

Original Research Article

Received : 05/09/2023 Received in revised form : 01/10/2023 Accepted : 11/10/2023

Keywords: Human papilloma virus, ELISA, Seroprevalence, Reproductive tract infections.

Corresponding Author: Dr. Aiswarya R, Email: draiswaryapkd@gmail.com

DOI: 10.47009/jamp.2023.5.5.206

Source of Support: Nil, Conflict of Interest: None declared

Int J Acad Med Pharm 2023; 5 (5); 1052-1059



SEROPREVALENCE OF HUMAN PAPILLOMA VIRUS (HPV) 16 AMONG WOMEN WITH SYMPTOMS OF REPRODUCTIVE TRACT INFECTION

Aiswarya R¹, Sreelatha S², Megha Jayaprakash³

¹Junior Resident, Department of Microbiology, Govt Medical College, Thrissur, Kerala, India. ²Associate Professor, Department of Microbiology, Govt Medical College, Idukki, Kerala, India. ³Additional Professor, Department of Obstetrics and Gynaecology, Govt Medical College, Thrissur, Kerala, India.

Abstract

Background: Human Papilloma Viruses (HPV) are major etiological agents associated with the development of cervical cancer. HPV 16 is the most carcinogenic among the 13 high-risk types of HPV considered to cause cervical cancer. Estimation of IgG antibodies against HPV16 can be used for large scale screening of women with reproductive tract symptoms. Materials and Methods: Hospital based cross sectional study was conducted among 200 women in the age group of 25-65 years for a period of one year. After collection of blood by venepuncture, separation of serum was carried out and IgG antibody to HPV16 was detected by ELISA. Result: The seroprevalence of HPV in this study was 2.5%. The higher prevalence was detected among the patients in the age group 26-35 years. 100 % of IgG positive patients belonged to below poverty line and were house wives. All the five seropositive patients had their first child birth between 20-25 years. HPV seropositivity was associated with diabetes mellitus, family history of cervical cancer and use of contraceptives like barrier contraceptives, intra uterine contraceptive device (IUCD), oral contraceptive device (OCP) and injectable contraceptives. Conclusion: Being a vaccine preventable cancer awareness should be created among people regarding the importance of HPV vaccination. Screening for cervical cancer should be encouraged in all women presenting with symptoms of reproductive tract infection in the hospital settings.

INTRODUCTION

Cervical cancer is the second most common cancer worldwide among women and leading cause of cancer mortality among them in developing countries. Globally every year 6,04,127 new cases are diagnosed and 3,41,831 patients succumb to the disease. A quarter of the global burden is experienced in India, where about 1,23,907 new cases and 77,348 deaths attributable to cervical cancer are estimated to occur each year.^[1] About 99% of cervical cancers are linked to infection with high-risk human Papilloma Virus (HPV). HPV can be grouped as low risk or high risk based on their epidemiological association with cancer. Genotypes belonging to high-risk groups are HPV -16, 18, 31, 33, 35, 39, 45, 51, 52, 54, 56, 58, 59 and 66 and those included under low-risk groups include HPV-6, 11, 34, 40, 42, 43 and 44.^[2] High risk types are associated with low grade and high grade cervical intraepithelial lesion and cervical cancer. Low risk types are associated with genital warts or condyloma acuminatum and recurrent respiratory papillomatosis.^[3]

Among the high-risk types HPV 16 is the most carcinogenic. HPV 16 has greater ability to escape immunosurveillance compared with other HPV types. Infection with HPV 16 has the highest tendency to persist and the highest probability of progression. At the same time HPV 16 has the lowest probability of self-healing.^[4]

Different screening methods available for early detection of cervical precancers and cancers are Pap smear cytology, visual inspection on acetic acid (VIA) and HPV DNA test.^[5] Cervical cytology is the most common screening method used in developed countries. Requirement of laboratory infrastructure, trained cytologist and considerable financial inputs makes cytology based cervical cancer-based screening program difficult in developing countries like India. VIA is one of the alternative screening methods widely used in India. Though it is inexpensive, it is not reproducible for the identification of precancerous lesions and it is less accurate. HPV DNA test has higher sensitivity and specificity, but its high cost makes it impracticable to be implemented in low-income countries.

