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ORIGINAL RESEARCH

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Colposcopic biopsy findings in ASCUS or normal cervical cytology patients with high-risk HPV positivity

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Abstract

The aim of the present study was to compare colposcopic biopsy results of women in Turkey with normal cervical cytology and Atypical Squamous Cells of Undetermined Significance (ASCUS) who had human papilloma virus (HPV) genotypes 16, 18 and combined 16/18. The overarching goal was to enhance the existing body of evidence on cervical cancer screening strategies, with an ultimate aim of refining HPV testing guidelines and improving patient management. In this retrospective study, we examined the medical records of 1121 patients from a tertiary health care setting who tested positive for HPV 16, HPV 18 or both, and who exhibited ASCUS or normal Pap smear findings. A detailed review of the patients' colposcopic biopsy outcomes was conducted, with particular attention to their HPV genotype status and the impact of smoking. The 1121 patients were classified based on HPV genotype into three groups: HPV 16 (78.5%), HPV 18 (15.8%), and co-infection with HPV 16 and 18 (5.7%). On the basis of smear characteristics, patients were categorized as normal (81.4%) and ASCUS (18.2%). Approximately 40% of patients infected with HPV 16, HPV 18 or both with no evidence of intraepithelial lesion or malignancy (NILM) or ASCUS on Pap smear had High-grade Squamous Intraepithelial Lesion cancer on biopsy. Notably, for those with normal smear results, the rate of Low-grade Squamous Intraepithelial Lesion biopsy was approximately 15% higher in the HPV 18 group than the HPV 16 group (59.6% *vs.* 45.8%; $p = 0.023$). Smoking prevalence was significantly higher in the co-infected HPV 16/18 group ($p = 0.013$). This study underscores the importance of vigilant HPV and cytology testing, especially for individuals with HPV 16/18, regardless of normal cytology findings.

Keywords

HPV 16/18; Colposcopic biopsy; HSIL; LSIL; Cytology screening

1. Introduction

Human Papilloma virus (HPV) continues to pose a major global health threat due to its direct link with various cancers, particularly cervical cancer. Among the many types of HPV, certain strains are classified as high-risk because of their potential to trigger malignant diseases. Specifically, HPV types 16 and 18 have drawn considerable scientific attention and have been the subjects of rigorous study, given their prevalent occurrence and robust correlation with the onset and progression of cervical cancer [1, 2]. The traditional screening method for cervical cancer includes cytological examination, encompassing the Papanicolaou (Pap) smear test, coupled with HPV DNA testing specifically targeting HPV 16 and 18. This combined approach, often referred to as co-testing, enhances the accuracy of detection and has become a cornerstone in cervical cancer prevention [2, 3].

In the context of cervical cancer prevention, several studies have highlighted the significance of HPV 16/18 and the value of co-testing methods. Saslow *et al.* [4] in a guideline supported by the American Cancer Society, American Society for Colposcopy and Cervical Pathology, and American Society for Clinical Pathology, concluded that the combination of HPV and Pap smear tests for women over 30 years of age was more effective than the Pap smear test alone. Schiffman *et al.* [5] found that HPV 16 significantly increased the risk of cervical pre-cancer and cancer compared to other high-risk HPV types and noted similar but less pronounced effects associated with HPV 18. Additionally, MacLaughlin *et al.* [6] demonstrated that the combination of Pap smear and HPV DNA testing achieved higher sensitivity in the early detection of cervical cancer, emphasizing that co-testing effectively identifies women at risk for cervical cancer.

Despite advances in screening technologies, the dilemma persists in clinical practice about the immediate necessity of biopsy in patients with certain cytological findings, especially when they test positive for HPV 16 or 18. The intricate relationship between these specific HPV types, cytological

results, and the progression of cervical disease presents a complex scenario that demands careful consideration and tailored approaches.

In this context, our study's principal objective is to delve into the association between HPV genotypes 16 and 18 and the outcomes of colposcopic biopsies in patients presenting with Atypical Squamous Cells of Undetermined Significance (ASCUS) or normal cytology. By concentrating exclusively on these particular HPV types, this retrospective study aims to enrich our understanding of HPV 16 and 18's impact on cervical disease evolution, alongside potential lifestyle variables such as smoking. The overarching aspiration is to bolster the existing scientific literature concerning cervical cancer screening techniques, ultimately refining HPV testing guidelines and enhancing patient management.

