

## Prevalence of Human Papilloma Virus infection Among Symptomatic Women with Positive Visual Inspection with Acetic Acid (VIA)

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### ABSTRACT

**Background:** HPV causes cancer among millions annually; the majority are cervical cancer in females. HPV subtypes 16 and 18 are associated with a higher risk of cervical cancer. VIA screening leads to early detection and intervention.

**Methodology:** This hospital based cross-sectional study done among 190 symptomatic multiparous women aged 30 to 50 years visiting a Gynaecology OPD. After informed consent, semi-structured questionnaire was administered to collect sociodemographic and reproductive health data. Gynaecologist performed a visual inspection with acetic acid (VIA), and positive were subjected for Real-time PCR detection of HPV subtypes.

**Results:** The age distribution was equal between 30-40 (48.9%) and 41-50 (51.1%), with a mean age of 40.73 ± 0.6 years. The mean age of menstruation was 13.96 ± 0.6. Vaginal discharge was the most common complaint (63.7%), and intermenstrual bleeding was less common (15%). The VIA positivity rate was 14.7% (28). The prevalence of the high-risk HPV subtypes 16, 18, and 12 was 1.6% (3), 1.1% (2), and 1.6% (3), respectively.

**Conclusion:** The married women aged 30 to 50 years had a moderate VIA positivity rate and low prevalence of high-risk HPV subtypes. This emphasizes on health education, better access to reproductive health care, and regular screenings.

**Key words:** VIA, married women of age 30 to 50 years, HPV

### INTRODUCTION:

Human Papilloma Virus (HPV) is the world's most common sexually transmitted disease, causing roughly 6 million cases of cancer (mainly cervical) in women and 70,000 in men each year <sup>1</sup>. In 2020, HPV will cause approximately 3.5 million cases <sup>2</sup>. In India, 6% to 37% of women have HPV on screening, primarily HPV 16 and 18, and 12.7% in Tamil Nadu, with the most frequent forms being HPV 16, 52, 58, 35, and 18 <sup>3</sup>. HPV causes cervical cancer, and high-risk HPV genotypes play an important role in the aetiology of cervical intraepithelial neoplasia (CIN) and invasive carcinoma. Visual inspection with acetic acid (VIA) is a low-cost screening tool used to detect cervical lesions in resource-limited situations. The prevalence of HPV infection among VIA-positive women is an important epidemiological factor impacting cervical cancer prevention <sup>4</sup>.

A study by Ghosh et al. <sup>5</sup> of women aged 30 to 50 years found 13.14% VIA-positive, statistically significant relationships with HPV infection <sup>5</sup>. Illiteracy, immunosuppression, smoking, higher parity, and poor sexual practices (e.g., sex at a young age) were all risk factors for HPV infection, which causes cervical cancer <sup>6</sup>. HPV vaccine (girls aged 9 to 14), health education (sex education), and annual VIA screening for all women aged 30 to 50 (5-year gap) significantly prevent cervical cancer <sup>7</sup>. VIA screening combined with HPV DNA testing resulted in early discovery and timely treatment. Cryotherapy or thermal ablation in VIA-positive women. High-grade lesions should be treated with the Loop

Electrosurgical Excision Procedure (LEEP) <sup>8</sup>. Further research is required to investigate VIA positivity and HPV genotype distribution in women, especially in rural areas, in order to optimize the screening strategy in high-risk groups.

**Objectives:** The objective of the study is to evaluate the prevalence of human papillomavirus infection among symptomatic women positive by visual inspection of acetic acid testing.

#### METHODOLOGY:

This cross-sectional study conducted for five months, from November 2023 to March 2024. The estimated sample size was 190, determined from a study by Pankaj et al. <sup>9</sup> that reported a 37% prevalence (p) of HPV among symptomatic women, with q (1-p) calculated as 63%, an absolute precision (d) of 7, and using the formula:  $n$  (sample size) =  $(Z_{1-\alpha})^2 pq / d^2$ , where  $(Z_{1-\alpha}) = 1.96$  is the constant for a 95% confidence interval. The study comprised 190 symptomatic women aged 30 to 50 years who visited the Gynaecology Outpatient Department (OPD). The study includes multiparous women of reproductive age (30-50 years) having any of the symptoms, such as vaginal discharge, intermenstrual bleeding, post-coital bleeding, and a clinically susceptible cervix. The study excludes women who are nulliparous, pregnant, have active vaginal bleeding, have frank cervical growth, or are known to be HIV positive.