Among the different HPV proteins expressed during the various phases of the virus life cycle, L1 major capsid protein is considered as a marker of cumulative exposure to HPV infection. Serological studies have demonstrated that majority of women infected with HPV 16 produce an IgG antibody response by 18 months.^[6] IgG antibodies against HPV capsid antigens (L1) are long lasting and hence a marker for past and persistent infection.^[7] Seroprevalence of IgG antibodies against L1 protein can be detected by Enzyme Linked Immunosorbent Assay (ELISA). This can be used for large scale screening of women with reproductive tract symptoms which is cost effective in developing countries like India where limited studies are available on the seroprevalence of HPV.^[8] The study was done to determine the prevalence of IgG antibody to HPV 16 among women with symptoms of reproductive tract infections and to assess the risk factors associated with HPV16 infection.

MATERIALS AND METHODS

Hospital based cross sectional study was conducted at Department of Microbiology Government Medical College, Thrissur for a period of one year from July 2021 to June 2022 after getting ethical clearance. Our study population included 200 patients between 25 -65 years of age who attended the Gynaecology OPD with reproductive tract symptoms such as abnormal uterine bleeding, post coital bleeding, intermenstrual bleeding, lower abdominal pain and persistent vaginal discharge. Patients with history of vaccination against HPV, pre malignant lesions of cervix, carcinoma cervix and pregnancy were excluded from the study. Sample size was calculated based on a study using the formula n=4xpq/d2 where p is prevalence (32%), q = 100-p (68%) and d = 20%of p (6.4) So, n= 4 x 32 x 68 / 40.96=200.^[8]

Specimen collection: 3ml intravenous blood sample was collected in a red top vacutainer by direct venepuncture from median cubital vein under aseptic precaution. The samples were labelled and transported to Microbiology laboratory without any delay. Blood samples were allowed to clot for 10-20 minutes at room temperature. Samples were centrifuged at 2000-3000 rpm for 20 minutes to separate the serum. The supernatant was collected without sediment. Serum samples were stored at -20°C until the tests were performed.

Detection of IgG antibody to HPV: IgG antibody to HPV16 was detected by Enzyme linked immunosorbent assay (ELISA). Shanghai Korain (BT Biotech Co.Ltd. Bioassay) Human papillomavirus type 16 L1-capsids (HPV16L1) antibody (IgG) ELISA kit was used. [9] This kit was used for the qualitative detection of Human papillomavirus type 16 L1-capsids IgG antibody in serum. Test was carried out according to manufacturer's instruction.

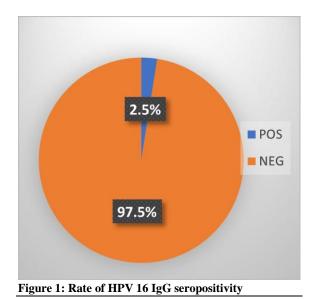
Assay Procedure: All reagents were brought to room temperature and mixed thoroughly before use. Wash buffer was diluted 25 times and strips containing 96 wells were inserted in microtiter plate. A blank well, 2 positive control wells and 2 negative control wells were set. 50µl of negative control was added to each of the negative control wells and 50µl of positive control to each of the positive control wells. 40µl of sample diluent was added to all sample wells followed by addition of 10µl of sample. Microtiter plate was covered with a plate sealer and incubated for 30 minutes at 37°C. After 30 minutes of incubation plate was washed with wash buffer 5 times using automated washer. Then the plate was blotted onto absorbent paper to remove all residual wash buffer. 50µl of HRP (Horseradish peroxidase) were added to each well (except blank well) and further incubated for 30 minutes at 37°C. Again, the plate was washed 5 times with wash buffer using automated washer. The plate was then blotted onto absorbent paper. 50ul substrate solution A followed by 50µl substrate solution B were added to each well and mixed well using the pipette. The plate was further incubated for 10 minutes at 37°C in the dark. The colour of the well would be changed to blue. Then 50µl of stop solution was added to each well. The colour of the plate changed from blue to yellow in positive control well and sample well containing antibody against the L1 capsid antigen. The optical density (OD value) of each well was determined immediately using a microplate reader set to 450 nm. Readings were taken within 15 minutes after adding the stop solution.

