Geographical distribution of HPV Genotypes and their association with Socio-demographic and Clinicopathological parameters among Normal, Pre-cancer and Cervical Cancer women in Chennai, India: A Cross-sectional study

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Abstract

Carcinoma of uterine cervix is a major worldwide wellness problem. Chronic infection with high-risk Human papillomavirus (HPV) is a major risk factor for the onset of invasive cervical cancer. The aim of this study is to investigate the HPV genotype distribution and its correlation with the socio demographic and clinic pathological features among healthy, cervical pre-cancer and cancer women in a defined region of Chennai, Tamilnadu, India. 160 cervical specimens consisting of 41 Squamous cell carcinoma(SCC), 4 Adenocarcinoma(AC), 35 precancerous lesions (LSIL/HSIL) and 80 samples from normal women were selected to perform detection and genotyping of high-risk HPV using PCR.

Out of 160 women, 80(50%) had normal Pap smear, 35(21.8%) had Precancerous lesions and 45(28.1%) had invasive cervical carcinoma. Among 80 women with abnormal cytology of cervix, HPV was found in 78 (97.5%) subjects. The predominant strains of HPV were HPV16 (41.02%) and HPV18 (38.4%). Several research investigations conducted in diverse geographical locations across India have demonstrated considerable variations in the frequency of infections caused by HPV and its genotype diversity. The present research provides a recent valuable baseline data regarding HPV distribution in this studied group.

Keywords: Human Papilloma virus (HPV), Cervical cancer, Genotyping.

Introduction

Uterine cervical cancer (CaCU) remains to be the 4th most prevalent malignancy among females worldwide, having about 660,000 new cases documented in 2022. The primary objective of global campaign for the eradication of cervical cancer is to prevent 74 million new cases and 62 million deaths by 2120[11]. The global population comprises of 2,972.8 million women who are 15 years and older that are considered to be susceptible for cervical cancer[4]. Cervical cancer accounts for 6-30% of all malignancies in women[21]. HPV (Human papilloma virus) is a sexually transmitted pathogen, with high-risk strains observed in 99.7% of CA cervix specimens[33]. In 1983, Durst et al[7] first demonstrated the association between HPV and cervical cancer. Unfortunately, in rural India, insufficient hygiene, knowledge, screening and immunisation have led to undetected HPV infections[9].

Human papilloma viruses are classified under A genus of the Papovaviridae family. Their genome consists of a circular double stranded DNA that spans 7000 to 8000 base pairs, encoding eight functional early proteins (E1-E8), 2 structural late proteins (L1 and L2) and a non-coding long terminal region (LTR)[20,23]. The Papilloma virus nomenclature committee categorizes each type into evolutionary lineages according to geographic distribution, pathogenicity, transcriptional regulation and immunological response[24]. The IARC and the World Health Organization (WHO) have classified 229 distinct types of human papillomavirus (HPV) including over 40 types that colonize the genital tract into three categories based on their carcinogenic potential.

High-risk HPV strains, primarily HPV16 and HPV18 from group 1, are significantly linked with onset of several malignancies including cervix of uterus, anogenital (penis, vulva, vagina) and head and neck cancers[25]. Globally, HPV 16 is responsible for more than 50% of squamous cell carcinomas whereas HPV 18 constitutes around 20% of these carcinomas, collectively representing 71% of cervical cancer cases[12]. Sixty different types are linked to mucosal epithelia and categorised into the genus Alphapapilloma virus (alpha-PV), Betapapilloma virus and Gammapapillomavirus[24]. The majority of HPV infections are resolved within few months due to the immune system's

reaction. Approximately 50% of HPV infections cure after 8 months, whereas roughly 90% are eradicated within 2 years.

The prevalence of illness in women reaches its peak at around 40 years of age, but the high incidence of infection by HPVs occurs around 20 years of age, leading to a significant latency interval between infection and the onset of cancer[16]. The frequency of human papilloma virus infection among sexually active females varies from 50% to 80% during their lives, establishing it as one of the most prevalent viral infections worldwide, with an estimated global prevalence of 11.7%[28]. Numerous research studies undertaken in various geographical regions of India have revealed significant disparities in the prevalence of infection by HPV and distribution of its genotype. Nonetheless, there is an absence of comprehensive national data about human papilloma virus infection and its type distribution that might facilitate the implementation of screening strategies and vaccination initiatives[26].

