Chapter 5: Updating the natural history of HPV and anogenital cancer

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Abstract

The major steps in cervical carcinogenesis include infection of the metaplastic epithelium of the cervical transformation zone with one or more of the 12–18 carcinogenic types of human papillomavirus (HPV) infection, viral persistence, clonal progression of the persistently-infected epithelium to cervical precancer, and invasion. Although these fundamental steps are established, several new epidemiologic studies have shed light on the factors that influence each of these transitions. The importance of the transformation zone in cervical cancer has been extended to other HPV-induced cancers such as anal or tonsillar cancers. Natural history studies show that HPV with normal cervical cytology and cervical intraepithelial neoplasia (CIN) grade 1 behave similarly, with the majority of both showing regression. Although these studies have demonstrated the importance of HPV persistence in the development of precancer CIN-3, the timing from infection to evidence of CIN-3 varies from 1 to 10 years. Whether equivalent lesions diagnosed later differ in their natural history remains unknown. Several factors have been implicated in enhancing persistence and/or progression. However, none are consistently associated with both except age: young women are less likely to show persistence and older women with persistence are more likely to be at risk of invasive cancer. Recent studies have also underscored the importance of the host immune response in clearance of established infections. Finally, data on non-cervical HPV infections, such as penile infections are limited to date compared to cervical infections. Several ongoing cohort studies should give us further insight into male infections in the near future.
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1. Introduction

Infection with HPV is necessary for the development of the vast majority of cases of cervical cancer and its immediate precursor (cervical intraepithelial neoplasia grade 3 or CIN-3). The major, sequential stages of HPV infection and cervical carcinogenesis are uniform regardless of population, thereby permitting a rational starting point for any discussion of optimal prevention efforts. Specifically, our improved understanding of the natural history translates into more accurate and stable transition-state models that are central to

health planning. However, these models remain affected by the technical limitations of HPV test methods, cytology, colposcopy, and histopathology which result in misclassifcation errors. The reader is referred to other chapters (7, 9 and 23) for discussion of these limitations.

2. Cervical carcinogenesis

The major steps in cervical carcinogenesis are shown in Fig. 1 before infection of the metaplastic epithelium of the cervical transformation zone with one or more carcinogenic types of HPV infection, viral persistence rather than clearance, clonal progression of the persistently-infected epithe-

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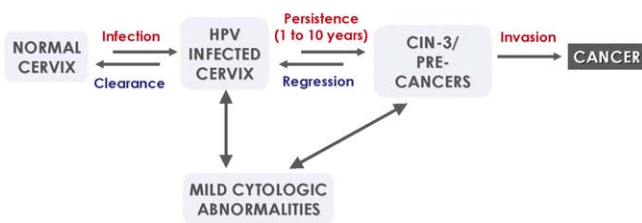## MAJOR STEPS IN CERVICAL CARCINOGENESIS

Fig. 1. Infection of the metaplastic epithelium of the cervical transformation zone with one of the carcinogenic types of HPV infection; this infection is either cleared quickly through either the innate immune system or other mechansisms. The majority of established infections which often manifest as microscopoic abnormalities are then either cleared at some point by host immune responses. Viral persistence leads to clonal progression of the persistently-infected epithelium and cervical intraepithelial neoplasia (CIN)-3/precancers arise; events which remain unknown lead infected cells to cervical invasion. Reprinted from [1] with permission from Oxford Press University.

lium to cervical precancer, and invasion [1]. Although these fundamental steps are established, each one merits discussion regarding subtleties and remaining questions regarding co-factors and host immune responses.

2.1. Importance of the metaplastic epithelium of the cervical transformation zone

For unknown reasons, HPV infection tends to cause cancer at areas referred to as "transformation zones". This is where one kind of epithelium contacts and gradually replaces another by literally transforming itself through a process referred to as metaplasia. The cervix, anus, and tonsils are examples of tissues with transformation zones prone to HPV carcinogenesis. In the case of the cervix, it is the area of columnar epithelium that transforms into squamous epithelium. The process in children is relatively dormant, whereas it becomes quite active around puberty. The zone of cervical HPV infection extends well into the vagina, but it is the area of cervical epithelium undergoing squamous metaplasia that is relevant to carcinogenesis. In order for the transformation zone to become infected with HPV, it is likely that sexual contact explains the majority of exposure routes. Although it is presumed that infection occurs through microscopic tears in the mucosal, other mechanisms may be in play specifically at the site of the cervical transformation zone.

