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Permalink

Journal

ISSN

Authors
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Publication Date

DOI

Peer reviewed

Redetection of Cervical Human Papillomavirus Type 16 (HPV16) in Women With a History of HPV16

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Background. The purpose of this study was to examine the rate of and risks for cervical human papillomavirus type 16 (HPV16) redetection in women with documented or suspected HPV16 infection.

Methods. A convenience sample of women aged 13–21 years were seen at 4-month intervals for HPV DNA testing and cytology. Serum samples were obtained at baseline and annually.

Results. A total of 1543 women entered the study. Of the 295 women with detection of HPV16 DNA and subsequent clearance, 18.1% had HPV16 redetected by 8.5 years (88% cleared this second detection by 3 years). Of the 247 women who had antibodies to HPV16 and were HPV16 DNA negative at baseline, 15.3% had HPV16 redetected by year 5. Risks for redetection included douching, current use of medroxyprogesterone, reporting >1 sex partner or having a new sex partner, and having a sexually transmitted infection. Development of cervical intraepithelial neoplasia 2/3 was rare in women with redetection, except for those with prevalent HPV16 infection.

Conclusions. Reappearance of HPV16 DNA was observed in 18% of women. Most are associated with sexual exposure and appear benign. Interpretation of the studies is more complex in women with prevalent infections as it appears that this small subset reflects women with persistence already present at entry.

Keywords. redetection of HPV16; reactivation; risk for acquisition; risk for clearance.

Human papillomavirus (HPV), the cause of cervical cancer, is an extremely common infection of the cervix with a lifetime risk of approximately 80% [1]. The majority of HPV infections in women "clear" over time with few progressing to cancer. One of the remaining controversial questions is whether women truly "clear" an HPV infection, or if HPV goes into a latent state where redetection of infection reflects reactivation rather than a new exposure [2]. The prevalence of HPV

DNA in the genital tract peaks by 20–25 years of age followed by a decline to a plateau by 30–35 years of age. Data from Central and South America, but not North America, show a second, albeit lower, peak of HPV DNA prevalence around 50–55 years of age [3]. This second peak has been considered a possible reactivation of a prior HPV infection associated with a loss of immune memory response with age. However, this age period also reflects a common time when divorce occurs with the increased possibility of reexposure via a new sexual partner. Trottier et al [4] found that in adult women, redetection after known clearance of HPV type 16 (HPV16) was strongly associated with the report of new sexual partners. Certainly, the frequency of redetection after a documented clearance will likely vary with age of cohort, interval time between testing, length of follow-up, and definition of clearance [4–6]. In a group of women reporting recent sexual activity, redetection of the same HPV type occurred in 19.5% of

women within 1 year. However, this study used only 1 intervening negative HPV test to define clearance, which may have resulted in misclassification and risks for reinfection not being determined [7]. Studies using HPV serology as a marker of previous infection also demonstrate that 8%–13% of women have HPV16 DNA identified on follow-up. Estimates of infection based on serology are problematic, as many women never seroconvert [4, 6, 8].

The purpose of this analysis was to examine the rate of and risks for cervical HPV16 redetection in a cohort of women with evidence of HPV16 infection and clearance. Two groups were examined: (1) women with observed cervical HPV16 DNA detection and clearance and (2) women with serological evidence of previous HPV infection and clearance, defined as positive HPV serology and negative HPV16 DNA at entry. Risks for HPV16 DNA detection included sexually transmitted infections (STIs) and self-reported sexual behaviors. Rates of and risks for the clearance of the redetected HPV16 infections were also examined.

MATERIALS AND METHODS

Subject Population
Since its inception in 1990, 1559 women have been recruited into the University of California HPV natural history study. Recruitment of these women has been previously published [9–12]. Although follow-up of the cohort is ongoing, we report here on data collected through September 2007.

This study was approved by the University of California, San Francisco and San Francisco State University institutional review boards. Women were interviewed on sexual and substance use behaviors and examined at 4-month intervals [10, 11, 13]. Examinations included cervical samples for HPV DNA testing using cervical vaginal lavages with normal saline, cytology, and wet mounts for diagnosis of *Trichomonas vaginalis*, yeast, and bacterial vaginosis [9, 11–13]. Samples for *Chlamydia trachomatis* and *Neisseria gonorrhoeae* were obtained at annual visits or if symptomatic. Lesions suggested of HSV were tested at commercial laboratories.