As of 2023, the worldwide market provides six preventative HPV vaccines: Cervarix, Cecolin, Walrinvax, Gardasil, Cervavax and Gardasil9. All of these vaccinations confer protection against the High-risk strains of HPV16 and 18(16). Newly established "HPV FASTER" guideline recommends administering HPV vaccine to women between the ages of 9 and 45, irrespective of their HPV status[5]. The worldwide strategy proposes for at least two lifetime screenings, including an exceptionally well HPV diagnosis at age 35 and again at age 45[12]. The implementation of sustainable and effective screening and vaccination programs, under governmental oversight, represents the most efficient strategy for reducing the prevalence and mortality associated with cervical carcinoma in underdeveloped nations[5].

Due to complex regional cultural disparities across different States of India, it is essential to delineate the geographical distribution of genotypes of HPV in cervix cancer patients and sample population from diverse representative community members prior to generalising these informations on implementation of National Cancer Prevention Strategies[29]. Previous research has demonstrated a significant association between persistent infection with high-risk HPVs and the global distribution patterns of HPV genotypes in the development of cervical cancer. However, data specific to this particular geographical region of Chennai remains limited and consequently this study aims to analyze the distribution of HPV genotypes within a defined population to better understand the prevalence and variations of the virus. Additionally, it sought to examine the correlation between HPV infection and various histological grades and different stages of cervical carcinoma subjects of this population.

Material and Methods

Study design, Setting and Participants: A cross-sectional research was done at the Department of Obstetrics and Gynaecology, SRM Medical College Hospital and Research Centre, Kattankulathur and at Department of Gynaecology and Preventive Oncology, Adyar Cancer Institute (W.I.A), Chennai, from April 2023 to April 2024. This research comprised of 160 subjects: 80 normal women, 35 with precancerous conditions and 45 with invasive malignancy. The control participants consisted of randomly selected healthy women who attended the hospital for routine examinations, with no prior history of cervical cancer or any other gynaecological malignancies, hysterectomy, abnormal cytological results, pregnancy, or past cancer therapy. Women aged 35 to 85 exhibiting symptoms such as leukorrhoea, severe abdominal pain and lower back pain were incorporated into the research.

Women who were unwilling or had treated cases of cervical cancer, as well as patients currently undergoing treatment, were excluded from the research. All enrolled women were provided with comprehensive information on the study's purpose and the research was made clear to those who participated using a participant information sheet (PIS). Informed consent was obtained in writing from all patients and their families. A detailed questionnaire was administered, encompassing sociodemographic data including education, parity status, age at marriage and socioeconomic level. The privacy and confidentiality of the information provided by participants were meticulously preserved during the study.

According to cytopathological and histopathological studies, all patients were classified into LSIL/HSIL and invasive cancer (SCC/AC) correspondingly. The staging of cervical cancer was carried out in accordance with the FIGO classification. The research obtained clearance by the Institutional Ethics Committee of both organizations (IEC No: 2365/IEC/2021 & IEC/2023/Sep10) and carried out in conformity with the principles of ethics outlined in the Helsinki's declaration.

Cervical sample collection and Handling: Pap smear was performed using an Ayre's spatula, rotated 360˚ within the endocervix and rapidly spread onto a glass slide and then placed in a Coplin jar with 95% ethanol. The fixed slides were subsequently transmitted for cytopathological evaluation and grading was conducted utilising the Bethesda system for reporting cervical cytology 2014. The sample for HPV DNA analysis was collected using a cytobrush, thereafter placed in a 0.5microlitre Eppendorf tube which contains 1X phosphate buffer solution and kept at 4˚C until further analysis. Due to the inaccessibility of pap smears in patients with invasive cancer, tissue biopsy samples were collected, placed in a small container with 10% formalin and subsequently dispatched for histopathological analysis.

Throughout the duration of this study, we ensured the preservation of clinical specimens quality, specifically cancer biopsy samples and Pap smears. The samples were collected, transported and stored following standardized

protocols. This approach minimized the risk of sample contamination or deterioration from the point of collection to the execution of the clinical and genotype analysis.