2.2. Intra-epithelial microscopic abnormalities

The spectrum of HPV-related epithelial abnormalities leading to cancer was formerly considered a step-wise progression of increasingly severe intraepithelial neoplasia—grade 1 to grade 2 to grade 3 (including carcinoma *in situ*) to cancer. This histopathology concept remains very important in clinical management, but for epidemiologic and preventive research, it makes increasing sense to

consider HPV infections as a single broad transition state between normal and precancer. The most important viral characteristics are genotype and length of persistence. It is important to realize that the natural history of HPV that presents as equivocal lesions is not markedly different from infections that present as classical koilocytotic changes, or CIN-1 [2]. In fact, HPV type is arguably more important than the presence or absence of mild microscopic evidence of infection in the prediction of risk of cancer [3] because noncarcinogenic types also frequently cause cytologic and histologic abnormalities, whereas HPV type predicts outcome. Some argue that the probability that a microscopic abnormality will be found is more a function of diagnostic variability and error than a separate natural history stage [4].

For example, HPV infection might be thought of as two separate states: low viral load infections without evident abnormalities as compared to higher viral load infections with microscopically evident abnormalities [5]. Clinically this concept may be translated to explain why HPV detectable only by PCR is often associated with microscopic normalcy and with low risk of subsequent precancer/cancer, whereas HPV detected by less sensitive tests, such as the commercially available Hybrid Capture 2, is more likely to predict subsequent precancer development [6,7].

However they are viewed, microscopic abnormalities are diagnosed in only a small minority of women with HPV detectable by DNA assays. This fraction depends on the thresholds of the molecular and microscopic tests and HPV genotype [8], and can range widely from 1/10 to 1/3 in terms of relative prevalence in cross-sectional studies. In longitudinal studies among cytologically normal women who are HPV-DNA-positive, the cumulative risk of incident, including equivocal abnormalities, rises to 25–40% at 1–3 years following detection (Fig. 2) [9]. Low-grade
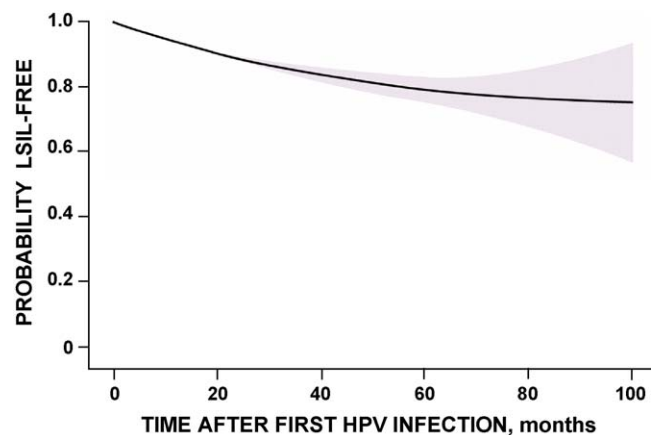

Fig. 2. Estimated distribution of time after first HPV infection participants remained free of low-grade squamous intraepithelial lesions (LSIL). Shaded area represents the 95% confidence interval. Reprinted from Moscicki AB, et al. JAMA 2001;285(23):2995–3002. Copyright ©American Medical Association. All rights reserved.

squamous intraepithelial lesion (LSIL) detection declines thereafter, returning to baseline after about 4 years [4,9]. Unfortunately, most of these studies predict cytologic abnormalities from baseline and do not account for other incident infections that most likely occur throughout the observed period.

2.3. Viral persistence versus clearance

Understanding the determinants of regional variation in age-specific HPV prevalence is an important remaining task that is tied to our need for a better understanding of viral persistence, clearance, and possible latency. HPV prevalence and incidence appears to peak in women under 20 years of age, with a decline noted in women 30 or older which is secondary to the clearance of HPV.

Cervical HPV infections (either with cytologic abnormality or not) tend to clear [5,10], as do warts anywhere on the body. The processes of HPV acquisition and clearance dynamically oppose each other in each cohort of women, to produce the characteristic age distributions, as infections are transmitted sexually when women have new partners and then cleared. To the extent that we can observe using existing cohorts, viral clearance is not often followed by reappearance of the same HPV genotype. When we see reappearance it is unclear whether these are new infections or resurgence of a poorly defined latent state. The high HPV prevalence among immunosuppressed HIV-positive individuals supports the proposal that a latent state of some kind exists [11]. These data are difficult to interpret since most women are diagnosed with HIV several years after HPV infection and many women in these studies did not stop having sex until the time of HIV diagnosis. Data from recently HIV infected adolescents show that even in women with normal CD4 counts, HPV persistence is prolonged compared to HIV-uninfected youth [5]. Consequently, clearance of any kind in this group may be impaired and not relevant to non-immunosuppressed groups. Possible additional evidence of re-activation of infection comes from populations in which prevalence rises secondarily among older women (see also Chapter 6). On the other hand, sexual behavior or cohort effects could explain secondary prevalence peaks [10].