Cut off value was Calculated: The average optical density value of the negative control wells plus 0.15 was taken as the cut off value. Any sample with an optical density value less than the calculated cut off value was interpreted as negative and sample with optical density greater than the cut off value was reported as positive.

Statistical Analysis: The data was entered into MS Excel and was analysed using Statistical Package for Social sciences (IBM SPSS) version 25.0. Categorical variables were analysed as proportions or percentages. Chi square test was used to analyse the relationship between study variables and HPV infection. The p value <0.05 was considered as statistically significant.

RESULTS

200 specimens from patients between 25 - 65 years of age with reproductive tract symptoms were included in the study. IgG positivity was detected in 5 patients (2.5%) of study participants. [Figure 1] In our study although maximum participants were from the age group 46-55 and minimum participation from the age group 26-35, higher seroprevalence (5.26%) was detected in the age group 26-35. 163 patients were below poverty line with a seropositivity of 3.07%. All 5 seropositive patients belonged to below poverty line. Out of the 5 seropositive patients two of them had secondary school education and three of them had higher secondary education. Seropositivity was 2.62% among house wives and no seropositive patient was there among working women. Association of the sociodemographic risk factors likeage, income status, educational status and occupational status with HPV 16 IgG positivity were analysed.and p value was found to be >0.05 [Table 1].



In our study although maximum participants were from the age group 46-55 and minimum participation from the age group 26-35, higher seroprevalence (5.26%) was detected in the age group 26-35. 163 patients were below poverty line with a seropositivity of 3.07%. All 5 seropositive patients belonged to below poverty line. Out of the 5 seropositive patients two of them had secondary school education and three of them had higher secondary education. Seropositivity was 2.62% among house wives and no seropositive patient was there among working women. Association of the sociodemographic risk factors likeage, income status, educational status and occupational status with HPV 16 IgG positivity were analysed.and p value was found to be >0.05 [Table 1].

Among the study participants, 85.5% patients were living with their partners and 14.5% patients were either divorced or widows. 4 patients who were tested positive for HPV IgG antibody lived with their partners and one of them was a widow.68% of the patients were married between 18-22 years and only 1% after 32 years of age. Three of the seropositive patients were married between the age of 18-22.56.5% of patients had their first child between 20-25 years, and only 1% patients had first child after 32 years. All the 5 seropositive patients had their first child birth between 20-25 years. In the present study 70% of patients had parity P2 and only 6.5% had parity greater than P3. Out of seropositive patients 3 were having parity P2 and 2 were with parity P3. The marital status, age at which marriage occurred, age at first coitus, age at first child birth and parity were assessed with the p value >0.05 [Table 2]

Out of 200 patients 44 patients were diabetic. Considering diabetes mellitus as a risk factor, patients who were diabetic hadmore chance of getting HPV infection compared to patients who were not diabetic and p value was found to be <0.05 [Table 3]. In our study 5% of patients had used barrier contraceptives. And none among themwere seropositiveand p value was >0.05 [Table 4].

Out of the 200 patients in our study 6.5% of patients had used IUCD. IgG positivity could not be detected among them and p value was >0.05 [Table 5].

In our study 1% of patients had used oral contraceptive pills (OCP) and 2 were seropositive. P value was found to be <0.05 [Table 6].

Out of the 200 patients in our study 1.5% of patients had used injectable contraceptives and 1 among them was IgG positive also with the p value <0.05 [Table 7]

Out of 200 patients, 7 (3.5%) patients had family history of cervical cancer. 4 patients (80%) who were tested positive for IgG antibody had family history of cervical cancer. Considering family history of cervical cancer as a risk factor, patients with the family history had more chance of getting HPV infection compared to women without any family history of cervical cancer and p value was found to be <0.05 [Table 8].

