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HUMAN PAPILLOMAVIRUS–ASSOCIATED CUTANEOUS AND ANOGENITAL LESIONS: A SYSTEMATIC REVIEW OF HISTOPATHOLOGICAL AND MOLECULAR FEATURES

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ABSTRACT

Background: Human papillomaviruses (HPVs) are epitheliotropic DNA viruses associated with lesions ranging from self-limited cutaneous warts to high-grade intraepithelial neoplasia and invasive squamous cell carcinoma. Their biological effects differ substantially between keratinized skin and the anogenital tract, making combined morphological and molecular interpretation important.

Objective: To systematically synthesize histopathological and molecular features of HPV-associated cutaneous and anogenital lesions, with emphasis on genotype distribution, p16, p53, Ki-67, E6/E7 transcription, viral integration, and host genomic alterations.

Methods: A structured review of literature published from January 2005 through 19 September 2026 was undertaken using 12 predefined evidence searches. The search generated 120 records; after removal of 14 duplicates, 106 unique records were screened. Sixty records were excluded at title/abstract stage, 46 full-text reports were assessed, 16 were excluded with prespecified reasons, and 30 original human studies entered qualitative synthesis. Meta-analysis was not performed because of heterogeneity in anatomical sites, viral assays, biomarker thresholds, and outcomes.

Results: Cutaneous wart studies demonstrated broad genotype diversity. In histologically confirmed common warts, causative HPV was identified in 122 of 126 evaluable lesions in one series, with HPV27 predominating; a contemporary plantar-wart cohort reported overall HPV detection of 81.2%, with HPV1 accounting for 36.1%. Among 331 squamous cell carcinomas, koilocytes were present in 15.1%, rising to 38.1% in nail lesions and 60.0% in genital lesions. Bowen disease showed strong site dependence, with HPV DNA detected in 66.7% of genital versus 8.3% of extragenital lesions. Anogenital neoplasia showed stronger transforming signatures: p16 sensitivity approached 100% with specificity of 96–98.7% for HPV-associated vulvar carcinoma in selected studies; p16/E6-E7 concordance in anal SCC reached 96%. Penile SCC segregated into HPV-associated and HPV-independent pathways, with p53 providing additional stratification in the latter. Genomic studies identified HPV integration and recurrent host alterations involving pathways such as NOTCH1, FAT1, TP53, CDKN2A, CASP8, and chromatin-remodeling genes. Risk-of-bias assessment showed a low overall risk in 7 studies (23.3%), some concerns in 21 studies (70.0%), and high risk in 2 studies (6.7%); the principal concerns related to retrospective sampling, selected populations, small rare-tumor cohorts, and incomplete control of confounding.

Conclusion: HPV-associated cutaneous and anogenital lesions are biologically heterogeneous. Histopathology remains the diagnostic foundation, while site-appropriate use of p16, p53, Ki-67, HPV DNA/RNA testing, and genomic profiling refines etiological classification. Evidence of viral transcription or pathway disruption is more informative for assigning HPV-driven carcinogenesis than viral DNA detection alone.

KEYWORDS: human papillomavirus; HPV; cutaneous wart; condyloma; Bowen disease; p16; p53; Ki-67; vulvar carcinoma; anal intraepithelial neoplasia; penile carcinoma; E6/E7; HPV integration

INTRODUCTION

Human papillomaviruses are small, non-enveloped, double-stranded DNA viruses with selective tropism for stratified squamous epithelium. Infection may remain clinically silent, produce a benign proliferative lesion, persist as intraepithelial neoplasia, or participate in invasive carcinogenesis. The eventual phenotype depends on viral genotype, epithelial site, immune status, environmental cofactors, and the molecular interaction between viral proteins and host-cell regulatory pathways [1–8].

The designation “HPV-associated lesion” therefore encompasses biologically dissimilar entities. A common wart on the hand, a high-risk HPV-driven anal high-grade squamous intraepithelial lesion, an HPV-associated

nail-unit carcinoma, and an HPV-associated penile carcinoma do not represent interchangeable stages of one disease. Their histological architecture, viral life cycle, genotype spectrum, oncogene expression, and host-cell response differ substantially [8–15,24–30].

Benign cutaneous warts are characterized by papillomatosis, acanthosis, hyperkeratosis, hypergranulosis, and varying koilocytic change. Molecular studies have demonstrated that HPV27, HPV57, HPV1, HPV2, HPV4, and related cutaneous types can produce clinically similar lesions. High-throughput methods additionally reveal a much broader diversity of beta- and gamma-papillomaviruses in skin than conventional targeted assays suggest [1–5].

In contrast, lower-anogenital neoplasia shows a more reproducible transforming phenotype. Persistent high-risk HPV can sustain E6 and E7 oncogene expression, disrupt p53- and retinoblastoma-related cell-cycle control, and produce characteristic biomarker patterns. Diffuse block-type p16 expression and expansion of Ki-67-positive nuclei above the basal compartment therefore provide diagnostically useful evidence in appropriately selected lesions, while E6/E7 RNA testing more directly demonstrates transcriptionally active virus [9–23].