2. Material and methods

2.1 Patient selection

Our study was a retrospective analysis of colposcopy and biopsy results conducted between January 2017 and December 2021. We examined and compared the results of patients who tested positive for HPV 16, 18 had ASCUS or normal Pap smear results, and underwent colposcopy and minor procedures (*e.g.*, cervical biopsy). Inclusion criteria: (1) women with normal or ASCUS cytology and Hr-HPV infection (2) women accepted colposcopic examination and underwent cervical biopsy under colposcopic guidance.

Exclusion criteria: (1) women with history of treatments to cervical lesions, such as cervical surgery or medical treatmen (2) women with cervical cancer (3) women with malignant tumors (4) women with autoimmune diseases or receiving immunotherapy (5) women with pregnancy. Demographic factors investigated included age, gravidity, parity and smoking status.

2.2 Cervical smear, HPV and colposcopic biopsy

Cervical smear samples were obtained using a plastic brush, placed in containers prepared for the centrifuge-based liquid cytology BD Surepath test method (Becton, Dickinson and Company, Sparks, MD21152 USA), and sent for pathological analysis. Liquid-based pap tests were reported according to the 2014 Bethesda System.

All colposcopic examinations were performed by one specialist in gynecological oncology, and the biopsy and final histological excision results were reviewed by one or two experienced gynecological pathologists following a double-blind method. To type the cervical lesions, we used a novel classification system, College of American Pathologists (CAP) and American Society of Colposcopy and Cervical Pathology (ASCCP) Lower Anogenital Squamous Terminology. In this system, cervical lesions are classified as either high grade (Cervical squamous intraepithelial neoplasia 2–3) or low grade (CIN 1) [7].

Smear samples obtained with cervical brushes were sent to our hospital microbiology laboratory in the Abbott Cervi-Colect Specimen Collection Kits. The samples were subjected to HPV DNA isolation by Polymerase Chain Reaction Cobas 4800 System Hr-HPV test (Roche Diagnostics, Basel, Switzerland) and reported as positive or negative for high-risk HPV, HPV-16 and HPV-18.

All individuals with HPV16+/18+/16+18/ASCUS, and HPV16+/18+/16+18/Normal cytology underwent colposcopy. The procedures were performed with a binocular Leica CLS 150 XC brand colposcope (Leica, Germany), capable of 20× magnification and equipped with a green filter. Colposcopic findings were described using the criteria of the International Federation of Cervical Pathology and Colposcopy [8]. Punch biopsies were obtained from acetowhite areas, mosaics, punctuations, erosions, leukoplakia, atypical vascular formations, and iodine-negative areas. Loop electrosurgical excision (LEEP) was performed for therapeutic purposes in patients with High-grade squamous intraepithelial lesion (HSIL) biopsy results. Endocervical curettage sampling was also performed.

2.3 Statistical evaluation

Patient information was extracted from the colposcopy files and transferred to SAS Studio (SAS Institute Inc. 2015. SAS/IML® 14.1 User's Guide. Cary, NC: SAS Institute Inc.). Continuous data are presented as the mean ± Standard Deviation, while categorical data are presented as percentages (%). Shapiro-Wilk test was used to investigate the normality of the data. To compare groups that did not exhibit a normal distribution, the Mann-Whitney U test was used for two groups and the Kruskal-Wallis H test was used for three or more groups. Pearson Chi-Square and Pearson Exact Chi-Square analyses were used to analyze cross tables formed. Statistical significance was set at $p < 0.05$.

3. Results

The study included a total of 1121 patients, with an average age of 41.3 ± 10.3 years and a median age of 40 years. The age range of the patients was 30–65 years. The overall average Body Mass Index (BMI) was found to be 24.5 ± 2.7 kg/m^2. Based on smear characteristics, the patients were divided into two categories: normal (n = 912, 81.4%) and ASCUS (n = 204, 18.2%).