Persistence can be broadly defined as detection of the same HPV type two or more times with a given time interval between the examinations. There is no agreed cut-off point between transience and persistence. The median time to clearance of prevalently detected and incidentally infected women is not very different in studies among very young women among whom all infections are relatively new, but the difference can be more important with age. The observed median time to clearance of prevalent infections ranges from four to six months to one to two years in different studies, depending on follow-up strategies and definitions (e.g., whether one or two negative tests are required to define clearance). Although time to clearance may vary between studies,

almost all show that approximately 90% clear a specific HPV type after two years of observation. When a specific HPV type is found consecutively, it is very likely to represent the same variant as well, thus suggesting true persistence and not sequential infections. The definition of HPV persistence differs from other viruses such as HIV or hepatitis B since, in contrast to these infections, most initially "persistent" HPV infections will eventually clear if followed long enough. In addition, length of observation is often truncated in studies when women develop CIN-2/3, although many of these lesions regress as well.

Some data suggest that HPV-16 persists longer, on average, than any other type [12]. Any other inter-type differences are subtle. Long-term persistence (5–7 years) is not strictly correlated with carcinogenicity since some noncarcinogenic types show long persistence as well (e.g., HPV-61) [12]. In population-based studies, HPV persistence shows the expected relationship with prevalence [12]. A central, critical understanding is emerging that a major determinant of HPV persistence is how long the infection has already lasted—the longer an HPV infection lasts, the more likely it is to last even longer. This might explain a variety of corollary observations. For example, infections in cross-sectional screening of older women might persist longer than in younger women because they are more likely to represent infections that are already persistent [13,14]. Clinical strategies requiring repeated abnormalities or repeated specific HPV detection lasting more than a year take advantage of this fact of natural history to sort out the transient infections and associated lesions from the persistent ones which pose the greatest risk to the patient [13,15].

2.4. Clonal progression of persistently infected epithelium to cervical precancer

"Precancer" is the intraepithelial precursor to invasive cancer. In scientific studies based on the notion of "surrogate end-points" for cancer, it is equivalent to the histologic diagnosis of CIN-3, which can be reliably distinguished from recently acquired HPV infection and it is a good indicator of subsequent cancer risk. In contrast, there is substantial heterogeneity in the microscopic diagnosis and biological meaning of CIN-2 lesions: some certainly represent acute HPV infections of particularly bad microscopic appearance that, however, are destined to regress, while others are incipient precancer and are destined to persist with high-risk of invasion. Some non-carcinogenic HPV infections are capable of producing lesions diagnosed as CIN-2, thereby showing that this level of abnormality is not a sufficient surrogate for cancer risk. High-grade CIN and carcinoma of the cervix are characterized by aneuploidy and genetic instability, and *in vitro* evidence shows that this is most likely a consequence of inappropriate high-risk (HR) HPV E6 and E7 expression with telomerase activation and centrosomal amplification: these changes provide a platform from which further mutations can arise and be selected [16].
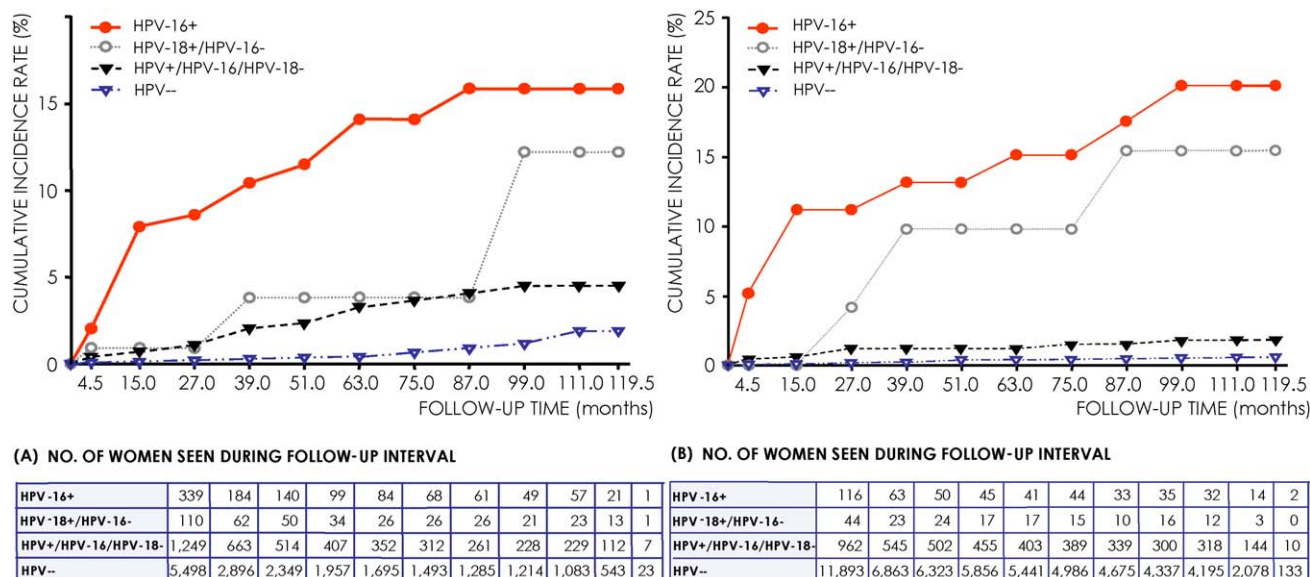
Fig. 3. Cumulative incidence of cervical intraepithelial neoplasia grade 3 and cancer (≥CIN-3) over a 10-year period in (A) 7285 women younger than 30 years of age and (B) 13,229 women 30 years old and older, according to oncogenic HPV status at enrollment. HPV status is defined hierarchically as: positive for HPV-16 (closed circles), else positive for HPV-18 (open circles), else positive for the non-HPV-16/18 oncogenic types in Hybrid Capture 2 (HC2) (closed triangles), else oncogenic HPV negative (open triangles). Reprinted from [3] with permission from Oxford University Press.