Genomic DNA extraction: DNA was extracted using FAVORGEN (Biotech Corp) FavorPrep^{TM} Tissue Genomic DNA Extraction Mini Kit for smear samples and QIAamp® DNA FFPE Kit (QIAGEN) for formalin fixed and paraffin embedded cancer biopsy specimens adhering to the manufacturer's guidelines. Isolated samples of DNA underwent electrophoresis on 1% agarose gel to assess DNA quality and the amount has been determined using a Nanodrop Spectrophotometer (Nanodrop, ND-1000).

HPV DNA Detection and Genotype: The DNA of HPV in cervical cells was identified by multiplex PCR utilising sequences derived from L1 consensual site of viral genome. Two distinct pairs of oligonucleotide probe sets, MY09/11 and GP5+/6+, were utilised as they identify a wide range of virus strains at subpicogram concentrations. All PCR reaction sets were conducted using positive and negative controls. The adequate quantity and purity of isolated DNA from each source for amplification of PCR were assessed by recognizing the human β globin gene as an internal standard. The PCR was conducted in a total volume of 20µL including 100ng DNA sample, 1X PCR buffer (10mM Tris Cl pH 8.3, 50mM KCl, 1.5mM $MgCl_2$), 5mM of both the forward and reverse primer, 4µL of dNTP mix (that includes 200µM of dATP, dTTP, dCTP and dGTP) and 1U of Taq DNA polymerase.

The HPV-positive samples underwent genotyping and type-specific PCR was conducted using primers specific to HPV 16 and 18. HPV genotypes were identified by analysing the amplified PCR products on 1.5% agarose gel dyed with ethidium bromide, thereafter documented using a gel documentation system. Due to the varying lengths of all amplified products, the viral genotypes were analysed using electrophoresis and visualised using a UV transilluminator. Robust standardization procedures, quality assurance and validation methods were employed at every stage of the investigation to guarantee the accuracy and dependability of the findings.

Statistical Analysis: The results of this study collected through different techniques were uploaded into Microsoft Excel on a personal computer and evaluated manually. The statistical analyses were conducted using SPSS software (version 25) (Chicago, USA) and Excel for Windows 7. The demographic data and clinical characteristics were organised using frequency and percentages. A two-sided p-value was calculated, with $p<0.05$ indicating statistical significance. The correlation between sociodemographic characteristics and clinical measures concerning either the presence or absence of infection with HPV was assessed using the conventional contingency table approach and Fisher's exact test. Logistic regression analysis was used for calculating odds ratio [OR] and 95% confidence interval [CI].

Results

Table 1 depicts the socio-demographic factors of study participants, comprising control, precancerous and cancer subjects. An interquartile range was utilized for the selection of age groups. The average age of subjects included was 58.91 years, with a standard deviation (SD) of 12.3. All women included in this study were married, with parity ranging from 1 to 7. The majority of participants belonged to low socioeconomic status, were illiterate and were married at a young age (under 20 years). Of the 160 women selected for this study, 80 (50%) showed normal Pap smear results, 35 (21.8%) presented with precancerous lesions and 45 (28.1%) were diagnosed with invasive cervical carcinoma. All samples tested positive for β-globin, confirming their suitability for subsequent genotyping. Figure 1 displays the gel images illustrating DNA concentration from control samples and formalin-fixed paraffin-embedded (FFPE) samples.

Among the 80 women exhibiting atypical cytology of uterine cervix, HPV was identified in 78 (97.5%) subjects whereas among women with normal cytology, only 10 (12.5%) were presented with positive infection. The most prevalent HPV genotypes include HPV16 in 32 (36.75%) subjects and HPV18 in 30(34.25%) women, followed by co-infection with HPV16/18 in 10(11.5%) and genotypes other than HPV16 and HPV18 in 6(7.5%) case groups. Figure 2 illustrates the distribution and prevalence of HPV infection among the normal, precancer and cervical cancer groups.

Table 2 displays the correlation between FIGO staging of invasive carcinoma and HPV genotype distribution. This study found that infection rate of HPV16 was elevated (44%) in patients with stage II and III, whereas infection rate by HPV18 was higher (53%) in stage III patients; moreover, HPV infection was lower (2%) in stage I patients. Table 3 represents the prevalence of HPV across various histological grades and different types of cancer tissues. Of the 45 cervical cancer biopsy samples analysed, 41 (91%) were diagnosed as Squamous cell carcinoma (SCC) and 4 (9%) as Adenocarcinoma (AC). Additionally, the distribution of grades was found to be 47% in grade I, 36% in grade II and 18% in grade III. A notable prevalence of HPV was observed in well-differentiated malignancies, although reduced with a decline in differentiation grading.