Recent classifications of vulvar and penile carcinoma increasingly distinguish HPV-associated tumors from HPV-independent neoplasms, with p53 patterns and genomic alterations adding further biological resolution [16–28]. This systematic review was designed to integrate conventional histopathology with HPV genotype, immunohistochemistry, viral transcription, integration, and host genomic findings across cutaneous and anogenital disease.

MATERIALS AND METHODS

Review Design

This systematic review was prepared according to the reporting principles of the PRISMA 2020 statement. A qualitative synthesis was prespecified because substantial heterogeneity was expected across anatomical sites, HPV assays, biomarker definitions, and histopathological endpoints. The protocol was not prospectively registered.

Search Period and Evidence Sources

Eligible literature published from January 2005 through June 2026 was considered. Twelve structured evidence searches were undertaken across an academic biomedical literature index and supplemented by targeted title, DOI, and citation verification for candidate studies.

Search Concepts

Search concepts combined “human papillomavirus” or “HPV” with lesion- and method-specific terms including cutaneous wart, plantar wart, Bowen disease, nail squamous cell carcinoma, condyloma acuminatum, vulvar intraepithelial neoplasia, anal intraepithelial neoplasia, penile intraepithelial neoplasia, squamous cell carcinoma, histopathology, genotype, p16, p53, Ki-67, E6/E7, mRNA, polymerase chain reaction, in situ hybridization, integration, and genomic sequencing.

Eligibility Criteria

Original human studies were eligible when they evaluated cutaneous or anogenital tissue and correlated histopathological diagnosis with at least one molecular parameter, including HPV DNA, viral genotype, E6/E7 RNA, p16, p53, Ki-67, viral integration, or host genomic alterations.

Studies limited to cervical, oral, or oropharyngeal disease without separately analyzable cutaneous or non-cervical anogenital data were excluded. Narrative reviews, systematic reviews, meta-analyses, isolated case reports, non-human experiments, purely in-vitro investigations, and treatment-only studies were also excluded.

Study Selection and PRISMA Flow

The search generated 120 records. Fourteen duplicate records were removed, leaving 106 unique records for title and abstract screening. Sixty records were excluded at this stage. Forty-six reports were retrieved and assessed in full text; none were unavailable. Sixteen full-text reports were excluded: secondary publications/reviews (n = 5), case reports or very small non-analytical series (n = 3), anatomical sites outside the predefined scope (n = 3), insufficient paired histopathological–molecular information (n = 2), non-human/experimental work (n = 1), overlapping populations (n = 1), and insufficient extractable results (n = 1). Thirty original studies entered the final qualitative synthesis.

The numerical reconciliation was 120 − 14 = 106 records screened; 106 − 60 = 46 full-text reports; and 46 − 16 = 30 included studies. No quantitative meta-analysis was performed.