2.5. HPV Infection, persistence and risk of precancer diagnosis

It is the persistence of one of the carcinogenic types that is strongly linked to precancer (Fig. 3) [3,12]. As discussed above, incident HPV infection may or may not be associated with microscopic abnormalities. Studies, however, have found that only a fraction of precancers arise from HPV infections in the absence of diagnosed mild or even equivocal microscopically evident abnormalities [17]. This might also represent misclassification of cytology or histology or rapid transit through the mildly abnormal phase. It has also been posited that precancers develop in HPV-infected mucosa independent and adjacent (internal) to what is called CIN-1, rather than as an internal sub-clonal event [17].

Using cross-sectional data, the modal time between HPV infection and CIN-3 has been calculated to be 7–15 years with infection occurring in the later teens or early twenties and CIN-3 diagnosis peaking around 25–30 years of age (Fig. 4) [18]. However, prospective cohorts with careful, intensive follow-up are documenting rapid development of CIN-2 and -3, sometimes within a few months after incident infection (Fig. 5) [19]. The biological meaning of these "early" CIN-3 diagnoses is unknown. Although it is plausible that many of these would regress, observational studies of these lesions would be unethical. Long cohort studies covering the full period between average age at first sexual intercourse and average age of CIN-3 diagnosis are required to define the average lag-time between the two natural history stages as short cohorts find only the earliest cases.

2.6. Influence of HPV type, co-infection with multiple HPV types and HPV viral load

It is believed that HPV type affects both the absolute risk of viral persistence (discussed above) and of progression to precancer given viral persistence. HPV 16 appears to be remarkably carcinogenic with an absolute risk of CIN-3 approaching 40% at 5 years of persistence. In comparison, other HPV types have absolute risks several fold lower.

The amount of HPV-DNA in the cervical epithelium is a complex sum of the number, size, and state of the HPV-associated lesions. Low viral loads are associated with microscopic normalcy and with low risk of subsequent precancer/cancer, but in the clinical setting the prognostic importance of increasingly high viral loads is not at all established [6]. Some of the highest viral loads are associated with ultimately resolving CIN-1 and low-grade squamous intraepithelial lesions (LSIL), producing large amounts of virus analogous to benign warts. Cervical cancers do not produce large amounts of intact virus, and this is probably linked to the disruption of the HPV virion that precedes genomic integration.