The results of five patients (0.5%) were not included in the study due to Inadequate tissue. The distribution according to the HPV genotype was as follows: HPV 16 (n = 875, 78.5%), HPV 18 (n = 177, 15.8%) and HPV 16/18 (n = 64, 5.7%). No statistically significant differences were found among the HPV genotype groups regarding educational and socioeconomic status ($p > 0.05$). The smoking addiction rate was approximately 20% higher for the combined HPV 16 and 18 types, showing a significant difference from other single HPV types ($p = 0.013$) (Table 1).

When examining the biopsy status of patients divided by smear type and HPV genotype groups, a significant difference was only found in the normal smear type with Low-grade Squamous Intraepithelial Lesion (LSIL) biopsy between HPV-16 and HPV-18 genotypes ($p = 0.023$) (Table 2). The rate of LSIL biopsy was approximately 15% higher in the HPV-

TABLE 1. Comparison of lifestyle and socioeconomic variables among patients with HPV genotypes 16, 18, and combined 16/18.

Variables	Subgroups	HPV Genotype			*p* Value
		HPV 16 (n = 875)	HPV 18 (n = 177)	HPV 16/18 (n = 64)	
			n (%)		
Education					
	High School	580 (66.3)	121 (68.4)	35 (54.7)	0.128
	University	295 (33.7)	56 (31.6)	29 (45.3)	
Socioeconomic					
	Low	574 (65.6)	114 (64.4)	34 (53.1)	0.131
	High	301 (34.4)	63 (35.6)	30 (46.9)	
Smoking					
	No	539 (61.6)	113 (63.8)	28 (43.8)	0.013
	Yes	336 (38.4)	64 (36.2)	36 (56.3)*	

*Pearson's chi square test used and $p < 0.05$ considered significant. *Indicates a statistically significant difference. HPV: Human Papilloma virus.*

18 group than in the HPV-16 group (59.6% *vs.* 45.8%). illustrates the distribution of various cervical biopsy outcomes, including LSIL, HSIL, No-Dysplasia and Cancer, among the HPV genotype subgroups. Approximately 40% of patients infected with HPV-16, HPV-18 or both with no evidence of intraepithelial lesion or malignancy (NILM) or ASCUS on Pap smear had HSIL or cancer on biopsy. In addition, the prevalence of HSIL or cancer was higher among those with ASCUS (44%) compared to those with NILM (36%). This difference almost reaches statistical significance ($p = 0.53$). The prevalence of HSIL and cancer was higher in women infected with HPV-16 (41%) than those infected with HPV-18 (23%). The distribution of HPV types according to biopsy results is shown in (Table 2). LSIL was found as a result of colposcopic biopsy of the patient with normal cervical cytology and HPV 16 positivity (Fig. 1). HSIL was found as a result of colposcopic biopsy of the patient with cervical cytology ASCUS HPV 16 positivity (Fig. 2).

There were no significant differences among the HPV genotype groups in smear type, parity score, postcoital bleeding, Post menapousal bleeding (PMB), Abnormal uterine bleeding (AUB), intrauterine device and condom use. However, a significant difference was found in the presence of smelly discharge between HPV 16 and 18 genotypes, with a higher rate observed for HPV 18 (41.2% *vs.* 28.6%) ($p = 0.002$). Oral contraceptive Pill (OCS) usage was significantly higher in the HPV 16 group than in the HPV 16 and 18 groups (28.2% *vs.* 15.6%) ($p = 0.031$). Sexual protection rates were significantly higher in the HPV 18 and HPV 16 and 18 genotype groups than in the HPV 16 group ($p = 0.045$) (Table 3).