Among the study population 13.5% patients had abnormal vaginal discharge and 51% patients had abnormal uterine bleeding.17.5% patients presented with lower abdominal pain and 30% patients had postmenopausal bleeding. Out of 200 partcipants only 0.5% had post coital bleeding. Out of the 5 patients tested positive, three of them had abnormal uterine bleeding, one of them had post-menopausal bleeding with p value >0.05. Onepatient who had post coital bleeding was IgG positive also and the p value was <0.05. [Table 9]

 Table 1: Distribution and HPV seroprevalence according to Sociodemographic risk factors and their statistical association

`Age group (Years)		%	IgG	+ positivity			χ2 Value	p Value
	n		Pos	itive	Negat	tive		
			n	%	n	%		
26-35	19	9.5	1	5.26	18	94.74	3.07	0.382
36-45	57	28.5	2	3.51	55	96.49		
46-55	72	36	0	0.00	72	100.00		
56-65	52	26	2	3.85	50	96.15		
Income status		%	IgG	positivity			χ2 Value	p Value
	n		Posi	itive	Negati	ive		

			n	%	n	%			
Below poverty line	163	81.5	5	3.07	158	96.93	1.16	0.281	
Above poverty line	37	18.5	0	0.00	37	100.00			
Educational status		%	IgG	positivity		•	χ2 Value	p Value	
	n		Pos	itive	Negati	ve			
		7	n	%	n	%			
Primary school	13	6.5	0	0.00	13	100.00	2.14	0.543	
Secondary school	35	17.5	2	5.71	33	94.29			
Higher secondary	143	71.5	3	2.10	140	97.90			
Degree	9	4.5	0	0.00	9	100.00			
Occupation status		%	IgG	positivity			χ2 Value	p Value	
	n		Pos	itive	Negati	ve		-	
			n	%	n	%			
House Wife	191	95.5	5	2.62	186	97.38	0.24	0.623	
Working	9	4.5	0	0.00	9	100.00			

Table 2: Distribution and seroprevalence of HPV according to marital and reproductive factors and their statistical association

Marital status		%	Ig(5 positivi	ity	χ2 Value	p Value	
	n		Pos	sitive	Nega	tive		
			n	%	n	%		
Living with partner	171	85.5	4	2.34	167	97.66	0.13	0.724
Divorced / widow	29	14.5	1	3.45	28	96.55		
Age at marriage (Years)		%	IgC	b positivity	y		χ2 Value	p Value
	n		Pos	Positive		ive		-
			n	%	n	%		
<18	11	5.5	0	0.00	11	100.00	1.91	0.753
18-22	136	68	3	2.21	133	97.79		
23-27	38	19	2	5.26	36	94.74		
28-32	13	6.5	0	0.00	13	100.00		
>32	2	1	0	0.00	2	100.00		
Age at first coitus (Years)		%	IgC	b positivity			χ2 Value	p Value
	n		Pos	sitive	Negat	ive		
			n	%	n	%		
<18	11	5.5	0	0.00	11	100.00	1.91	0.753
18-22	136	168	3	2.21	133	97.79		
23-27	38	19	2	5.26	36	94.74		
28-32	13	6.5	0	0.00	13	100.00		
>32	2	1	0	0.00	2	100.00		
Age at first child birth (Years)		%	IgC	b positivity	у		χ2 Value	p Value
	n		Pos	sitive	Negat	ive		
			n	%	n	%		
<20	51	25.5	0	0.00	51	100.00	3.95	0.267
20-25	113	56.5	5	4.42	108	95.58		
26-31	34	17	0	0.00	34	100.00		
>32	2	1	0	0.00	2	100.00		
Parity	% IgG positivity			χ2 Value	p Value			
-	n		Pos	Positive		ive		-
			n	%	n	%		
P1	20	10	0	0.00	20	100.00		0.31
P2	140	70	3	2.14	137	97.86		
P3	27	13.5	2	7.41	25	92.59	3.59	
>P3	14	6.5	0	0.00	13	100.00	7	

Table 3: Distribuiton and seroprevalence of HPV based on diabetes mellitus and their statistical association								
Diabetes mellitus		%	IgG	positivity		χ2 Value	p Value	
	n		Posi	tive	Negativ	e		
			n	%	n	%		
No	156	78	2	1.28	154	98.72	4.32	0.038
Yes	44	22	3	6.82	41	93.18		

Table 4: Distribution and seroprevalence of HPV according to usage of barrier contraceptive and their statistical association

Barrier contraception	n	%	IgG	positivity			χ2 Value	p Value
			Posi	tive	Negativ	e		
			n	%	n	%		
Yes	10	5	0	0	10	100	0.27	0.603
No	190	95	5	2.63	185	97.37		