Further complicating the measurement of viral load is the common occurrence of multiple HPV types due to sexual co-transmission. Cervical cancer is typically a monoclonal event related to a single HPV type. However, the surrounding cervical epithelium can still be infected with other types. As measured by sensitive DNA detection methods, more than 20–30% of women with cervical infections have more than one type, regardless of stage of pathology [5]. Because of great variability and uncertain interpretation, the role of viral
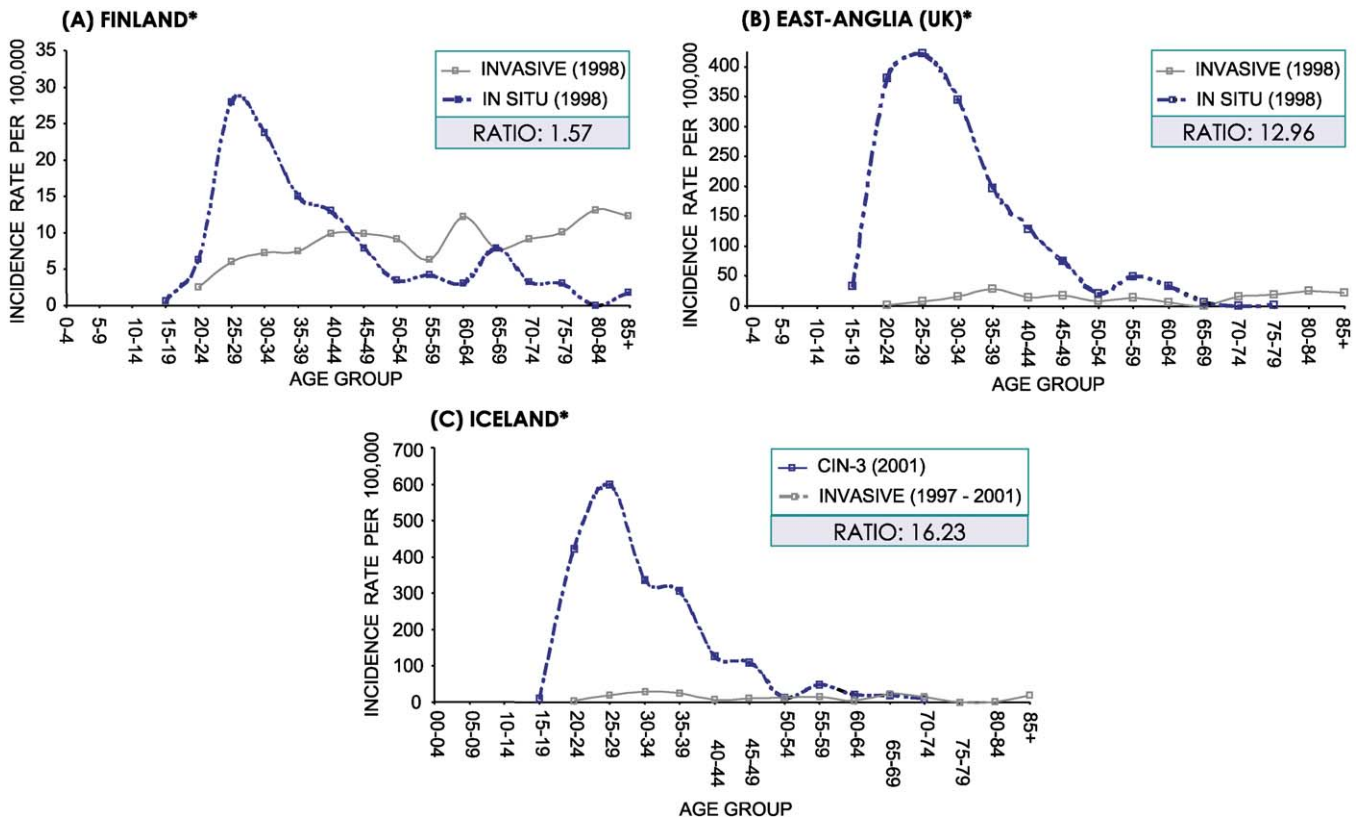
Fig. 4. Age-specific incidence rate of cervical intra-epithelial neoplasia (CIN)-3/carcinoma *in situ* and of invasive cervical cancer in selected populations. Ratio: number of cases of CIN-3/*in situ* carcinoma of the cervix over the number of cases of invasive cancer diagnosed in the same population over the same time period across all age groups. *Data provided courtesy of the Finnish Cancer Registry, East Anglian Cancer Registry, and the Iceland Cancer Registry. Reprinted from [18] with permission from Oxford Press University.

load in driving the natural history of HPV remains uncertain. In addition, it is still not clear whether infection with multiple HPV types interferes, either directly or immunologically, with the persistence of a given HPV type or with progression

[1,4,5]. Lastly, it also remains unclear to what extent variants of HPV types have different natural histories.

2.7. Invasive cervical cancer

CIN-3 lesions tend not to regress over short-term follow-up; however, the risk and timing of invasion versus eventual regression is probabilistic. While the median age of women with CIN-3 is 27–30 years, the median age of women with invasive cancers is skewed to much older ages. The median age of cancer moves toward even older ages as the quality of screening decreases. Even women with screen-detected cases of invasive cancer tend to be more than 10 years older, on average, than women with CIN-3, which suggests a long average sojourn time in the precancerous CIN-3 state (Fig. 4). Unfortunately, epidemiologic studies have not been able to suggest risk factors for invasion. Rapidly invasive, sometimes fatal cancers among young women are rare, but exert a profound influence on prevention strategies in many regions.

2.8. Cofactors for persistence and progression

Interpreting the literature on co-factors associated with persistence has been difficult. Given the predominating tran-
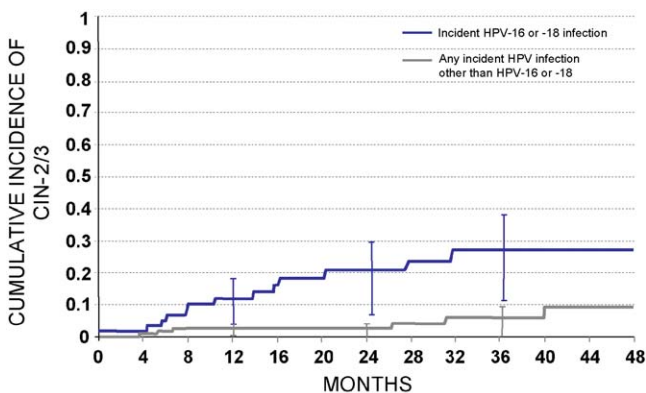