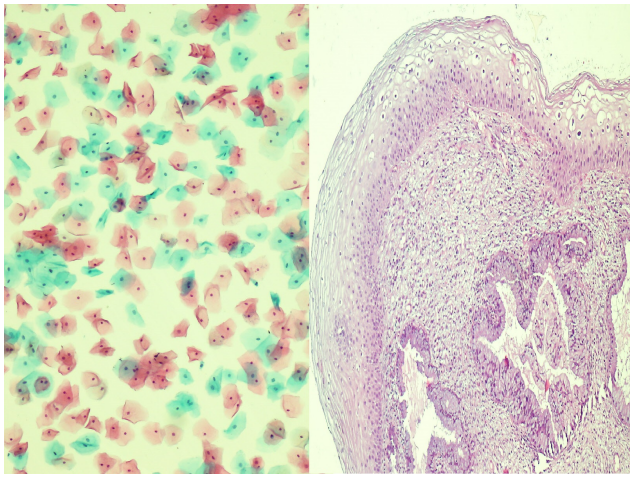

FIGURE 1. Normal cytology + HPV 16 positive + LSIL biopsy.

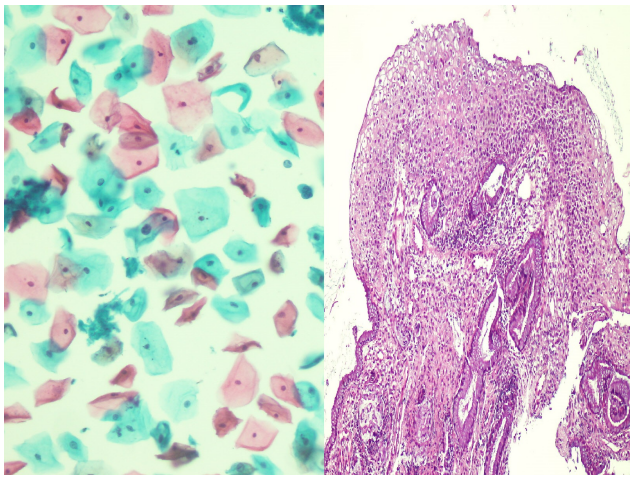

FIGURE 2. ASCUS + HPV 16 positive + HSIL biopsy.

4. Discussion

The present study explores an extensive patient base of 1121 individuals aged between 30 and 65 years, depicting a broad representation of the populace. With the average age being 41.3 ± 10.3 years and a median of 40 years, our cohort reflects

TABLE 2. Comparison of cervical biopsy outcomes across HPV genotype subgroups by smear result.

Biopsy	HPV Groups						*Post hoc* comparison *p*-value					
	HPV-16 (n = 875)		HPV-18 (n = 177)		HPV 16&18 (n = 64)		HPV-16 *vs.* HPV-18		HPV-16 *vs.* HPV-16&18		HPV-18 *vs.* HPV-16&18	
	NI	Ascus	NI	Ascus	NI	Ascus	NI	Ascus	NI	Ascus	NI	Ascus
LGSIL	321 (45.6)	73 (42.7)	89 (59.7)*	15 (53.6)	27 (57.4)	9 (52.9)	0.023*	0.508	0.327	0.563	0.838	0.917
HGSIL	267 (37.9)	66 (38.6)	30 (20.1)	10 (35.7)	18 (38.3)	6 (35.3)	0.061	0.867	0.977	0.667	0.241	0.766
No-Dysplasia	93 (13.6)	26 (15.2)	30 (20.1)	2 (7.1)	2 (4.3)	1 (5.9)	0.418	0.790	0.745	0.886	0.626	0.967
Cancers	20 (2.8)	6 (3.5)	1 (0.7)	1 (3.6)	0 (0)	1 (5.9)	0.896	1.000	0.868	0.823	0.962	0.912
SCC	16 (80.0)	4 (66.7)	1 (100)	0 (0)	0 (0)	0 (0)	0.619	0.212				
Adenocarcinoma	4 (20.0)	2 (33.3)	0 (0)	1 (100)	0 (0)	0 (0)	0.619	0.212				