The convenient sampling technique was employed, with all eligible and consenting women who visited the Gynaecology OPD during the study period being enrolled until the sample size was met. This method is feasible and widely utilized in hospital-based cross-sectional research. All the participants provided with informed written consent. A gynaecologist did the clinical examination. Prior to the study, the Institutional Ethics Committee provided clearance. A pre-tested, semi-structured questionnaire in the participants' native Tamil language was used to collect sociodemographic, reproductive, and clinical information. This increases the reliability of the collected data. Data was collected after informed consent by trained professionals. Visual Inspection with Acetic Acid (VIA) by a gynaecologist examined each subject using a speculum to detect the squamocolumnar junction. Using a big cotton swab to apply a 3-5% acetic acid solution to the cervix. After at least one minute, the cervix was checked for acetowhite alterations. The persistent acetowhite alterations in the transformation zone with well-defined boundaries were regarded as a positive outcome. The absence of persistent acetowhite alterations was documented as a bad outcome. Women who tested positive for VIA underwent cervical cell sampling. Samples were gathered using a tiny brush and stored in a preservation solution. The samples were delivered to the Molecular Laboratory for analysis. Real-time PCR was used to detect HPV types 16, 18, and other high-risk strains (31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68). The data collected were entered in a Microsoft Excel sheet. Analysis done using SPSS software version 16.0. The relevant statistical methods (e.g., descriptive statistics) were used to evaluate HPV prevalence and risk variables. The categorical data (such as VIA positivity) were presented as proportions and continuous data (such as age) were presented as means and standard deviations.

#### RESULTS:

Out of 190 study participants, 93 (48.9%) belong to the 30–40 years age group, and 97 respondents (51.1%) belong to the 41–50 years age group. The mean  $\pm$  SD age of the participants was  $40.73 \pm 0.6$  years. The majority (79.1%) had menarche at age 14, followed by 10.5% at age 15, 5.7% at age 12, 4.2% at age 13, and 0.5% at age 16. The average age of menarche was  $13.96 \pm 0.6$  (Table 1). The most common symptom is vaginal discharge (63.7%), but intermenstrual bleeding is less prevalent (15%). The majority (92%) had their first sexual intercourse after the age of 18, indicating that sexual initiation was delayed in this group. Only 10 (5.3%) used OCP. Fewer than 1.1% had an HPV-positive spouse, indicating a low level of education or prevalence in the sample (Table 2). Out of 190 study participants, 28 (14.7%) had VIA positive (Figure 1). Among the study participants, high-risk subtype HPV 16 were positive in 3 (1.6%), HPV 18 in 2 (1.1%), HPV 12 subtype 3 (1.6%) and no one tested positive for both HPV 16 and 18 (Table 3). HPV 16 and HPV 12 accounted for 11% (3) each, and HPV 18 accounted for 7% (2) among the VIA-positive cases (28). There were no combined HPV 16 and 18 found (Table 4).

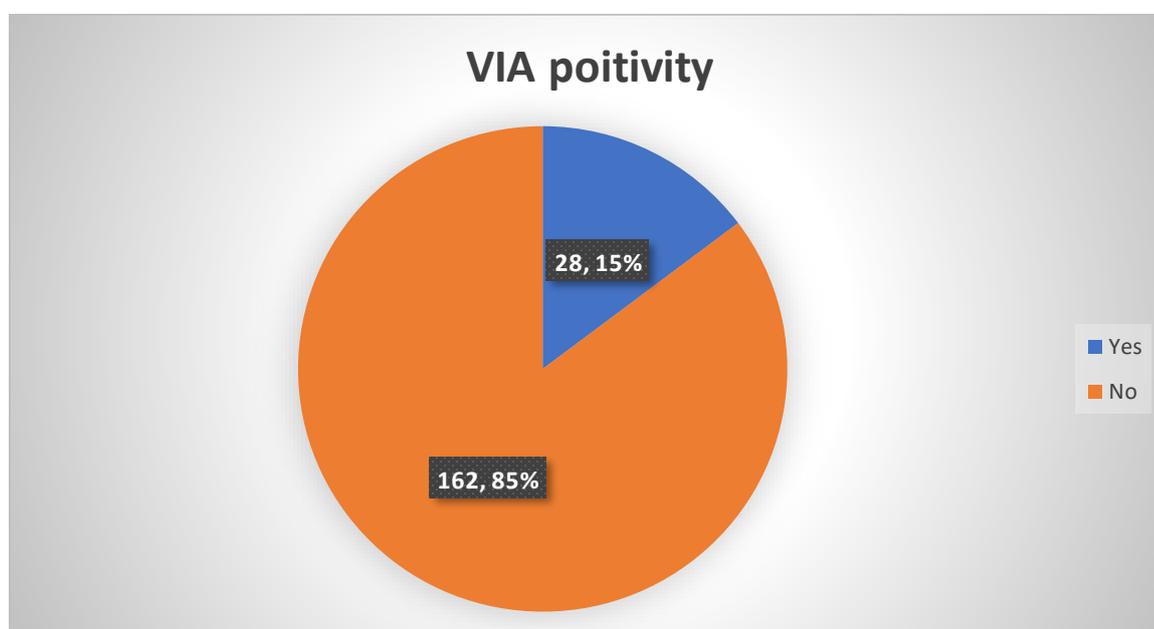
**Table 1: Age distribution and Age at Menarche of the study participants: N = 190**