Fig. 5. Cumulative incidence of developing cervical intraepithelial neoplasia grade 2 and 3 (CIN-2/3) among women with incident HPV-16 or -18 infection (thick blue line; $n = 60$) or with any incident HPV infection other than HPV-16 or -18 (thin grey line; $n = 137$). The x-axis shows the no. of months from time of detection of first incident HPV-16 or HPV-18 infection or first infection with any type other than HPV-16 or HPV-18. Reprinted from [19] with permission from University of Chicago.
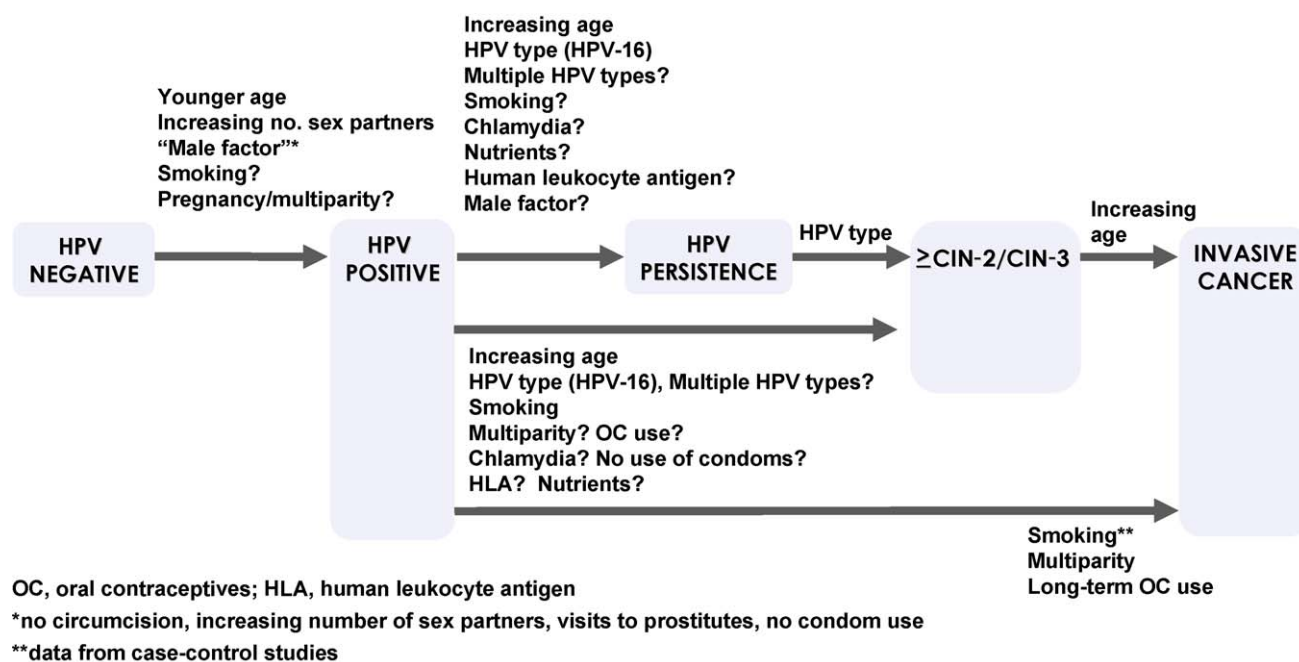
Fig. 6. Overview of factors most consistently reported to play a role at different stages in the natural history of HPV and cervical neoplasia.

sient nature of the infection, it is clear that other factors also play a role. However, it is not clear at which step(s) in the carcinogenic process these factors are most involved. Factors identified at the different steps are shown in Fig. 6.

Given the ubiquity of HPV, the most critical step in cervical carcinogenesis is not acquisition of the infection, but rather the step involving progression to clinically important lesions (i.e. CIN-3). There have been virtually no studies that try to separate risk factors for viral persistence from those for neoplastic progression. The two phenomena are not identical but thus far they are so closely linked that epidemiologists are only now beginning to disentangle them. It is believed that long-term persistence without progression is uncommon, but with the lack of exact knowledge about how to define transient, persistent, and long-term persistent HPV infection, it may still be valuable to try to assess risk factors for progression given persisting ("longer-lasting") HPV infection.

As the step from HPV positivity to ≥CIN-3 progression also includes the intermediate step to HPV persistence, there is a large overlap between the risk factors identified for HPV persistence and those found for progressing from HPV positivity (transient + persisting) to ≥CIN-3 (Fig. 6). These include, first of all, characteristics of the infection itself (HPV type, single/multiple infections, viral load) as discussed earlier. Multiple studies have identified different host factors and behavioral factors that play a role in the persistence of HPV and the risk of progression to precancerous lesions.