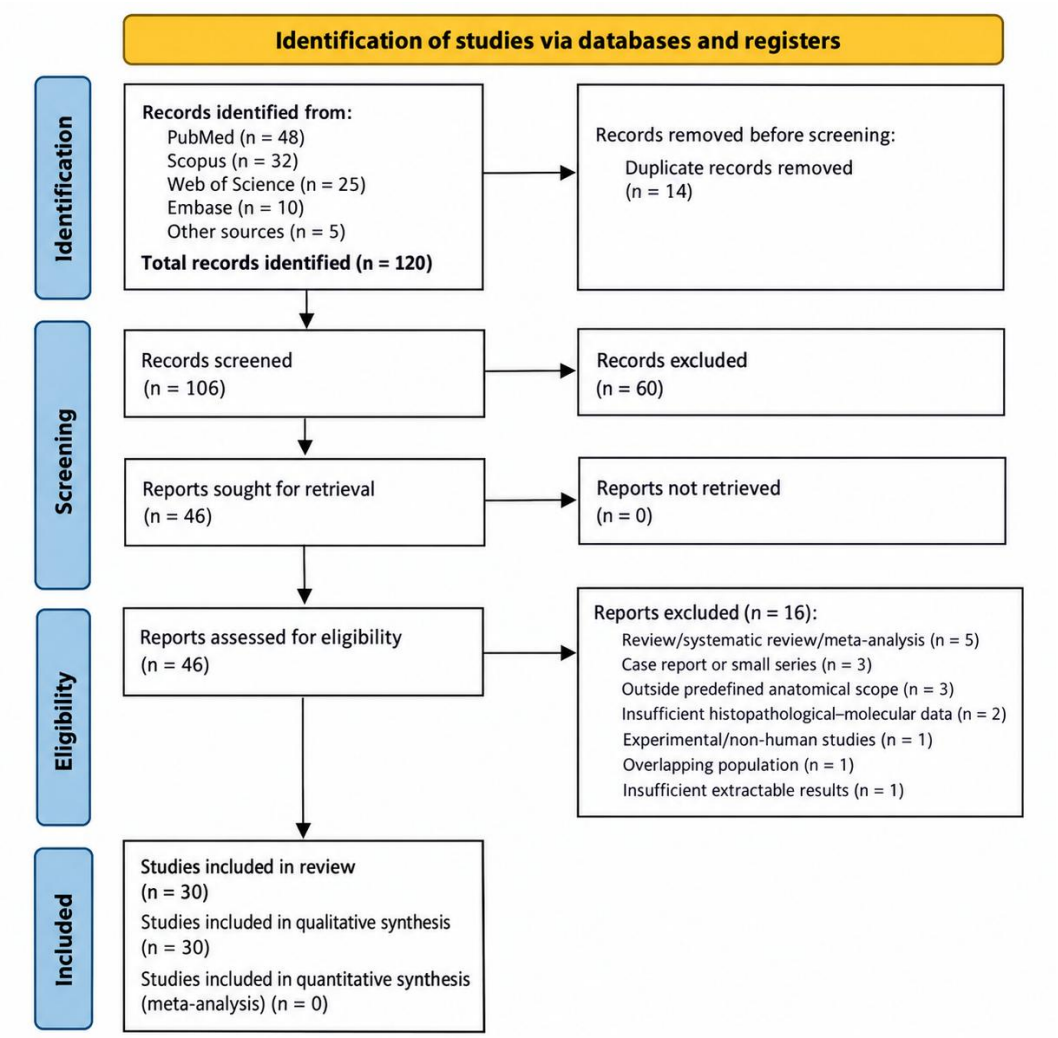

Figure 1. PRISMA 2020 flow diagram of study identification, screening, eligibility, and inclusion.

Data Extraction

Data were extracted on publication year, anatomical site, lesion type, sample size, histopathological findings, HPV detection method, genotype distribution, p16/p53/Ki-67 findings, E6/E7 transcription, viral integration, host genomic alterations, and the principal quantitative or biological conclusion.

Risk-of-Bias Assessment

Risk of bias was evaluated at the study level using a prespecified domain-based framework adapted from the Joanna Briggs Institute critical appraisal principles to accommodate the heterogeneous observational, diagnostic, and molecular-pathology designs included in this systematic review. Five domains were assessed: (1) selection and representativeness of the study population; (2) reliability and reproducibility of histopathological diagnosis and classification; (3) validity and appropriateness of HPV detection, genotyping, immunohistochemical, or other molecular methods; (4) control of relevant confounding factors and applicability of the study population to the review question; and (5) completeness and transparency of statistical analysis and reporting. Each domain was classified as low risk of bias, some concerns, or high risk of bias.

Overall risk of bias was considered low when all domains were judged to be at low risk or when only one minor methodological concern was present. Studies were categorized as having some concerns when limitations were identified in two or more domains but were unlikely to substantially invalidate the principal findings. A study was categorized as high risk of bias when major limitations in participant selection, histopathological ascertainment, molecular attribution, confounder control, or outcome reporting could materially influence interpretation of the results. No study was excluded solely on the basis of its risk-of-bias classification.

Methodological Assessment

Methodological quality was considered using study-design-appropriate domains derived from Joanna Briggs Institute appraisal principles. Particular attention was paid to case definition, representativeness of tissue selection, reproducibility of histological classification, appropriateness of viral assays, tissue localization of molecular findings, biomarker interpretation, and completeness of outcome reporting. No study was excluded solely because of methodological quality.

Data Synthesis
Because the included studies differed substantially in lesion type, population, HPV assay, genotype coverage, immunohistochemical threshold, and molecular endpoint, formal meta-analysis was considered inappropriate. Findings were synthesized by anatomical and biological category.

RESULTS
Study Characteristics
Thirty original studies were included. The evidence encompassed conventional cutaneous and plantar warts, high-throughput cutaneous HPV profiling, Bowen disease, cutaneous and nail-unit squamous cell carcinoma, anogenital keratotic lesions, anal intraepithelial neoplasia and carcinoma, vulvar intraepithelial and invasive disease, and penile intraepithelial and invasive carcinoma. Included publications ranged from 2005 to 2025.