Table 4 illustrates correlation between sociodemographic factors and prevalence of various HPV genotype distribution among the research participants. Among the 78 women in the case category who were confirmed positive for HPV infection, 16 (21%) were in the age group 35-49 years, 20 (26%) were aged 49-59 years, 21 (27%) were in age group 59-68 years and the remaining patients were over 68 years. The highest positivity rates were observed in women over 49 years of age. Regarding parity, 74 (95%) of the women with more than three children exhibited a high incidence of HPV infection. In terms of literacy, 55 (70.5%) of the illiterate women had a greater incidence of HPV positivity.

Additionally, 72 (92%) of the women who married at an early age (under 20 years) showed a stronger association with HPV infection. Furthermore, 73 (93%) of the subjects residing in rural areas demonstrated increased susceptibility to HPV infection.

Table 1
Various Sociodemographic factors of normal, precancer and cervical cancer women

Variables	No. of Control Samples	No. of Pre-cancer cases (LSIL/HSIL)	OR (95% CI)	P-value	No. of Invasive cancer cases	OR (95% CI)	P-value
Age							
35-49	21(26%)	5(14%)	1 (reference)		12(26%)	1 (reference)	
49-59	17(21%)	8(23%)	1.976 (0.545-7.161)	0.300	12(27%)	1.235 (0.444-3.440)	0.686
59-68	20(25%)	7(20%)	1.470 (0.400-5.398)	0.562	15(33%)	1.312 (0.495-3.481)	0.585
>68	22(27%)	15(43%)	2.864 (0.884-9.278)	0.079	6(13%)	0.477 (0.151-1.504)	0.207
Literacy							
Literate	34(42%)	12(34%)	1 (reference)		11(24%)	1 (reference)	
Illiterate	46(56%)	23(66%)	1.417 (0.620-3.239)	0.409	34(76%)	2.285 (1.015-5.144)	**0.046**
Parity Status							
<3	4(5%)	2(6%)	1 (reference)		2(4%)	1 (reference)	
>3	76(95%)	33(94%)	0.868 (0.152-4.977)	0.874	43(96%)	1.132 (0.199-6.435)	0.889
Age at time of marriage							
>20	10(13%)	4(11%)	1 (reference)		3(7%)	1 (reference)	
<20	70(87%)	31(89%)	1.107 (0.322-3.804)	0.872	42(93%)	2.000 (0.521-7.682)	0.313
Socioeconomic status							
Urban	12(15%)	3(8%)	1 (reference)		2(4%)	1 (reference)	
Rural	68(85%)	32(91%)	1.882 (0.496-7.139)	0.352	43(96%)	3.794 (0.809-17.78)	0.091

LSIL-Low Squamous intraepithelial lesions, HSIL- High Squamous intraepithelial lesions, OR-Odds ratio, CI-Confidence interval

Table 2
Association between FIGO staging of Invasive cancer and HPV Genotype

FIGO staging	Invasive cancer (n=45)	HPV(+ve) (n=44)				HPV(-ve) (n=1)	P-Value
		HPV16 (n=18)	HPV18 (n=17)	HPV16/18 (n=6)	HPV other than 16/18 (n=3)		
I	1(2%)	-	-	-	-	1(100%)	0.266
II	14(31%)	8(44%)	5(29%)	1(17%)	-	-	
III	23(51%)	8(44%)	9(53%)	4(67%)	2(67%)	-	
IV	7(15%)	2(11%)	3(18%)	1(17%)	1(3%)	-	

FIGO-International Federation of Gynecology and Obstetrics, HPV-Human Papilloma virus

Table 3
Association of HPV (Human papillomavirus) distribution with different histological type/grades of cervical cancer tissues

Histological Type	Total (n=45)	HPV(+ve) (n=44)	HPV(-ve) (n=1)	P value
Squamous cell carcinoma (SCC)	41(91%)	40(91%)	1(100%)	0.115
Adenocarcinoma (AC)	4(9%)	4(9%)	-	
Histological Grade				
Grade I (Well differentiated)	8(18%)	7(16%)	1(100%)	0.420
Grade II (Moderate differentiated)	16(36%)	16(36%)	-	
Grade III (Poorly differentiated)	21(47%)	21(48%)	-	

