between populations (eg. social background, sexual habits etc.) but it may also be caused by difficulties in diagnosing viral lesions, particularly co-existing with CIN (Bosch & Munoz, 1989). The present study findings reveal that amongst the women with inflammatory smears who attended the gynaecology out patient department for various gynaecological complaints and contraception counselling the frequency of HPV infection was found to be 6.6 per thousand women screened. The criterion for diagnosis of HPV was based on cytology ie. based on the detection of koilocytotic and dysharyocytes. Diagnosis of HPV infection based on cytological criteria alone underestimates the number of women with HPV infection (Hirschowitz et al, 1992). The detection of HPV through Pap smears seems to be the only feasible method applicable to large scale mass screening programmes.

When age-specific infection rates have been studied, it has generally been found that the rate peaks in the age group 20-25 years. The prevalence of HPV DNA by age groups was extensively discussed and there was a consensus that, among women with normal cytology, the peak detection rate was in the young age groups (around 20 years of age) and decreased with age to a fairly constant level (below 10%) after the ages of 45-50 years. This observation was independent of the hybridization method used, and a similar pattern were observed for all HPV types (Bosch, 1992). In the present study women in 20-29 years revealed slightly higher frequency of HPV infection as compared to women in other age groups. However the differences observed were not found to be statistically significant (p>0.05). The reason for not observing change in the infection rates among different age groups may be due to inaccuracies in the age reported by women. Women with educational level of high school certificate & above showed comparatively low frequency of infection of HPV (5.0 per 1000) compared to women with low educational status (7.1 per 1000). This could be attributed to the fact that the educated women probably maintain better genital hygiene practices as compared to the illiterates (Murthy et al, 1990). It has been shown that poor

genital hygiene facilitates transmission of STD's (Wasserheit & Holmes, 1992). In the present study increasing frequency of HPV infection was observed with increasing parity. The frequency of infection among nulliparous women was 5.4 per 1000 as compared to 8.7 per 1000 among women with more than 5 parity. Multiple pregnancies have been found to elevate the risk of HPV infection (Kjaer et al, 1990).

Increasing number of reports also indicates that HPV infections should share many of the risk factors of cervical cancer if the virus is a true causal agent of cervical cancer. It has been reported that the risk factors for genital HPV infections seem very similar, if not identical to those described as risk factors for CIN, CIS and invasive cancer (Villa & Franco, 1989). In the present study increased frequency of HPV infection was observed among women who had signs and symptoms consistent with increased risk for cervical cancer (bleeding symptoms, unhealthy cervix). A prospective study carried out over a period of 6 years on women with cytological evidence of HPV infection in Pap smears revealed a 15.6 fold higher risk for the development of CIN III when compared to the general population (Mitchell et al, 1986).

One of the major limitations of this study is that the diagnosis of HPV infection was on the basis of the presence or absence of HPV induced changes in cervical smears and not on the basis of HPV DNA detection. However, this is likely to be independent of the risk factors studied. This kind of misclassification may reduce the prevalence rate of infection. Secondly, our cases represent the prevalent rather than incident cases. Thus some of the cases may represent persistent or recurrent infections rather than first infection.

Our observation reveals a need for proper counselling and education on practices related to genital hygiene besides treatment to control and prevent the above problem in the community.

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