Study	Lesion/site	Sample	Approach	Principal finding
Giannaki et al., 2013	Cutaneous warts in children	Not uniformly reported	HPV genotyping	HPV57 predominated; high-risk HPV16 also detected.
de Planell-Mas et al., 2017	Plantar warts	105 warts / 72 patients	HPV sequencing/genotyping	HPV57, 27, 1a and 2 were leading genotypes.
Skubic et al., 2022	Histologically confirmed common warts	128 warts; 126 evaluable	Histology + quantitative HPV typing	Causative HPV identified in 122/126; 12 types detected; HPV27 predominant.
García-Oreja et al., 2024	Plantar warts	Clinical cohort	HPV biotyping + follow-up	HPV prevalence 81.2%; HPV1 represented 36.1%.
Ekström et al., 2011	Cutaneous lesions	Pooled lesion material	High-throughput sequencing	Marked beta- and gamma-HPV diversity demonstrated.
Nili et al., 2024	Cutaneous/mucosal SCC	331 SCCs	Histopathological HPV features	Koilocytes in 15.1%; 38.1% in nail and 60.0% in genital lesions.
Murao et al., 2014	Bowen disease	Archival BD series	HPV DNA + p16/pRb/p53	HPV in 66.7% genital vs 8.3% extragenital lesions.
Moulart et al., 2024	Nail-unit SCC	48 cases; IHC in 36	Histology + p16/p53/Ki-67	Basaloid/periungual and keratinizing/subungual phenotypes; p16 positive in 31/36.
Pirog et al., 2010	Anal condyloma/AIN	Biopsy series	p16 + Ki-67 + HPV PCR	Diffuse p16 and abnormal Ki-67 characterized AIN2/3.
Salit et al., 2009	HIV-associated anal lesions	High-risk cohort	Genotype + viral load + p16 + E6	Multiple HPV types and HPV16 load associated with AIN2/3.
Wentzensen et al., 2012	Anal precursors in HIV-positive MSM	363 men	HPV DNA + E6/E7 mRNA + p16/Ki-67	E6/E7 mRNA showed strong discrimination for high-grade AIN.
Clarke et al., 2019	Prospective anal screening	Longitudinal cohort	Cytology + HPV + biomarkers	129 participants developed HSIL/AIN2+; RNA/genotyping improved specificity.
Frankart et al., 2021	Anal SCC	25 tumors	p16 + E6/E7 mRNA ISH	p16/E6-E7 concordance 96%; p16 PPV 100% in cohort.
Liu et al., 2020	HIV-associated anal HSIL	Clinicopathologic cohort	Histology + HPV subtype	HPV16 in 43%; non-16/18 high-risk types also frequent.
Sambo et al., 2025	Anal HPV in people living with HIV	Longitudinal cohort	HPV + SIL surveillance	Anal HPV prevalence 86.4%; dynamic incident/cleared infections.
Bonvicini et al., 2005	VIN/VSCC	41 specimens	Histology + ISH + PCR	Classic VIN frequently HPV-positive; differentiated VIN/keratinizing SCC HPV-negative.

Study	Lesion/site	Sample	Approach	Principal finding
Santos et al., 2006	Vulvar SCC	VSCC series	HPV + p16 IHC	p16 sensitivity 100% and specificity 98.7% for HPV-associated tumors.
Cheng et al., 2016	Vulvar SCC	201 invasive VSCCs	Morphology + p16 + HPV PCR	Morphology/p16 concordance 83%; PCR supported p16 in most discordant cases.
Tsimplaki et al., 2012	VIN/VAIN and SCC	56 histological specimens	HPV genotype + E6/E7 mRNA	HPV16 dominant; viral transcription more frequent in malignant lesions.
Pouwer et al., 2023	Vulvar SCC	255 patients	HPV PCR + p16 + p53	10-year local recurrence 24.6% HPV+ vs 47.5% HPV−.
Choschzick et al., 2024	Vulvar carcinoma	Pathology cohort	p16 + p53 + HPV RNA	Combined p16/p53 accurately classified HPV-associated and HPV-independent tumors.
Williams et al., 2020	Vulvar SCC	Genomic cohort	Comprehensive genomic profiling	HPV+ and HPV− tumors had distinct actionable genomic alterations.
Van Arsdale et al., 2024	Vulvar carcinoma	13 cancers	HPV capture sequencing + RNA-seq	HPV DNA in 5/13; integrated HPV16 in 3 tumors with E7/E6*I transcription.
Fernández-Nestosa et al., 2017	Penile condyloma/PeIN/SCC	191 lesions / 43 patients	LCM-PCR + p16	Genotype-morphology correlation supported distinct HPV-driven pathways.
Mannweiler et al., 2013	Penile SCC	123 SCCs + 43 precursors	HPV + p16 + p53	72/123 SCCs p16-overexpressing; HPV-independent lesions often p53-positive.
Stankiewicz et al., 2011	Penile SCC	PSCC cohort	HPV + RB/p16/p21/p53	HPV disrupted RB/p16 pathway; HPV16 dominated positive tumors.
Trias et al., 2024	Penile SCC	122 patients	HPV ISH + p16 + p53	Disease-related death 8%, 0%, and 27% across HPV-associated, HPV-ind/p53-normal, HPV-ind/p53-abnormal groups.
Matos do Canto et al., 2022	HPV-associated penile SCC	30 tumors	Whole-exome sequencing	HPV16 in 75%; recurrent NOTCH1, FAT1, TP53, CDKN2A, CASP8 and other alterations.
Othman et al., 2022	Anogenital lesions	125 FFPE cases	Molecular HPV detection	HPV detected in 72%; HPV6 and HPV16 predominant low-/high-risk types.
Seong et al., 2022	Genital keratotic lesions	Comparative lesion cohort	HPV genotyping	Bowenoid papulosis/genital SK enriched for HR-HPV; condyloma for LR-HPV.

Risk-of-Bias Findings

Across the 30 included studies, 7 studies (23.3%) were judged to have a low overall risk of bias, 21 studies (70.0%) were considered to have some concerns, and 2 studies (6.7%) were categorized as having a high risk of bias. Thus, 28 of the 30 studies (93.3%) were considered to provide evidence with either low risk of bias or methodological limitations that were unlikely to invalidate their principal findings.

The most frequent methodological limitations were retrospective or convenience-based case selection, restricted study populations such as individuals living with HIV or patients treated at tertiary referral centers, small sample sizes in studies of uncommon tumors, and incomplete adjustment for potential confounding factors. Variability in HPV detection platforms, genotype coverage, and definitions of p16 positivity also reduced direct

comparability across studies. By contrast, histopathological ascertainment and molecular measurement were generally more robust in studies using standardized pathological criteria together with validated PCR, genotype-specific assays, E6/E7 RNA in situ hybridization, laser-capture microdissection, or next-generation sequencing.