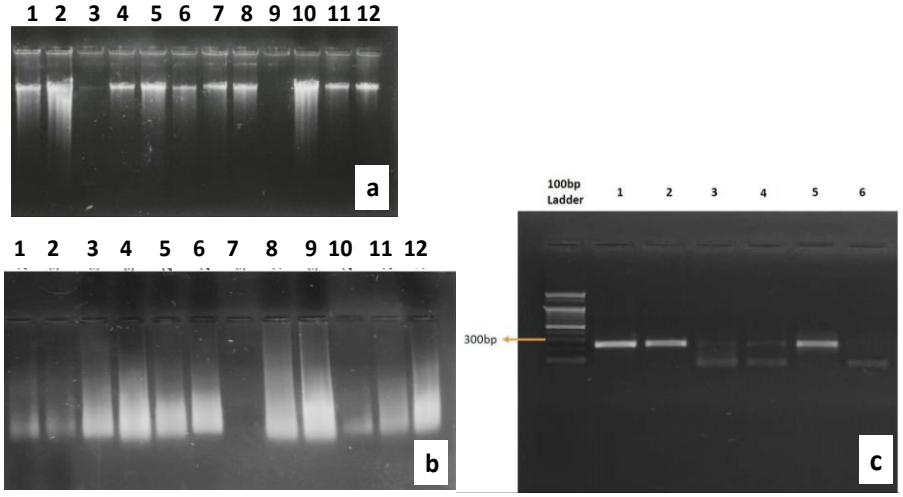

Figure 1: Gel images showing DNA concentration from a) Lane 1-12=Control Pap smaer samples, b) Lane 1-12=FFPE(Formalin fixed paraffin embedded samples, c)Beta globin primer Lane 1 and 2-Control samples, 3 and 4-FFPE samples,Lane 5-Human DNA,Lane 6-Water control

Table 4
Association between Socio demographic characteristics and HPV infection in Precancerous and cancer women

Socio demographic factors	Cases(n=80)		HPV(+ve) (n=78)				HPV (-ve) (n=2)	P-Value
	LSIL/ HSIL (n=35)	Invasive cancer (n=45)	HPV 16 (n=32)	HPV 18 (n=30)	HPV 16/ 18(n=10)	HPV Other than 16/18 (n=6)		
Age Group								0.99
35-49	5(14%)	12(26%)	6(19%)	7(23%)	2(20%)	1(16%)	1(50%)	
49-59	8(23%)	12(27%)	8(25%)	9(30%)	2(20%)	1(16%)	-	
59-68	7(20%)	15(33%)	8(25%)	8(26%)	4(40%)	1(16%)	1(50%)	
>68	15(43%)	6(13%)	10(31%)	6(20%)	2(20%)	3(50%)	-	
Literacy								0.930
- Literate	12(34%)	11(24%)	9(28%)	11(37%)	2(20%)	1(17%)	-	
- Illiterate	23(66%)	34(76%)	23(72%)	19(63%)	8(80%)	5(83%)	2(100%)	
Parity status								1.000
<3	2(6%)	2(4%)	3(9%)	1(3%)	-	-	-	
>3	33(94%)	43(96%)	29(91%)	29(97%)	10(100%)	6(100%)	2(100%)	
Age at time of marriage								0.817
- >20	4(11%)	3(7%)	2(6%)	4(13%)	-	-	1(50%)	
- <20	31(89%)	42(93%)	30(93%)	26(87%)	10(100%)	6(100%)	1(50%)	
Socio-economic status								0.399
- Urban	3(8%)	2(4%)	1(3%)	3(10%)	1(10%)	-	-	
- Rural	32(91%)	43(96%)	31(97%)	27(90%)	9(90%)	6(100%	2(100%)	

LSIL-Low Squamous intraepithelial lesions, HSIL- High Squamous intraepithelial lesions, HPV-Human Papilloma virus.

Discussion

Cervical carcinoma represents a substantial major threat to health worldwide, especially in countries with low or middle incomes (LMICs), whereby the burden is disproportionately high[27]. Geographical variations in the distribution of HPV, particularly HPV 16, may be influenced by several factors including differences in sample sizes, cultural attributes, ethnic diversity and heterogeneity across studies[2,10]. This study is likely to make a meaningful contribution to high-risk HPV screening and to enhance our understanding of disease susceptibility by systematically evaluating related risk factors. The reliability of our findings is reinforced by comprehensive participant recruitment strategies and rigorous statistical analysis.