Table 5: Distribution and seroprevalence of HPV according to usage of IUCD and their statistical association								
IUCD	n	%	IgG J	positivity			χ2 Value	p Value
			Positive Negative				1	
			n	%	n	%		
Yes	13	6.5	0	0	13	100	0.36	0.550
No	187	93.5	5	2.67	182	97.33		

Table 6: D OCP	istribution a	nd seropr %	1	evalence of HPV according to usage of OCP and their statistical association IgG positivity χ2 Value p Value								
			Positi	ive	Negative		· · ·	•				
			n	%	n	%						
Yes	2	1	2	100	0	0	78.79	< 0.05				
No	198	99	3	1.52	195	98.48						

Table 7: Distribution and seroprevalence of HPV according to usage of injectable contraceptives and their statistical association

Injectable contraceptive	n	%	IgG positivity				χ2 Value	p Value
			Positive		Negative			
			n	%	n	%		
Yes	3	1.5	1	33.33	2	66.67	11.88	< 0.05
No	197	98.5	4	2.03	193	97.97		

 Table 8: Distribution of family history of cervical cancer among study participants seroprevalence of HPV cancer and the statistical association

Family history of cervical cancer		%	% IgG positivity		y		χ2 Value	p Value
	n		Pos	sitive	Negat	ive		
			n	%	n	%		
Yes	7	3.5	4	57.14	3	42.86	88.86	< 0.05
No	193	96.5	1	0.52	192	99.48		

Discharge PV		%	IgG	positivity		χ2 Value	p Value	
_	n		Pos	itive	Negat	tive		_
			n	%	n	%		
No	173	86.5	5	2.89	168	97.11	0.80	0.849
Yes	27	13.5	0	0.00	27	100.00		
Lower abdominal pain		%	IgG	positivity		-	χ2 Value	p Value
	n		Posi	tive	Negat	ive		
			n	%	n	%		
No	165	82.5	5	3.03	160	96.97	1.09	0.780
Yes	35	17.5	0	0.00	35	100.00		
Abnormal uterine bleeding		%	IgG	positivity			χ2 Value	p Value
	n		Posi	tive	Negat	ive		
			n	%	n	%		
No	98	49	2	2.04	96	97.96	1.96	0.580
Yes	102	51	3	2.94	99	97.06		
Post-menopausal bleeding		%	IgG	positivity			χ2 Value	p Value
	n		Posi	tive	Negat	ive		
			n	%	n	%		
No	140	70	4	2.86	136	97.14	1.97	0.578
Yes	60	30	1	1.65	59	98.3		
Post coital bleeding		%	IgG	positivity			χ2 Value	p Value
	n		Positive		Negative			1
			n	%	n	%		
No	199	99.5	4	2.01	195	97.99	39.20	< 0.05
Yes	1	0.5	1	100.00	0	0.00		1

DISCUSSION

200 patients between 25 - 65 years of age with reproductive tract symptoms who attended the Gynaecology OPD were included in the study. As per the results of our study the seroprevalence of HPV16 was 2.5%. In studies conducted by KP Chacho et al, in Tiruchirappalli and M Aminu from reproductive health clinic of university teaching hospital, Nigeria seroprevalence was found to be higher than the present study.^[10,11] Low seroprevalence in the present study may be due to lower number of participants and antibody against HPV 16 was only studied. Hence caution must be taken in generalising these findings to entire population.

In our study although maximum participants were from the age group 46-55 and minimum participation from the age group 26-35, seroprevalence of HPV was found to be higher in 26-35 age group. In the studiesconducted by Kuruvila P Chacho in Tiruchirapalli and Megan A Clarke in University of Mississipi the prevalence of HPV infection was more among younger age group.^[10,12] The lower seroprevalence in older age group may be due to mature stable transformation zone in cervical epithelium in older people making them less prone to acquire new HPV infection.^[13] In the present study 81.5% of patients were below poverty line and 18.5% were above poverty line and the seroprevalence of HPV was higher (3.07%) in people belonging to below poverty line. In the study conducted by Cherian et al in Trivandrum and Catherine Sauvaget in Maharashtra the prevalence of HPV infection werefound to be higher in low income group.^[14,15] The prevalence of HPV infection was higher among people with only school level education compared to zero prevalence in patients having degree level education. In a study conducted by Cherian Varghese seroprevalence of HPV was found to behigher (7.1%)among illiterate women.^[14] Study done by Catherine Sauvaget et al in Maharashtra showed 73.2% HPV positivity in women who were illiterate.^[15] In the present study prevalence of HPV infection was 2.62% among house wives and IgG positivity was not detected in working women. In studies conducted by Cherian et al in Trivandrum and Catherine Sauvaget et al, in Maharashtra higher prevalence among house wives were seen.^[14,15]