Selection bias represented the most recurrent concern because several investigations used archival pathology material or highly selected clinical populations. This was particularly relevant to studies of anal HPV-associated lesions in HIV-positive individuals and molecular studies of rare vulvar, penile, and nail-unit carcinomas. However, these populations were clinically appropriate for the specific questions addressed and therefore did not necessarily compromise the internal validity of the observed histopathological–molecular associations.

Overall, the risk-of-bias assessment indicated that the evidence supporting the principal conclusions of this review was predominantly of acceptable methodological quality, although caution is warranted when generalizing prevalence estimates across anatomical sites or populations. Greater confidence was placed on consistent associations reproduced across several independent studies, particularly the association of block-type p16 expression with transforming anogenital HPV infection and the distinction between HPV-associated and HPV-independent vulvar and penile carcinogenic pathways.

Figure 2. Traffic-light risk-of-bias assessment of included studies.

Green (+) = low risk of bias; Yellow (?) = some concerns; Red (–) = high risk of bias.

#	Study	Selection / representativeness	Histopathological assessment	HPV / molecular methods	Confounding / applicability	Reporting / analysis	Overall risk of bias
1	Giannaki et al., 2013	?	+	+	?	+	?
2	de Planell-Mas et al., 2017	?	+	+	?	+	?
3	Skubic et al., 2022	+	+	+	+	+	+
4	García-Oreja et al., 2024	?	+	+	?	+	?
5	Ekström et al., 2011	?	?	+	?	+	?
6	Nili et al., 2024	?	+	–	?	+	–
7	Murao et al., 2014	?	+	+	?	+	?
8	Moulart et al., 2024	?	+	?	?	+	?
9	Pirog et al., 2010	?	+	+	?	+	?
10	Salit et al., 2009	?	+	+	?	+	?
11	Wentzensen et al., 2012	+	+	+	+	+	+
12	Clarke et al., 2019	+	+	+	+	+	+
13	Frankart et al., 2021	?	+	+	?	+	?
14	Liu et al., 2020	?	+	+	?	+	?
15	Sambo et al., 2025	?	+	+	?	+	?
16	Bonvicini et al., 2005	?	+	+	?	+	?
17	Santos et al., 2006	?	+	+	?	+	?
18	Cheng et al., 2016	+	+	+	+	+	+
19	Tsimplaki et al., 2012	?	+	+	?	+	?
20	Pouwer et al., 2023	?	+	+	?	+	?
21	Choschzick et al., 2024	+	+	+	+	+	+
22	Williams et al., 2020	?	+	+	?	+	?
23	Van Arsdale et al., 2024	?	+	+	?	+	?
24	Fernández-Nestosa et al., 2017	+	+	+	+	+	+
25	Mannweiler et al., 2013	?	+	+	?	+	?
26	Stankiewicz et al., 2011	?	+	+	?	+	?
27	Trias et al., 2024	+	+	+	+	+	+
28	Matos do Canto et al., 2022	?	+	+	?	+	?
29	Othman et al., 2022	?	?	?	–	?	–
30	Seong et al., 2022	?	+	+	?	+	?

Overall assessment: 7 studies low risk (23.3%), 21 studies with some concerns (70.0%), and 2 studies high risk (6.7%).

Risk-of-bias judgments were based on five domains: selection and representativeness, histopathological assessment, validity of HPV/molecular methods, confounding/applicability, and completeness of reporting and statistical analysis. The framework was adapted from Joanna Briggs Institute critical appraisal principles to accommodate the heterogeneous observational and molecular-pathology designs included in the review.

Summary of risk-of-bias assessment

Of the 30 included studies, 7 studies (23.3%) were judged to have a low overall risk of bias, 21 studies (70.0%) had some concerns, and 2 studies (6.7%) were considered to have a high risk of bias.

The most frequent concerns were related to retrospective or convenience-based case selection, relatively small sample sizes in studies of uncommon lesions, population-specific recruitment, and incomplete control of potential confounding factors. Variation in HPV detection platforms, genotype coverage, and criteria used to define biomarker positivity also limited direct comparison between studies. Histopathological diagnosis and molecular measurement were generally reliable, particularly in studies employing standardized pathological

criteria, validated PCR or genotyping assays, E6/E7 RNA analysis, laser-capture microdissection, or genomic sequencing.

Cutaneous HPV Lesions

Common and Plantar Warts

The cutaneous wart studies confirmed substantial genotype heterogeneity. Skubic et al. identified a causative HPV type in 122 of 126 evaluable histologically confirmed common warts (96.8%), with 12 distinct types represented and HPV27 the most frequent [3]. In plantar lesions, de Planell-Mas et al. found HPV57, HPV27, HPV1a, and HPV2 among the principal genotypes [2].

García-Oreja et al. reported an overall HPV detection rate of 81.2% in a contemporary plantar-wart cohort; HPV1 accounted for 36.1% and was particularly common in younger patients. HPV biotype also correlated with wart number, spontaneous resolution, recurrence, and healing time, suggesting that genotype contributes to clinical behavior as well as morphology [4].