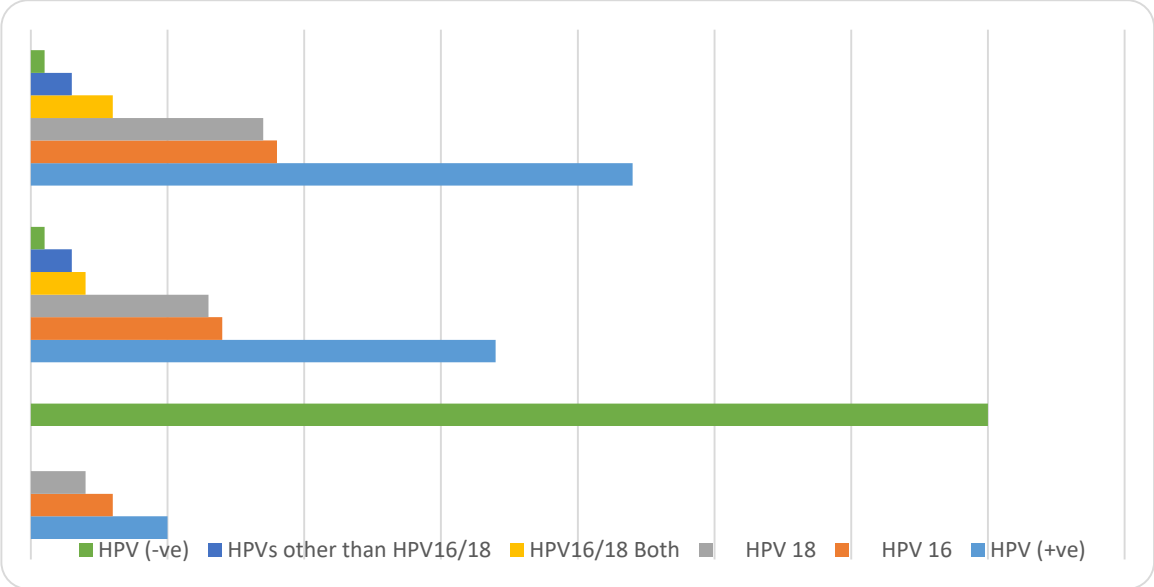

Figure 2: HPV distribution among different groups of studied population

Our study reveals a high-risk HPV genotype prevalence of 97.7% among invasive cancer patients, corresponding to previous research reporting 96.6%[14]and 93.3%[9]. Analysis of many research studies revealed that the most frequently mentioned and discovered genotypes were HPV16 and HPV18. The frequency of HPV 16 is prominent in the current study, consistent with findings from other studies[3,22,30]. The prevalence of SCC in our research is 91.1%, whereas AC is 9%, which aligns with findings from Parekh et al[22] reporting SCC at 91% and AC at 7.2%. Study by Gupta et al[9] observed 97.6% for SCC. Multiple investigations demonstrate that patients may be co-infected with various genotypes of HPV. The current research identified infections with several HPV types in 11.5% women, comparable to the 8.2% reported by Vinodhini et al[32].

A prior research conducted in Chennai established a substantial link among the incidence of cervical cancer and inadequate hygienic circumstances, among various other factors[8]. Our study offers more comprehensive insights into the relative incidence of high-risk infections (HPV 16, 18, 16/18 and other HPV types) in relation to sociodemographic and clinicopathological parameters, compared to other research conducted in this region. This study revealed a significant relationship between illiteracy in women (66%) and the development of cervical cancer ($P<0.046$). Nonetheless, no association was detected between the stage, grade, or histologic classification of illness and age of women about HPV prevalence in this community, similar to the findings of Munagala et al[19] who also reported no correlation with any sociodemographic parameters.

Previous research in South India reveals a significant prevalence of high-risk HPV among cervical carcinoma patients with about 87.8% in Andhra Pradesh according to Sowjanya et al[29] and 99.4% as recorded by Franceschi et al[8] in Chennai. In contrast, a research by Maheshwari et al[17] reported the overall incidence of HPV DNA to be at 40.98%, which is significantly low. Given inadequate awareness, illiteracy and social stigma, many women exhibit reluctance towards routine gynaecological check-ups and for Pap smear examination.