HPV Testing
HPV DNA typing for cervical samples used the polymerase chain reaction (PCR)–based PGMY09/11 primer system as previously described using denatured biotin-labeled PCR product hybridized to an array of immobilized oligonucleotides [11, 12, 14]. Ongoing quality assurance (QA) shows a reproducibility of HPV16 DNA detection of 91%. Samples negative for HPV16 DNA sandwiched between first and second detection were reamplified and retested. At baseline and at annual visits, all women were asked but not required to have their blood drawn. For those with serum samples available at baseline, HPV16 serology testing was performed using an HPV16 L1 antibody binding assay, using glutathione S-transferase fusion proteins on a Luminex platform [15–19]. Testing was performed in the laboratories of 2 of the authors (M.P., D.A.G.).

Statistical Approach
We followed 2 analytic approaches in examining cervical HPV redetection. The first included all women (group 1) with either incident or prevalent (defined as by HPV16 DNA status at enrollment) HPV16 infection detected by HPV DNA testing, who also had at least 2 follow-up visits after HPV16 was detected. In these women, we first estimated the distribution of time to clearance as defined by 2 consecutive negative tests for HPV16 DNA, taking the initial positive visit as the time origin. Estimates were based on the Kaplan-Meier method. Among women observed to clear according to the above definition, we also estimated the distribution of time to next incident HPV16 DNA detection, taking the time of the first of the 2 consecutive negative tests as the time origin.

In our second analysis, we estimated the distribution of time to first detection of HPV16 DNA in women (group 2) observed to be HPV16 DNA negative at both baseline and the next consecutive visit but who were also seropositive for HPV16 antibodies—a surrogate marker for a previous HPV16 DNA infection. The baseline visit was the assumed time origin for this analysis. Because the sensitivity of HPV16 serology to detect all HPV16 infections is known to be low, we also estimated the analogous distribution in the HPV16 seronegative group [6]. Kaplan–Meier estimates were also used to summarize the cumulative probability of redetection and clearance of the redetection.

Two-sample *t* tests and χ^2 tests were used to evaluate differences in sociodemographic characteristics between women with prevalent and incident infections (group 1), between women with and without serology test results, and between seropositive and seronegative women (group 2). Crude redetection rates were estimated using person-time methods and expressed as the number of HPV16 redetection events per 1000 woman-years of observation. Confidence intervals (CIs) for crude redetection rates were calculated using the Poisson distribution. Cox proportional hazards regression models were used to examine associations between both fixed and time-varying predictors and HPV16 redetection and subsequent clearance. Candidate predictors for regression models with marginal associations significant at the 10% level or less were retained for further analyses. Variables of interest are listed in the corresponding tables. All models were adjusted for age, condom use, and, for group 1, HPV16 prevalence. Because of the low observed redetection rate, too few cases of clearance after detection were observed to allow for regression modeling. Only marginal associations are reported for this outcome. All analyses were repeated using 3 consecutive negative tests as a definition for clearance. Results were similar (data not shown).

Table 1. Demographics of Groups 1[a] and 2[b]

Demographic	Group 1 HPV16 DNA Positive[a] (n = 460)	Group 2	
		HPV16 DNA Negative/Seropositive[b] (n = 247)	HPV16 DNA Negative/Seronegative[b] (n = 640)
Age, y, mean ± SD	19.2 ± 2.25	19.76 ± 2.14[c]	18.93 ± 2.06[c]
Age of first sexual intercourse, y, mean ± SD	16.11 ± 1.98	16.18 ± 2.09	16.25 ± 1.91
Age of menarche, y, mean ± SD	12.71 ± 1.31	12.68 ± 1.35	12.53 ± 1.36
No. of lifetime partners, mean ± SD	6.66 ± 7.17	6.60 ± 5.94[c]	5.08 ± 6.64[c]
No. of days in study, mean ± SD	2038 ± 1476	2305.3 ± 1530.8[c]	1813.3 ± 1146.3[c]
Race[c]			
White	200 (43%)	111 (45%)	265 (41.4%)
Black	61 (13%)	37 (15%)	65 (10.2%)
Hispanic	100 (22%)	48 (19.4%)	158 (24.7%)
Asian	55 (12%)	31 (12.6%)	123 (19.2%)
Other	44 (10%)	20 (8.0%)	29 (4.5%)
Current smoker	143 (32%)	67 (28%)	199 (31.4%)
Drinks alcohol at least weekly	107 (24%)	46 (18.9%)	148 (23.4%)
Smokes marijuana at least weekly	70 (15.6%)	36 (14.8%)	108 (17.1%)

Abbreviations: HPV16, human papillomavirus type 16; SD, standard deviation.