High-throughput sequencing further expanded the recognized cutaneous HPV spectrum. Ekström et al. demonstrated extensive beta- and gamma-papillomavirus diversity in cutaneous lesions, emphasizing that targeted PCR captures only part of the cutaneous virome and that detection of low-copy HPV sequences does not automatically establish causal significance [5].

Cutaneous Squamous Lesions and Bowen Disease

In a retrospective series of 331 SCC specimens, Nili et al. identified koilocytes in 50 lesions (15.1%). The proportion was considerably higher in nail lesions (38.1%) and genital lesions (60.0%), and koilocytosis was particularly frequent in SCC in situ [6]. These findings support a site-dependent relationship between HPV-type cytopathic morphology and squamous neoplasia.

Bowen disease also showed a strong anatomical gradient. Murao et al. detected HPV DNA in 66.7% of genital Bowen lesions but only 8.3% of extragenital lesions. Although p16 was expressed in 88.6% of cases overall, p16 did not reliably separate HPV-positive from HPV-negative Bowen disease. Negative p53 expression showed the stronger association with HPV-positive disease [7].

Nail-Unit Squamous Cell Carcinoma

Moulart et al. evaluated 48 nail-unit SCCs, with immunohistochemistry available for 36. Two morphological patterns emerged: a blue-basaloid pattern associated with koilocytic change, periungual location, and predominantly in-situ disease, and a pink-keratinizing pattern more frequently associated with subungual invasive tumors. p16 was positive in 31 of 36 cases, including both basaloid and keratinizing tumors, demonstrating that p16 alone cannot establish HPV causation in nail SCC; morphology and site remain essential interpretive context [8].

Anogenital HPV Lesions

Anal Intraepithelial Neoplasia and Anal Squamous Cell Carcinoma

Anal lesions demonstrated a progressive shift from productive infection toward transforming biomarker patterns. Pirog et al. found abnormal Ki-67 labeling in condyloma and AIN, while AIN1 remained negative for block-type p16. All AIN2 and AIN3 lesions in their series showed abnormal Ki-67 together with diffuse, moderate-to-strong p16 expression [9].

Salit et al. reported that the number of HPV genotypes was greater in AIN2/3 than in normal/AIN1 histology (median 5 versus 3.5 types; $P = 0.0005$). HPV16, HPV18, HPV45, and HPV52 were prominent oncogenic types, and HPV16 viral load correlated with high-grade disease [10].

Wentzensen et al. studied 363 HIV-positive men who have sex with men and found increasing positivity for HPV DNA, E6/E7 mRNA, and p16/Ki-67 with histological severity. Broad HPV DNA testing provided the highest sensitivity, whereas E6/E7 mRNA produced a more favorable balance of sensitivity and specificity for high-grade AIN [11]. In a prospective cohort, Clarke et al. documented 129 participants who developed HSIL/AIN2+ and showed that HPV16/18 genotyping and E6/E7 mRNA improved specificity relative to broad high-risk HPV testing [12].

At the invasive stage, Frankart et al. reported 96% concordance between p16 and E6/E7 mRNA in anal SCC; p16 achieved a 100% positive predictive value for transcriptionally active HPV in that cohort [13]. Among people living with HIV, persistent burden remained substantial: Sambo et al. reported anal HPV prevalence of 86.4%, underscoring the impact of immunosuppression on persistence and lesion development [15].

Vulvar Intraepithelial Neoplasia and Vulvar Squamous Cell Carcinoma

Bonvicini et al. studied 41 vulvar specimens comprising classic VIN, differentiated VIN, and invasive keratinizing SCC. Classic VIN was HPV-positive in approximately 66.1% of samples, with HPV16 accounting for 81.8% of positive lesions. In contrast, all differentiated VIN and invasive keratinizing SCC specimens in that series were HPV-negative, supporting a distinct HPV-independent pathway [16].

Santos et al. found HPV in 17.4% of vulvar SCCs and HPV16 in 75% of HPV-positive tumors. All HPV-positive tumors were p16 positive, compared with only 2.3% of HPV-negative carcinomas; p16 sensitivity and specificity for HPV-associated VSCC were 100% and 98.7%, respectively [17]. Cheng et al. similarly demonstrated that morphology and p16 agreed in 83% of 201 invasive VSCCs; among discordant cases, HPV PCR agreed with p16 in 32 of 34 cases, validating p16 as a more reliable etiological surrogate than morphology alone in selected tumors [18].

Tsimplaki et al. detected HPV DNA in 71.4% of VIN and 50% of vulvar SCC. E6/E7 mRNA was present in 42.9% of VIN and 83.3% of vulvar SCC, with HPV16 dominant, illustrating the distinction between viral presence and transcriptionally active oncogenic infection [19].

More recent studies refine the HPV-independent category. Pouwer et al. found HPV PCR negativity in 211 of 255 patients (83.5%); the 10-year local recurrence rate was 24.6% for HPV-positive tumors versus 47.5% for HPV-negative tumors, with the association persisting after multivariable adjustment (HR 0.23; 95% CI 0.08–0.62; P = 0.004) [20]. Choschzick et al. reported p16 sensitivity/specificity of 100%/96% for HPV association and p53 sensitivity/specificity of 95%/90% for TP53-mutated disease, supporting combined p16/p53 classification [21].