*Pearson's Chi Square used and $p < 0.05$ considered significant. LGSIL: low-grade squamous intraepithelial lesion; HGSIL: high-grade squamous intraepithelial lesion; SCC: Squamous cell carcinoma; NI: Normal and/or inflammation; HPV: human papilloma virus. *Indicates a statistically significant difference.*

the average age of HPV infected individuals as reported in previous literature [9]. Furthermore, the overall average Body Mass Index (BMI) was well within the healthy range, as per World Health Organization (WHO) guidelines (24.5 ± 2.7 kg/m^2), indicating that the influence of BMI on the development of cervical pathology is potentially minimal, aligning with a similar finding reported by Poorolajal *et al.* [10]. The cohort was split into two distinct categories based on smear characteristics: Normal (81.4%) and ASCUS (18.2%). This categorization offers a new perspective on the analysis of outcomes, aligning with the less frequent use of this approach in prior research, thereby increasing the novelty and uniqueness of our findings [11]. The HPV genotype distribution predominantly consisted of HPV 16 (78.5%), followed by HPV 18 (15.8%) and a minority with co-infection of both HPV 16/18 (5.7%). These figures correlate with earlier studies that reported a higher prevalence of HPV 16 over HPV 18 [9, 12]. Our data reinforces these findings while providing a more precise understanding of the genotype distribution within the two different categories of patients. Several lifestyle and social factors were also analyzed, including the level of education, socioeconomic status, and smoking habits. It's noteworthy that smoking prevalence was significantly higher in the co-infected HPV 16/18 group, a finding that supports previous studies which demonstrate an elevated susceptibility to HPV infection among smokers, potentially due to the damaging effects of smoking on immune function [13].

Comparison with existing research can offer valuable insights into HPV genotypes' epidemiology and their associated factors. However, direct comparisons can sometimes pose challenges due to the varying population characteristics, HPV testing methodologies, and statistical analysis methods employed across different studies [13, 14]. Large-scale, multicenter studies would serve to substantiate our findings and establish stronger associations.

A noteworthy discovery was that the prevalence of LSIL was significantly higher among the HPV-18 group as compared to the HPV-16 group for the normal smear type. This finding adds a new dimension to the existing literature which often associates HPV 16 with higher HSIL and cancer than HPV 18 [9, 14]. In addition, the prevalence of HSIL or cancer was higher among those with ASCUS (44%) compared to those with NILM (36%). This difference almost reaches statistical significance ($p = 0.53$). The prevalence of HSIL and cancer was higher in women infected with HPV-16 (41%) than those infected with HPV-18 (23%) [9]. The reason for the difference seen in our study could potentially be attributed to variations in population characteristics, or due to the different rates of progression associated with different HPV types. In their extensive study on adolescents, Moore *et al.* [15] undertook a comprehensive evaluation of both cytological and histopathological aspects of cervical dysplasia. The results indicated a high incidence of CIN grade 2 or higher (CIN-2+) in 32.5% of their cohort. A majority of these patients were initially identified with Low-grade Squamous Intraepithelial Lesion or lower on their cytology reports. The increased risk for cervical dysplasia in adolescents is often attributed to their sexual behavior patterns and heightened exposure to high-risk human papilloma viruses. There is ongoing debate suggesting that adolescent dysplasia often follows a transient course, proposing that a more conservative approach might be sufficient in management. However, the significant presence of CIN-2+ in the adolescent cohort of this study indicates that dysplasia in this age group is not an insignificant finding and thus requires

TABLE 3. Comparison of lifestyle and clinical factors among HPV genotype groups.