Studies assessing the risk of CIN-3 or cervical cancer among HPV-positive women have been consistent in finding smoking as a co-factor (see Chapter 1), and this has been confirmed in recent prospective studies [20,21]. In contrast, the association between smoking and persistence of HPV is less

consistent [22–24]. Thus, smoking seems to be an important co-factor, but it is not entirely clear at which stage in cervical carcinogenesis smoking interferes.

The findings related to oral contraceptive (OC) use as a co-factor for the development of high-grade cervical lesions among HPV-positive women are more equivocal [25]. None of the prospective studies have found an association between OC use and CIN-3 in HPV-positive women [4,24].

Early studies did not find an effect of condom use on HPV persistence, whereas one recent follow-up study found prolonged duration of HPV infection associated with infrequent condom use (below median level) during an HPV infection [26], and two other studies reported that condom use is related to decreased persistence of high-risk [23] and low-risk HPV types [24]. Similarly, two cohort studies have found that condom use reduces the risk for high-grade lesions among HPV-positive women [22,27], and, finally, it has been reported that condom use increases HPV clearance and CIN regression in women [28].

It has been suggested that other sexually transmitted diseases may act as co-factors for HPV. The role of *Chlamydia trachomatis* (CT) has been studied intensively, and the majority of studies restricted to HPV-positive women have demonstrated an association with high-grade cervical lesions and invasive cancer, as reviewed by Castle and Giuliano [29]. CT has also been reported to be associated with increased High-risk (HR) HPV persistence in two recent prospective studies [23,30]. In contrast, the findings related to other sexually transmitted infections like herpes simplex virus and *Trichomonas vaginalis* have been much less consistent [29].

Other factors like different nutrients (e.g. Vitamin E, lutein, lycopene, beta-carotene, and Vitamin C), intake of

fruit and vegetables, as well as alcohol intake have also been suggested as modulating the natural history of HPV/cervical neoplasia, but the results do not yet permit any firm conclusions.

3. The role of immunity in altering the natural history of HPV

Host immunity is a complex army of defense artillery that includes innate and adaptive immunity. It is generally thought that innate responses are responses that are relatively nonspecific to the pathogen. In the vaginal/cervical mucosal environment, these types of responses are extremely complex since the pool of nosocomial microorganisms that exist is immense. In addition, the innate system is responsible for triggering or activating adaptive immunity, which is exquisitely specific with memory. Antibody-mediated humoral immunity clears free virus particles and can prevent re-infection. Cell-mediated immune responses are important for clearance of viral-infected cells and generation of immune memory. Although, intuitively, the control of HPV should include all three, the evidence to date remains difficult to interpret since few studies employ these serological assays in natural history studies. In addition, the variability of target antigens and assays has contributed to the confusion.

3.1. Innate immunity and HPV

Since HPV particles are freed through normal desquamation processes, there is no cytopathic death involved. Hence, little inflammation occurs in which to trigger the innate immune system [31]. However, several lines of clinical and laboratory evidence show that innate infections are important in HPV control. For examples, women with transient HPV infections are less likely to develop antibody responses or cell-mediated responses than women with persistent infections [32] suggesting that the innate immune response can quickly eliminate antigens before there is a chance to develop memory responses. Emerging data underscore the complexity of the innate immune system which includes complex patterns of pathogen recognition (e.g. Toll-like Receptors), innate effector mechanisms (e.g., inflammation and chemotaxis), and the innate immune system's role as a bridge to adaptive immunity [33]. Studies are needed in epidemiologic cohorts to better understand these mechanisms as they relate to HPV persistence.

3.2. Measures of humoral response to HPV

Most studies support the notion that humoral responses to naturally occurring infections exert little protective effect against HPV persistence or disease. Rather, the development of HPV-specific antibody characterizes women by history of HPV exposure (see Chapter 12 and 23). As mentioned, some women most likely clear through innate mechanisms before there is adequate antigen exposure to develop adaptive immune responses. Among those who develop a response, there is a lag of several months that underscores the ability of HPV to evade the immune response for a period of time. The fact that recurrence of the same type after clearance is uncommon suggests, however, that humoral responses do give some protection [34].

3.3. Adaptive immune responses

The appearance of persistent HPV infection demonstrates that the innate system is often circumvented. The period between infection of the basal keratinocyte and the appearance of lesions is highly variable in natural history studies, thus demonstrating the ability of HPV to effectively evade the immune system for several months or even years [9]. There is relatively good clinical evidence that cell-mediated immune responses are critical in viral clearance after infection is established. Targets thought to be important in HPV control include the oncoproteins E6 and E7 and the gene E2 [31].

3.4. CD4 cell-mediated immune responses

Initial observations of an altered clinical course of HPV infection suggested that CD4 cells play a critical role in its natural history [35]. Although high rates of HPV persistence and squamous intraepithelial lesions are observed in HIV-infected women and men, the rate of invasive cancers among these individuals is much lower than expected. Also, HPV persistence and progression to HSIL has been shown to occur in HIV-infected young women with normal CD4 counts, which suggests [4] that immune factors other than CD4 cells play a role in HPV control.