Association of the sociodemographic risk factors with IgG positivity was not found to be statistically significant in the present study. Income status, educational status and occupational status are indicators of socioeconomic status of a person. Higher seroprevalence of HPV was observed in people belonging to low socioeconomic status. Most of them may not be aware about HPV infection, the role it plays in occurrence of carcinoma cervix and its prevention. They also might not be able to afford necessary healthcare or might be living in places with poor access to healthcare facilities, which can lead to vulnerability for the development of disease.

85.5% of study participants were living with their partners and the prevalence of HPV infection was 2.34% among them.14.5% patients were either divorced or widowed and the prevalence was 3.45%. In the study conducted by Franceschi et al, in Dindigul, Tamil Nadu the prevalence of HPV infection was 16.3% among women who were married and 24.3% among women who were widowed similar to our findings.^[16] Widow or separated women are socially vulnerable and economically disadvantaged. There are more chance of them being sexually abused making them susceptible to sexually transmitted diseases. 68% of patients were married between 18-22 years and only 1 patient was married after 32 years. The prevalence of HPV infection was 2.21% in women married at an

age between 18-22 and 5.26% in those married between 23-27. In a study conducted by Aminu in Nigeria 25.7% of women had first sexual intercourse in the age 20-23 and 5.7% had their first sexual debut after 27 years.^[11] In a study conducted by Usha Sarma in Guwahati Medical College, the prevalence of HPV infection was found to be higher in women who married before 16.^[17] A study conducted by Temesgen MM et al in women aged 21 to 49 years in Northern Ethiopia showed almost similar results.^[18] In the present study 56.5% patients hadtheir first child between 20-25 years and only 1% patients had the first child after 32 years. All 5 women who were positive for IgG had their first child between 20-25 years. In the study by Aminu et al most of the women had their first child birth in the age 23-26 and in 4.6% women the first child birth was after 30 almost similar to our study.^[11] 70% patients were of parity P2 and only 6.5% had parity greater than P3 in the present study. The prevalence of HPV infection was more (7.41%) among women with higher parity(P3) compared to 2.14% in women with lower parity (P2). According to the study by Aminu et al about 33.7% of patients had a parity 4-6 and about 3.4% had parity more than 9.^[11] The high parity among these women might be because they had never used any form of contraception. Shikha Srinivastava et al in her study showed a prevalence of 10.3% when the parity was 0 to 1 and it increased to 12.2% when the parity was more than 4.^[19] Higher seroprevalence of HPV on increasing parity may be due to long duration of exposure to risk factors and also due to more cervical injury. In our study number of women with parity more than 3 were less. This may be because people are aware of family planning and the importance of contraceptive usage in controlling the population. No statistical association was found between HPV seroprevalence and marital and reproductive factors in our study.

Out of 200 patients 22% patients were diabetic in the present study. The prevalence of HPV infection was 6.82% in women with diabetic compared to 1.28% in women who were not diabetic. In a study conducted by Yue et al in China it was observed that 9.62% participants were diabetic and the prevalence was 36.69% in diabetic patients compared to 33.14% in those who were not diabetic.^[20] The higher prevalence of HPV infection in diabetics may be due to reactivation of latent infection caused by human papillomavirus in immunosuppressed people. Association between diabetes mellitus and the seropositivity of HPV was statistically highly significant.