Variables	Subgroups	HPV Genotype			*p* Value
		HPV 16 (n = 875)	HPV 18 (n = 177)	HPV 16/18 (n = 64)	
			n (%)		
Smear					
	Normal	710 (80.7)	149 (84.2)	53 (82.8)	0.618
	Ascus	165 (18.8)	28 (15.8)	11 (17.2)	
Parity					
	0	60 (6.9)	14 (7.9)	3 (4.7)	0.315
	1	248 (28.4)	50 (28.2)	14 (21.9)	
	2	388 (44.4)	82 (46.3)	26 (40.6)	
	3	146 (16.7)	26 (14.7)	20 (31.3)	
	4	29 (3.3)	5 (2.8)	1 (1.6)	
	5	3 (0.3)	0 (0)	0 (0)	
Smelly Discharge					
	No	624 (71.3)	104 (58.8)	40 (62.5)	0.002
	Yes	251 (28.6)*	73 (41.2)	24 (37.5)	
Postcoital Bleeding					
	No	696 (79.5)	145 (81.9)	56 (87.5)	0.258
	Yes	179 (20.5)	32 (18.1)	8 (12.5)	
PMB					
	No	823 (94.1)	174 (98.3)	59 (92.2)	0.050
	Yes	52 (5.9)	3 (1.7)	5 (7.8)	
AUB					
	No	753 (86.1)	163 (92.1)	58 (90.6)	0.064
	Yes	122 (13.9)	14 (7.9)	6 (9.4)	
OCS					
	No	629 (71.9)	138 (78.0)	54 (84.4)	0.032
	Yes	246 (28.1)	39 (22.0)	10 (15.6)*	
Sexual Protection					
	No	541 (61.8)	93 (52.5)	35 (54.7)	0.048
	Yes	334 (38.2)*	84 (47.5)	29 (45.3)	
Intrauterine Device					
	No	681 (77.8)	143 (80.8)	49 (76.6)	0.648
	Yes	194 (22.2)	34 (19.2)	15 (23.4)	
Condom					
	No	772 (88.2)	157 (88.7)	55 (85.9)	0.837
	Yes	103 (11.8)	20 (11.3)	9 (14.1)	

*Pearson's chi square test used and $p < 0.05$ considered significant. PMB: postmenaposal bleeding; AUB: abnormal uterine bleeding; OCS: Oral Contraceptive Pill; HPV: Human Papilloma Virus. *Indicates a statistically significant difference.*

vigilant monitoring and management [15]. Torres-Ibarra and colleagues have demonstrated that the combination of HPV DNA testing and Papanicolaou (Pap) cytology screening offers nearly 100% sensitivity and negative predictive value for detecting HSIL. This highlights the potential utility of this combined screening strategy for optimizing cervical cancer prevention programs. In the context of our discussion, their findings support the need for co-testing, as it not only improves detection rates but also offers significant prognostic assurance [16].

Interestingly, the absence of cancer cases in the combined HPV 16/18 group stood out in contrast to previous studies suggesting an increased risk of cancer with co-infections [17, 18]. The study conducted by Liaoa and colleagues identified an intriguing preference for co-infection between HPV 16/18 and types 31, 52 and 58. This finding underscores the potential necessity for developing and promoting prophylactic HPV vaccines that provide protection against a broader range of genotypes [18]. The research conducted by Tantengco *et al.* [19] exclusively identified the presence of HPV-18 and HPV-52 in patients with cervical cancer. Furthermore, within the HPV-positive cohort, there was a noted co-infection rate of 22.73% with Ureaplasma spp. and 9.09% with Mycoplasma spp. This observation contributes to our understanding of potential co-infections that may influence the progression of HPV-related cervical pathologies [19]. The contrast findings in our study might be due to the relatively smaller size of the co-infected group in our study, reinforcing the need for more expansive research to validate this finding.

Additionally, our study analyzed the association of various factors like smear type, parity score, smelly discharge, post-coital bleeding, PMB, AUB, intrauterine device use, condom use and HPV genotype groups. Of note, the presence of smelly discharge was significantly higher in the HPV 18 group ($p = 0.002$). This could potentially indicate the presence of a co-existing vaginal or cervical infection [19]. Our study, therefore, underscores the importance of thorough clinical examination and routine screening.

The use of oral contraceptives OCS was higher in the HPV 16 group than in the HPV 18 and HPV 16/18 groups ($p = 0.031$). While HPV-16 is often associated with precancerous lesions, the association with OCs is a finding that calls for more detailed exploration in larger studies. Our research reveals a connection between an increase in longer-term use of OCs, and HPV exposure, particularly HPV-16. Consequently, these findings underscore the importance of vigilant cytologic screening and ongoing epidemiological studies for young women using OCS. Additionally, they observed a potential cohort effect in the incidence of cervical lesions suggesting a trend of escalated risk with extended OCS use and an increased risk associated specifically with HPV-18. These observations underscore the necessity to comprehend and continuously monitor the intricate interplay between HPV infection, contraceptive utilization, and transformations in cervical cells [20].