Clinical studies using T-cell proliferation assays (CD4 functional assays) have given confusing results, with some showing association with clearance and others with persistence. Two recent studies [34,36] have found CD4 responses to E6 to be lacking in women with invasive cancer and recurrence of CIN after treatment.

3.5. CD8 cell-mediated immune responses

Evidence is mounting for a major role for MHC I-restricted CD8 CTL in modulating HPV infection and HPV-associated disease. In a prospective study of women with HPV clearance, cell-mediated immune (CMI) responses were found against HPV-16 E6 [37], and therapeutic vaccines have already begun to target CMI response to E6 and E7 [31].

HSIL and carcinomas are found to have a higher number of HPV genomes integrated into the host genome than LSIL, and integration of E6 and E7 promotes expression of these oncoproteins [38]. Studies have shown that CMI responses can be found in some women with long-term persistence but with normal cytology [31,37]. Since viral integration in this group has not been found [38], it may be that immune control

through anti-E6 and -E7 effects may stop integration or at least disease progression. Responses to L1 are not thought to be important in viral clearance once HPV infections are established [37], thus underscoring the nontherapeutic nature of the new vaccines based on L1.

3.6. Cytokines and CMI (the Th1/Th2 model)

The now well-established Th1/Th2 model of immunoregulation describes a phenotypic dichotomy among activated helper T (Th) cells according to their cytokine-producing profiles, and thus the type of "help" they deliver. Th type 1 (Th1) effector/memory cells produce predominately IFN-γ and promote CMI responses directed against intracellular pathogens; while Th2 cells produce IL-4, -5, and -13 (but not IFN-γ) and promote humoral responses. Several studies, including natural history studies [37] and cross-sectional studies of healthy women [34], have now shown that Th1 responses seem important in HPV control.

The mucosal immune complex is made up of not only what is considered innate and adaptive but also consists of locally secreted immunoglobins that are important in immune defense. Unfortunately, clinical studies of IGA and IGG and HPV control continue to show mixed results [39,40]. Although we assume that genetic influences on host responses to HPV are important to the natural history of infection, no specific host genetic marker, including HLA, has yielded strong and consistent results.

4. Noncervical HPV infections

Compared to the cervix, there are few reports on other anatomical sites, namely the vulva, vagina, penis, and anus, with the possible exception of studies of HPV in the anus of HIV-positive men and women [41]. This correlates with the fact that tumors in these sites are much rarer than in the cervix, with vaginal cancer and vaginal intraepithelial neoplasia being among the rarest of all female gynecological cancers.

The rarity of these cancers relative to cervical cancer also reflects the importance of the cervical and possibly anal squamocolumnar metaplastic epithelium because the observed carcinogenic HPV prevalence in all these anatomical locations is similar, ranging between 9 and 50% [41].

Concerning the penis, results should be interpreted with caution because they may be influenced by the site (shaft, urethra, foreskin) and collection procedure. Interestingly, the distribution of HPV types seems to be different, with HPV-82 being a common finding in penile smears while it is rare in cervical ones. Recent reports are exploring new possibilities for detection of HPV in men, including self-collected samples [42,43]. However, prospective studies of HPV infection and risk of penile intraepithelial neoplasia are just starting to generate data that are potentially important for the knowledge of the natural history of these infections in men. In a cohort

of young Danish men, HPV persistence was associated with having a high-risk type at enrollment, having multiple HPV types at enrollment, and being a current smoker. Short-term follow-up data show that 11–58% of men with HPV have persistence 8–12 months later [44,45]. In one study, circumcised men had a lower risk for persistent HPV infection than uncircumcised men [45]. The sexually transmitted nature of HPV infections between men and women underscores the need to consider them in effective cervical cancer prevention strategies, including HPV vaccine programs.

5. Future studies

Several questions remain concerning when CIN-3 develops after infection and whether CIN-3 lesions detected shortly after infection are similar to CIN-3 lesions diagnosed at a later stage. Since following these lesions is ethically questionable, other studies will be needed, such as affimetrix studies of cancer markers. Study of the role of immunity and genetics remains critical since the effect of the vaccine on CIN-3 and cancer will not be seen until two to three decades later. Several clinical studies have demonstrated the importance of immune control, and preventative vaccines aim the immune system towards humoral responses. The critical role of CMI in controlling established infections gives hope for therapeutic vaccines. After vaccines are implemented, it will also be important to see if the natural history of types other than HPV 16 and 18 are altered. If the local mucosal immune system is non-specifically stimulated by the vaccine to recognize other HPV-like pathogens, it may result in rapid regression of other HPV types.

Disclosed Potential Conflicts of Interest

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