As per our present study, 5% patients had used barrier contraceptives, 6.5% intrauterine contraceptive device (IUCD), 1% oral contraceptive pills (OCP) and 1.5% injectable contraceptives. M Aminu et al in her study conducted in Nigeria observedthat 9.4% women were using oral contraceptives, 10.6% using injectable contraceptives.^[11] In our study out of the 5 women who were seropositive for HPV two of them

were using OCP as a method of contraception and one was using injectable contraceptive. The prevalence of HPV infection was higher in women using OCP and injectable contraceptives. Estrogenic exposure causes ectropion of the cervix i.e, cells inside the cervix will be exposed outside and this region will be more susceptible to viral infection. This leads to persistence of HPV infection and its progression to cancer. None of the patients using IUCD and barrier contraceptives were positive for HPV infection. IUCD and barrier contraceptives have protective effect on HPV infection. Cherian et al, in his study conducted in Trivandrum showed a prevalence of 2.9% when barrier contraception was used compared to 6.2% when no contraceptive was used.^[14] Association between OCP usage, injectable contraceptive usage and HPV seroprevalence was found to be statistically significant.

3.5% of our study participants had family history of cervical cancer. Out of the 5 women who were IgG antibody positive for HPV 4 of them had family history of cervical cancer and the prevalence was 57.14%. In the study conducted by Sulaiya Husaiyin among women aged more than 30 years in China, 4.4% of women were having family history of cervical cancer.^[21] In a study conducted by Manga et al among women in Nort-Eastern Nigeria the prevalence of HPV infection was 56% in women who had family history of cervical cancer and 47% who did not have any family history of cervical cancer.^[22] This may be because some women have an inherited tendency of decreased ability to clear HPV infection. Hence this infection become persistent leading to development of cancer.[23]

Among the study population 13.5% patients had abnormal vaginal discharge and 51% patients had abnormal uterine bleeding. 17.5% patients presented with lower abdominal pain, 30% patients with postmenopausal bleeding and only 0.5% patient had post coital bleeding. Higher prevalence of HPV infection was observed in women who were having symptoms like abnormal uterine bleeding, post-menopausal bleeding and post coital bleeding. Out of the 200 participants only one of them had post coital bleeding and she been positive for HPV infection. Aminu et al in her study in Nigeria observed 40% of the study participants had abnormal vaginal discharge, 22.3% and 6% women had complaints of vaginal itching and vaginal rashes respectively.^[11] In a study conducted by Mishra R et al the seroprevalence of HPV was higher (69.6%) in women with symptoms like pain in abdomen, vaginal discharge and irregular menstruation compared to 30.4% prevalence in women who were asymptomatic.^[13] Association between IgG positivity and post coital bleeding was found to be statistically significant .But no statistical association was found between seropositivity and clinical features like discharge PV, lower abdominal pain, abnormal uterine bleeding and post-menopausal bleeding.

CONCLUSION

Screening by cervical cytology, HPV DNA test and VIA are difficult to implement in low income countries like India . IgG antibodies against HPV capsid antigens (L1) are long lasting and hence a marker for past and persistent HPV infection. The advantage of using serological methods like ELISA for screening to detect IgG antibody is that large number of samples can be tested at a single time and is cost effective. Patient compliance will be good in taking blood samples rather than taking cervical samples. Hence serological method like ELISA to detect IgG antibody will be a promising assay for large scale screening of women in low resource settings.