Variables	Frequency	Percentage (%)	Mean age	SD
<b>Age (in years)</b>				
30 to 40	93	48.9	40.73	0.6445
41 to 50	97	51.1		
<b>Age at Menarche (in years)</b>				

12	11	5.7	13.96	0.633
13	8	4.2		
14	150	79.1		
15	20	10.5		
16	1	0.5		

**Table 2: Characteristics of the study participants: N = 190**

Variables	Frequency	Percentage (%)
<b>Symptoms</b>		
Discharge P/V	121	63.7
Inter menstrual bleeding	29	15.3
Postcoital bleeding	40	21
<b>Age at first sex</b>		
<18 years	15	8
>18 years	175	92
<b>OCP Use</b>		
Yes	10	5.3
No	180	94.7
<b>Partner with HPV</b>		
Yes	2	1.1
No	188	98.9



**Figure 1: VIA positivity among the study participants: N = 190**

**Table 3: HPV subtypes among study participants: N = 190**

HPV genome	Frequency	Percentage (%)
<b>HPV 16</b>		
Yes	3	1.6
No	187	98.4
<b>HPV 18</b>		
Yes	2	1.1
No	187	98.9
<b>HPV 12</b>		
Yes	3	1.6
No	187	98.4
<b>HPV 16 &amp; 18</b>		
Yes	0	0
No	190	100

**Table 4: HPV subtypes among VIA positives: N = 28**

HPV genome	Frequency	Percentage (%)
<b>HPV 16</b>		
Yes	3	11
No	25	89
<b>HPV 18</b>		
Yes	2	7
No	26	93
<b>HPV 12</b>		
Yes	3	11
No	25	89

**DISCUSSION:**

This study was conducted to determine the prevalence of HPV by VIA screening, as well as the reproductive characteristics of 190 symptomatic women aged 30 to 50. According to cervical cancer screening criteria, the study population was roughly equally divided into two age groups: 30-40 and 41-50. The mean age at menarche was  $13.9 \pm 0.6$  years, with the majority (79.1%) experiencing it at the age of 14. The study by Meher and Sahoo et al. <sup>10</sup> showed 66% of the South Indian women had a mean age at menarche of 13.7 years, which is similar to our study results. Early menarche has been associated with greater gynaecological risk.

In this study, 92% of the participants initiated sexual activity (Age at first sex) at an age beyond 18, which is parallel to the NFHS 5 <sup>11</sup> findings on sexual initiation among Indian women at 18 years. In most circumstances, delayed sexual beginning is associated with a lower risk of HPV infection and cervical cancer, which helps to explain the study's relatively low HPV prevalence. The prevalence of oral contraceptive pills (OCP) utilization is 5.3% which is similar to the study by Nair et al. <sup>12</sup> (2.4%) in rural areas of Tamil Nadu. The cultural attitudes, limited availability, and lack of understanding could all be contributing factors to our study subjects' poor use. Vaginal discharge was the most common

symptom (63.7%). Intermenstrual haemorrhage was less common (15%), possibly reflecting fewer cases of advanced cervical pathology or other gynaecological disorders.

The VIA positivity rate was 14.7% (10%-20.6%), indicating a moderate frequency of cervical precancerous lesions in this cohort. This is comparable to Poli et al.<sup>13</sup> (2017) rural settings study, which found 10.8% VIA positive rates among the same age groups. The prevalence of HPV 16, HPV 18, and HPV 12 subtypes were 1.6%, 1.1%, and 1.6%, respectively. The HPV 16 and 18 co-infection was not detected, which could be due to minimal exposure or effective immune clearance in this cohort. These findings are consistent with international HPV prevalence data, which show that HPV 16 and 18 are the most common high-risk forms but vary greatly by geography and population subgroup. The study (2019) by Bruni et al.<sup>14</sup> reported that HPV 16 prevalence ranged from 2% to 10% in the general population. Also, in the study<sup>15</sup> done in South India reported prevalence of HPV 16 was 2.75% and HPV 18 was 22%. The low rate of HPV-positive spouses (1.1%) also indicates a low risk of transmission in this cohort, which could be due to restricted exposure to sexual partners or low community prevalence. The prevalence of HPV 16 and 18 among VIA-positive women (11% and 7%, respectively) is lower than in the study conducted by Vinodhini et al. 16 among Tamil Nadu women, where HPV 16 accounts for more than 30-40% of cases. This signifies the importance of VIA screening at the grassroots level.

#### CONCLUSION:

The current investigation found a moderate burden of cervical precancerous lesions (14.7% VIA positive) and a low incidence of high-risk strains of HPV in women aged 30 to 50. The majority reported a delayed onset of menarche at 14 years, delayed initiation of sexual engagement, and low use of oral contraceptives. These elements may help to explain HPV's relatively low incidence. Thus, the findings emphasize the importance of culturally tailored health education, increased access to reproductive health care, and cervical cancer screening as a regular practice for lowering cervical cancer risk among this population. Additional large-scale research is recommended to confirm these findings and identify impediments to contraception and HPV vaccine use.

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**Conflict of Interest: NIL**

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