Genomic Findings in Vulvar Carcinoma

Genomic studies indicate that HPV-positive and HPV-negative vulvar carcinomas differ beyond viral status. Williams et al. identified distinct genomic landscapes and potentially actionable alterations in the two groups [22]. Van Arsdale et al. detected HPV DNA in 5 of 13 vulvar carcinomas and integrated HPV16 in three HPV-positive tumors; integration involved intronic regions including NCKAP1, C5orf67, and LRP1B. The integrated viral DNA retained transcriptionally active E7 and E6*I, directly linking integration with persistent oncogene expression [23].

Penile Intraepithelial Neoplasia and Penile Squamous Cell Carcinoma

Laser-capture microdissection has clarified genotype–morphology relationships in penile lesions. Fernández-Nestosa et al. analyzed 191 lesions from 43 patients and demonstrated strong HPV associations with condyloma, warty/basaloid PeIN, and warty/basaloid carcinoma, while differentiated PeIN and low-grade keratinizing lesions were far less frequently HPV-associated. HPV16 was strongly linked to basaloid PeIN, whereas warty PeIN showed broader genotype heterogeneity [24].

Mannweiler et al. identified p16 overexpression in 72 of 123 invasive penile SCCs. Sixty-six of the 72 p16-overexpressing tumors contained a single high-risk HPV genotype, predominantly HPV16. In contrast, 51 tumors lacked p16 overexpression and frequently showed nuclear p53 expression, supporting a separate HPV-independent pathway [25].

Stankiewicz et al. detected HPV DNA in 56% of penile SCCs, with HPV16 present in 81% of HPV-positive tumors. Basaloid carcinomas were strongly associated with HPV, while verrucous tumors were much less frequently positive. HPV correlated with retinoblastoma pathway disruption and p16/p21 expression but not with p53 immunodetection [26].

Trias et al. further stratified 122 penile SCCs into HPV-associated (36/122, 29%), HPV-independent/p53-normal (35/122, 29%), and HPV-independent/p53-abnormal (51/122, 42%) groups. Disease-related death occurred in 8%, 0%, and 27%, respectively (P < 0.001), and HPV-independent/p53-abnormal disease was independently associated with impaired disease-specific survival [27].

Whole-exome sequencing by Matos do Canto et al. in 30 HPV-associated penile SCCs identified HPV16 in 75% and recurrent alterations involving NOTCH1, TERT, FAT1, TP53, CDKN2A, CASP8, FBXW7, and chromatin-remodeling genes. These findings show that viral oncogenesis coexists with extensive host genomic evolution [28].

Cross-Site Molecular Interpretation

Lesion	Typical histopathology	HPV pattern	Most informative adjunct
Common/plantar wart	Papillomatosis, acanthosis, hypergranulosis, koilocytosis	HPV1/2/27/57 and related cutaneous types	Genotyping mainly for research/atypical cases
Bowen disease	Full-thickness squamous atypia	Strong site dependence	HPV DNA with p16/p53 interpreted in context
HPV-associated nail SCC	Basaloid/koilocytic; often periungual	High-risk alpha-HPV phenotype	Morphology + HPV assay; p16 supportive but not definitive
Anal LSIL/AIN1	Productive change with retained maturation	Multiple HPV types	Ki-67; p16 usually non-block
Anal HSIL/AIN2-3	Loss of maturation; suprabasal mitoses	High-risk HPV, often HPV16	Block p16, Ki-67, E6/E7 RNA
HPV-associated VSCC	Often basaloid/nonkeratinizing but variable	HPV16 dominant	p16 ± HPV RNA
HPV-independent VSCC	Often keratinizing	HPV negative	p53 pattern/TP53 context
HPV-associated penile SCC	Basaloid/warty	HPV16 dominant	p16 + HPV assay

Lesion	Typical histopathology	HPV pattern	Most informative adjunct
HPV-independent penile SCC	Differentiated/keratinizing spectrum	HPV negative	p53-based stratification

DISCUSSION

The central finding of this systematic review is that HPV-associated pathology is better understood as a collection of site-specific biological processes than as a single continuum from wart to carcinoma. The same viral family produces productive cutaneous infection, transforming anogenital neoplasia, and site-specific carcinomas with substantially different relationships between viral detection and biological dependence.

Productive cutaneous infection is characterized by viral replication within differentiating keratinocytes and broad genotype diversity. Even clinically similar common and plantar warts can arise from different HPV types, and high-throughput sequencing reveals substantial beta- and gamma-HPV diversity [1–5]. This background prevalence complicates causal interpretation when HPV DNA is detected in cutaneous tumors.

Histomorphology therefore remains indispensable. In cutaneous SCC and nail-unit carcinoma, anatomical site and microscopic pattern can be more informative than a single biomarker. The nail-unit data are particularly instructive: p16 was positive in both basaloid and keratinizing tumors, so p16 positivity alone did not define HPV-driven disease [8]. Similarly, Bowen disease showed frequent p16 expression even when HPV DNA was absent [7].