[a] Group 1 were women with HPV16 DNA detection at baseline (prevalent) or during follow-up (incident).

[b] Group 2 were women who were HPV16 negative at baseline and were HVP16 seropositive or seronegative.

[c] Significant differences ($P < .01$) between seropositive and seronegative women in group 2.

RESULTS

A total of 1543 women completed a baseline visit. Supplementary Figure 1 demonstrates the number of women eligible for each of the analysis. None of the women received the HPV vaccine. Demographics of the cohort are described in Table 1 by statistical approach (group 1 and 2). Group 1 included 460 women: 250 with prevalent and 210 with incident HPV16 infections. Compared to women with incident infections, women with prevalent infections were less likely at baseline to smoke marijuana (11.7% vs 20%; $P = .01$), were slightly older (mean, 19.4 vs 18.9 years; $P = .01$), and had less follow-up (mean days in study, 1867 [SD, 1521] vs 2219 [SD, 1407]; $P < .001$). No other behavioral differences were found.

In the second analysis (group 2), 1293 women were cervical HPV16 DNA negative at baseline and the following consecutive visit. Of these women, 406 women refused a blood draw. However, women with serology available were more likely to have longer follow-up (1951.96 days [SD, 1284.8 days]) vs those who refused blood draws (mean days in study, 1096.71 [SD, 1034.0]; $P < .001$). No other behavioral differences were found. None of the women received the HPV vaccine.

Of the 887 women with serology, 247 (27.8%) were seropositive. Baseline demographics of the women are given in Table 1. Overall, characteristics of group 1 women and the seropositive women in group 2 were similar.

Rate of Cervical HPV16 Redetection in Group 1

Of the 460 women with a documented cervical HPV16 DNA infection, 52.9% (95% CI, 47.7%–58.2%) cleared within 1 year and 83.2% (95% CI, 78.3%–87.5%) cleared within 3 years. Of the 295 women who cleared and had follow-up, none had HPV16 DNA redetected within 1 year, 9.1% (95% CI, 6.0%–13.4%) within 3 years, and 18.1% (95% CI, 12.5%–25.7%) within 8.5 years. No differences were found for clearance or redetection between prevalent and incident cases (Figure 1A and 1B). The redetection of HPV16 DNA was 21.96 per 1000 women-years (Table 2).

Of the 33 women with redetection, 75.6% (95% CI, 58.0%–89.9%) of women cleared their second detection within 1 year and 87.8% (95% CI, 76.0%–96.9%) cleared within 3 years. No differences were found between incident and prevalent cases. (Figure 1C). However, if we defined clearance as no further detection of HPV16 DNA during follow-up, prevalent cases were less likely to clear than incident cases. The mean observation time for both groups was 9.2 years. Figure 1D shows that none of the incident cases that cleared their second detection had a reappearance, whereas 7.7% (95% CI, 1.1%–43.4%) of prevalent cases had a third detection by 3 years and 50.4% (95% CI, 22.8%–85.1%) by 12 years ($P = .04$).