Consequently, late detection and poor screening rates lead to a significant number of cases being discovered at advanced stages, resulting in an increased prevalence of HPV. This elucidates the elevated prevalence (53%) of stage III invasive cancer patients in the population we studied, consistent with Parekh et al[22] (58%).

A significant proportion of patients presented with advanced-stage illness, explaining the increased mortality and disability attributed to cervical cancer in our country relative to global figures. HPV remains a significant component consistently linked to cervical cancer; yet, it is inadequate on its own for the advancement of cervical preneoplastic lesions to invasive cervical cancer[30]. A chronic infection, characterized by the incorporation of the HPV DNA into the chromosomal genetic material of cervical cell epithelium, along with alterations of human cancer genes, impairment of immune mechanisms promotes the development of uterine cervical cancer[32]. HPV16 and 18 were detected in every age group. This may be a unique characteristic of the general population in which HPV infection continues into their middle years, therefore adding to the prevalence of cervical carcinoma. However, women aged 30 years or older with a mature stable transformation zone are less susceptible to acquiring new HPV infections; yet, they may test positive for HPV DNA owing to a long-standing persistent infection that has not been immunologically eradicated[18]. This underscores the necessity of sampling women aged 30 years or younger for HPV testing, especially in a country like India, where the age of marriage and the age of cervical cancer diagnosis are notably lower than in developed countries.

The potential influence of the host's genetic factors seemed more apparent when considering age-specific general HPV prevalence patterns, necessitating an exploration of the natural history and biology of human papillomavirus[3]. A study conducted by Venceslau et al[31] noted variations in HPV DNA detection rates that may be attributed to differences in sample types (e.g. smears, frozen specimens, paraffin-embedded tissues), anatomical localization and oligonucleotide design.

Furthermore, a hospital-based study suggested that HPV self-sampling, which demonstrates a sensitivity of 95%, can help women overcome practical and emotional barriers to screening[15].

This research have both strength as well as limitations. A notable limitation was the relatively restricted sample size belonging to a confined region of the State, which could impact the generalizability of the results and might not fully represent the diversity of Tamilnadu's wider population. Although our investigation offers valuable insights, it is crucial to interpret the findings cautiously due to the constrained sample size and potential regional influences. Our study population's genotype distribution exhibits both similarities and distinct variations when compared to other areas, necessitating additional research and area-specific health initiatives. In contrast to some previous research, our results indicate a reduced occurrence of certain high-risk HPV genotypes and we did not observe a notable presence of specific low-risk HPV genotypes in this area.

The uniqueness of this research lies in its focus on a small, previously understudied region, providing fresh insights into HPV genotype distribution and its potential impact on public wellbeing. The findings in this study suggest several recommendations for cervical cancer prevention and public health policies. The primary focus should be on expanding and prioritizing HPV vaccination programs, with a particular emphasis on reaching adolescent girls before they become sexually active to ensure maximum efficacy. To enhance the economic feasibility of HPV vaccination initiative programs in India, it is crucial to comprehend the regional genotypic prevalence of HPV, which is necessary for directing targeted vaccination strategies and the development of innovative vaccines with broader value[13].

Educational campaigns should be launched within communities to increase understanding of HPV, its connection to cervical cancer and the significance of vaccination and routine screenings[6]. The detection of various HPV strains responsible for illness has led to enhanced diagnostic, screening and preventive strategies within the medical society[1].

Moreover, training for healthcare professionals should be improved to ensure accurate sample collection, result interpretation and patient guidance. Lastly, ongoing monitoring of HPV genotype distribution in this population is vital to track changes over time and modify prevention strategies as needed.

Conclusion

Despite advancements in treatment methodologies, the burden of cervical cancer persists, necessitating further investigation into its causative factors and contributing elements. The varied distribution pattern of HPV genotypes across different geographical regions of India presents an opportunity to update the development of targeted HPV vaccination programs. This research makes a notable contribution to the current knowledge base by offering detailed information on the distribution of HPV genotypes within this specific area and our study strongly supports the use of HPV DNA analysis as an efficacious diagnostic method for management.

Additionally, the identification of various high-risk HPV genotypes in our research provides a valuable baseline for future studies and post-vaccination surveillance. It is essential to address psychological, spiritual and regional challenges before initiating the awareness programs, particularly in settings with limited resources.

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