REFERENCES

- Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2021;71(3):209–49.
- Jacobs MV, de Roda Husman AM, Van den Brule AJ, Snijders PJ, Meijer CJ, Walboomers JM. Group-specific differentiation between high-and low-risk human papillomavirus genotypes by general primer-mediated PCR and two cocktails of oligonucleotide probes. J Clin Microbiol. 1995;33(4):901–5.
- Organization WH. Human papillomavirus laboratory manual. World Health Organization; 2010.
- Castle PE, Burk RD, Massad LS, Eltoum IE, Hall CB, Hessol NA, et al. Epidemiological evidence that common HPV types may be common because of their ability to evade immune surveillance: Results from the Women's Interagency HIV study. Int J Cancer. 2020;146(12):3320–8.
- Mustafa RA, Santesso N, Khatib R, Mustafa AA, Wiercioch W, Kehar R, et al. Systematic reviews and meta-analyses of the accuracy of HPV tests, visual inspection with acetic acid, cytology, and colposcopy. Int J Gynecol Obstet. 2016;132(3):259–65.
- Stanley M. Immune responses to human papilloma virus. Indian J Med Res. 2009 Sep 1;130:266–76.
- Ho GY, Studentsov YY, Bierman R, Burk RD. Natural history of human papillomavirus type 16 virus-like particle antibodies in young women. Cancer Epidemiol Biomarkers Prev. 2004;13(1):110–6.
- Verma RR, Sriraman R, Ponnanna NM, Rana SK, Srivastava S, Roy JK, et al. Seroprevalence of Antibodies to HPV L1 in a Limited Population study determined by the GST-capture ELISA. Curr Trends Biotechnol Pharm. 2015;9(2):97–106.
- Tognon M, Tagliapietra A, Magagnoli F, Mazziotta C, Oton-Gonzalez L, Lanzillotti C, et al. Investigation on spontaneous abortion and human papillomavirus infection. Vaccines. 2020;8(3):473.
- Chacho KP, Nagarajan P, Ivan RA, Chandran G. Prevalence of Human papilloma viral infection from women in tertiary care teaching hospital, Tiruchirapalli, India. Int J Curr Microbiol App Sci. 2014;3(7):713–9.
- Aminu M, Gwafan JZ, Inabo HI, Oguntayo AO, Ella EE, Koledade AK. Seroprevalence of human papillomavirus immunoglobulin G antibodies among women presenting at the reproductive health clinic of a university teaching hospital in Nigeria. Int J Womens Health. 2014;6:479.
- Clarke MA, Risley C, Stewart MW, Geisinger KR, Hiser LM, Morgan JC, et al. Age-specific prevalence of human papillomavirus and abnormal cytology at baseline in a diverse statewide prospective cohort of individuals undergoing cervical cancer screening in Mississippi. Cancer Med. 2021;10(23):8641–50.
- 13. Mishra R, Bisht D, Gupta M. Distribution and Prevalence of High-risk Human Papillomavirus Infection in Women of

Western Uttar Pradesh, India: A Hospital-based Study. J South Asian Fed Obstet Gynaecol. 2022;14(2):91–4.

- Varghese C. Prevalence and determinants of human papillomavirus (HPV) infection in Kerala, India. Tampere University Press; 2000.
- Sauvaget C, Nene BM, Jayant K, Kelkar R, Malvi SG, Shastri SS, et al. Prevalence and determinants of high-risk human papillomavirus infection in middle-aged Indian women. Sex Transm Dis. 2011;38(10):902–6.
- Franceschi S, Rajkumar R, Snijders PJF, Arslan A, Mahe C, Plummer M, et al. Papillomavirus infection in rural women in southern India. Br J Cancer. 2005;92(3):601–6.
- Sharma U, Mahanta J, Borkakoty BJ, Talukdar KL, Gogoi R, Yadav K. Demographic Characteristic of HPV infection in women-A hospital based study from Guwahati, India. Natl J Med Res. 2013;3(01):1–4.
- Temesgen MM, Alemu T, Shiferaw B, Legesse S, Zeru T, Haile M, et al. Prevalence of oncogenic human papillomavirus (HPV 16/18) infection, cervical lesions and its associated factors among women aged 21–49 years in Amhara region, Northern Ethiopia. PloS One. 2021;16(3):e0248949.

- Srivastava S, Gupta S, Roy JK. High prevalence of oncogenic HPV-16 in cervical smears of asymptomatic women of eastern Uttar Pradesh, India: a population-based study. J Biosci. 2012;37(1):63–72.
- Yue C, Zhang C, Ying C, Jiang H. Diabetes associated with HPV infection in women aged over 50 years: A crosssectional study from China's largest academic woman's hospital. Front Endocrinol. 2022;13:972963.
- Husaiyin S, Han L, Wang L, Ma C, Ainiwaer Z, Rouzi N, et al. Factors associated with high-risk HPV infection and cervical cancer screening methods among rural Uyghur women aged> 30 years in Xinjiang. BMC Cancer. 2018;18(1):1–9.
- 22. Manga MM, Fowotade A, Abdullahi YM, El-Nafaty AU, Adamu DB, Pindiga HU, et al. Epidemiological patterns of cervical human papillomavirus infection among women presenting for cervical cancer screening in North-Eastern Nigeria. Infect Agent Cancer. 2015;10(1):1–9.
- Beskow AH, Gyllensten UB. Host genetic control of HPV 16 titer in carcinoma in situ of the cervix uteri. Int J Cancer. 2002;101(6):526–31.