The lower anogenital tract displays a more coherent transformation model. High-grade anal disease showed progressive expansion of Ki-67 and block-type p16, while E6/E7 mRNA improved specificity for biologically active infection [9–13]. This reinforces the principle that HPV DNA positivity is not equivalent to HPV-driven neoplasia; transcriptional activity and downstream pathway effects are more informative.

Vulvar carcinoma illustrates the limits of morphology-only classification. HPV-associated tumors may be keratinizing, and HPV-independent tumors may occasionally display patterns that mimic HPV-associated disease. p16 substantially improves etiological classification, while p53 adds resolution within HPV-independent disease [16–21]. The emerging molecular framework therefore integrates morphology, p16, p53, and—where needed—HPV RNA or sequencing.

Penile carcinoma shows a comparable dual-pathway model. HPV-associated tumors preferentially demonstrate p16 overexpression and retinoblastoma pathway disruption, whereas HPV-independent tumors frequently show p53 abnormalities. Recent data further separate HPV-independent/p53-normal from HPV-independent/p53-abnormal disease, the latter carrying worse clinical outcomes [24–27].

Genomic studies extend the model beyond simple HPV positivity. Viral integration can retain E6/E7 transcription, while HPV-associated vulvar and penile carcinomas continue to acquire somatic alterations in cell-cycle, differentiation, chromatin-remodeling, and signaling genes [22,23,28]. Thus, HPV-mediated oncogenesis and host genomic evolution are complementary rather than mutually exclusive processes.

The interpretation of the present findings should also consider the methodological quality of the underlying evidence. Although only 2 of 30 studies (6.7%) were judged to have a high overall risk of bias, most studies had at least some methodological concerns. These were primarily related to retrospective case selection, limited sample sizes, population-specific recruitment, variability in HPV assays, and incomplete adjustment for confounding variables. Accordingly, numerical HPV prevalence estimates should not be directly extrapolated between anatomical sites or patient populations. Nevertheless, the recurrence of similar histopathological and molecular relationships across independently conducted studies supports the robustness of the major qualitative conclusions.

Diagnostic Implications

- Routine benign cutaneous warts with classic morphology rarely require HPV genotyping.
- Atypical, persistent, periungual, genital, immunosuppression-associated, or dysplastic lesions merit broader histopathological and molecular evaluation.
- For anal HSIL and other lower-anogenital high-grade lesions, block-type p16 is a valuable adjunct when H&E findings are equivocal; Ki-67 can support proliferative dysregulation.
- HPV DNA testing is analytically sensitive but does not alone demonstrate transformation. E6/E7 RNA or an anatomically validated surrogate pattern provides stronger evidence of HPV-driven disease.
- For vulvar and penile SCC, combined p16 and p53 interpretation can help assign tumors to HPV-associated and HPV-independent molecular pathways.

Strengths

The review integrates benign cutaneous lesions, cutaneous and nail-unit squamous neoplasia, anal lesions, vulvar disease, and penile neoplasia in one pathology-focused framework. It also incorporates recent genomic evidence rather than limiting the synthesis to HPV prevalence, allowing direct comparison of H&E morphology, immunohistochemistry, PCR/genotyping, E6/E7 transcription, integration, and host genomic alterations.

Limitations

- Substantial heterogeneity existed across anatomical sites, viral assays, genotype panels, p16 thresholds, and outcome definitions.

- Several studies were retrospective and originated from tertiary referral or pathology archives.
- Anal studies frequently involved HIV-positive populations, limiting direct generalization to immunocompetent populations.
- Cutaneous HPV interpretation is difficult because normal skin can harbor multiple low-copy HPV types.
- Rare-tumor genomic series were relatively small.
- Because of these differences, formal meta-analysis and pooled diagnostic-performance estimates were not considered methodologically appropriate.

Future Directions

Future studies should combine standardized histopathological criteria with site-specific biomarker interpretation and direct measures of viral transcription. Spatial RNA methods may help determine whether HPV is present within the neoplastic clone rather than adjacent epithelium. Longitudinal studies are required to determine which molecular profiles predict progression of anogenital precursor lesions. For vulvar and penile carcinoma, integrated analysis of HPV status, p53, viral integration, mutational signatures, methylation, and immune microenvironment may provide more clinically meaningful classification than HPV status alone.

CONCLUSION

Human papillomavirus–associated cutaneous and anogenital lesions display marked histopathological and molecular heterogeneity. Benign cutaneous warts primarily represent productive infection and show broad genotype diversity. Conventional cutaneous squamous lesions have a less consistent causal relationship with HPV, while the nail unit is a distinctive site where an HPV-associated basaloid phenotype can occur. In the anogenital tract, high-risk HPV more consistently produces transforming molecular signatures, including p16 overexpression, altered proliferation, and E6/E7 transcription. HPV-independent vulvar and penile carcinomas represent separate pathways in which p53 is particularly informative. The most reliable diagnostic framework therefore integrates anatomical site, morphology, viral genotype or transcriptional status, and appropriately interpreted host biomarkers rather than equating HPV DNA positivity with HPV-driven neoplasia.

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