Our results draw attention to the potential role of various factors in the progression of cervical abnormalities alongside HPV genotype. The implications of these findings could potentially guide the development of comprehensive prevention and control strategies for HPV infection and associated conditions.

Finally, it is worth highlighting the importance of patients with normal cytological findings who are HPV 16/18 positive. Even though our study reported a high occurrence of normal cervical cytology across all three HPV genotype groups, the fact that HPV infection can often progress without detectable cervical cytological abnormalities, especially in HPV 16/18 positive patients, underlines the need for continuous vigilance and follow-up.

The natural history of HPV infection is complex. Some individuals can clear the infection, while in others, it can persist and potentially lead to precancerous lesions and, eventually, invasive cancer [9, 12, 13]. Particularly in the case of high-risk HPV types like HPV 16 and 18, the risk of progression is significantly higher [1, 3, 5].

Moreover, cytology screening, although crucial in cervical cancer prevention, is not flawless. Normal cytology does not guarantee the absence of a high-grade lesion or cancer. The effectiveness of the Pap test in detecting cervical cancer has often led to a misperception of its infallibility. However, it is essential to note that the test isn't flawless. The sensitivity of the Pap test in identifying HSIL falls between 70 and 80 percent. The test's sensitivity is limited due to various factors, such as the minuscule size of a lesion, a lesion's inaccessible location, unsampled lesions, the scant presence of abnormal cells on the slide, diminutive size of the abnormal cells, or visualization obscured by inflammation and/or blood. Even in meticulously optimized screening programs, false-negative results inevitably occur and cannot be completely eradicated [21, 22]. In our study, despite the normal cytology, the substantial presence of HPV 16/18 underlines the possibility of occult cervical intraepithelial neoplasia (CIN) or cancer, which may not be detectable in a single round of screening.

Limitations of our study include the lack of longitudinal follow-up, which could provide valuable insights into the progression and clearance of HPV infections. Future studies should incorporate this aspect to further our understanding of the natural history of HPV infection, particularly in individuals with normal cytology findings but positive for HPV 16/18.

5. Conclusions

In conclusion, our study emphasizes the importance of integrated HPV and cytology ("co-testing") or primary HPV testing, particularly for those with HPV 16/18 positivity, even if they exhibit normal cytological findings. This strategy could lead to earlier detection of precancerous lesions or cervical cancer, thus allowing for timely intervention and potentially improving patient outcomes.

This study provided that the prevalence of LSIL was significantly higher among the HPV-18 group as compared to the HPV-16 group for the normal smear type. This finding adds a new dimension to the existing literature which often associates HPV 16 with higher HSIL and cancer than HPV 18. Moreover, the prevalence of HSIL or cancer was higher among those with ASCUS (44%) compared to those with NILM (36%). The prevalence of HSIL and cancer was higher in women infected with HPV-16 (41%) than those infected

with HPV-18 (23%). Our findings indicate a high prevalence of normal cervical cytology across all HPV groups, illustrating the stealthy progression of HPV infection, especially in those carrying high-risk genotypes. Moreover, we have noted potential associations between HPV genotypes and specific clinical symptoms like smelly discharge and an observed influence on lifestyle behaviors such as contraceptive use. In the realm of public health, the findings underscore the necessity for robust HPV vaccination programs and sexual health education, particularly in populations infected with HPV 16. Overall, this study signifies a progressive step forward in our understanding of the implications of different HPV genotypes on lifestyle and clinical manifestations, potentially impacting future patient management and preventive strategies.

AVAILABILITY OF DATA AND MATERIALS

The datasets used during the current study are available from the corresponding author on reasonable request.

AUTHOR CONTRIBUTIONS

EK and ÖE—protocol/project development; manuscript writing/editing. EDO and GG—acquisition the data. MB and MS—analyzed the data. DA—immunohistochemical experiments. All authors contributed to the study conception, design and analysis, drafting and writing the paper, searching related articles and interpretation, Conceived and designed the experiments.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Ethical approval for this study was obtained from the Local Ethics Committee of the Tepecik Training and Research Hospital (decision number 2023/03-40). All patients provided written informed consent.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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