Development of Cervical Intraepithelial Neoplasia 2/3

None of the 16 women with incident infection who experienced redetection (depicted in Figure 1C) developed
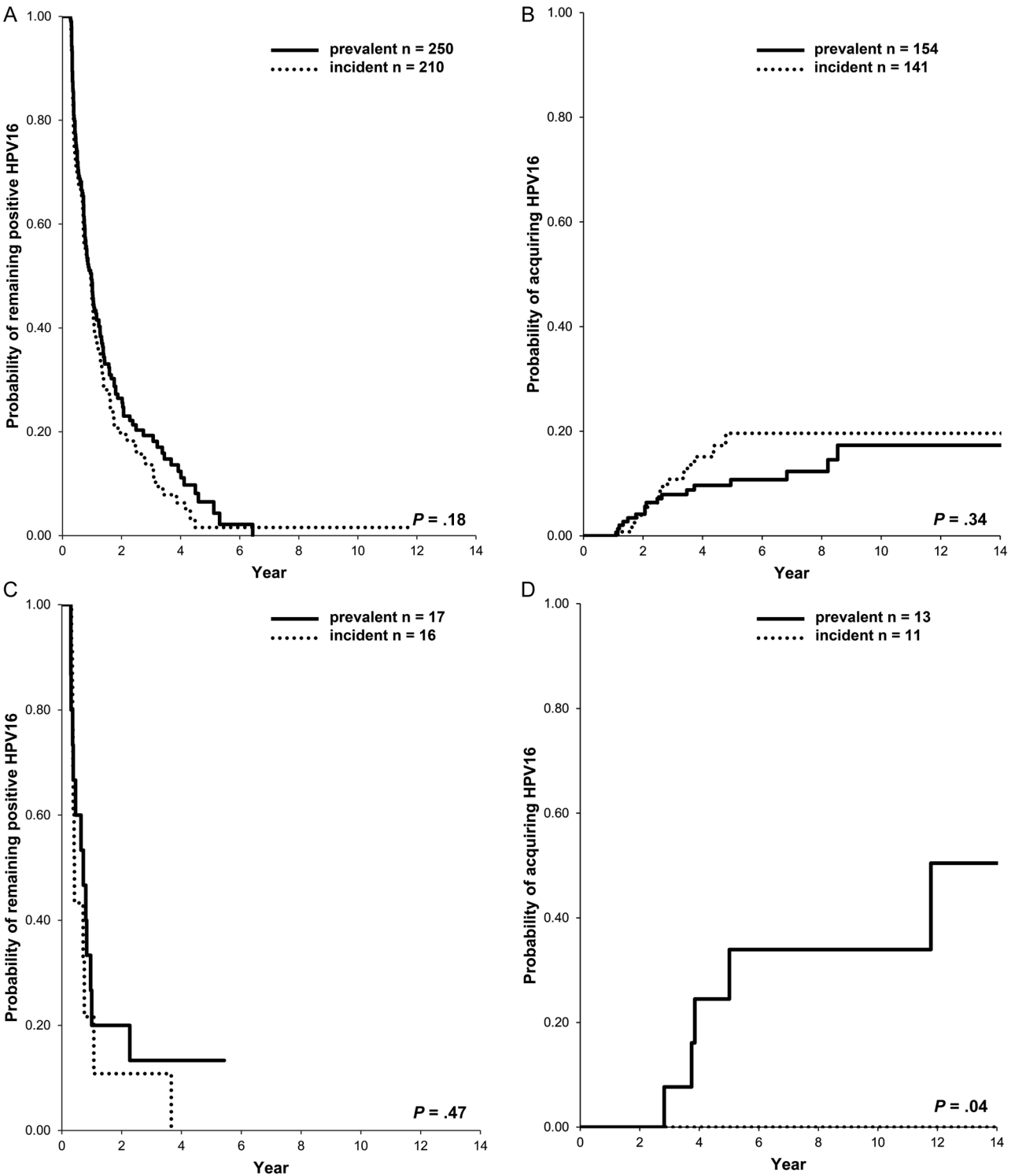
Figure 1. Among women with human papillomavirus type 16 (HPV16) DNA detection (group 1), either prevalent or incident, time to clearance (*A*), time to second detection of HPV16 DNA among women who cleared the virus (*B*), time to clearance of the second HPV16 DNA detection (*C*), and time to a third detection among women who cleared their second HPV16 DNA detection (*D*) are shown. Women with prevalent infections are noted by a solid line and incident infections by a dashed line. *P* values are based on log-rank test. Abbreviation: HPV16, human papillomavirus type 16.

HPV16-associated cervical intraepithelial neoplasia (CIN) 2/3. However, 1 woman developed HPV51-associated CIN 2/3

three years after her HPV16 clearance. Of the 17 women with prevalent infection who had redetection, 2 were immediately

Table 2. Redetection Rate of Human Papillomavirus Type 16 DNA per 1000 Women-years

Group	Women-years	Rate[a]	95% Confidence Interval
Group 1[b]	45 209	21.96	15.12–30.72
Group 2 seropositive[c]	5083	17.76	3.6–51.72
Group 2 seronegative[d]	9527	31.44	15.12–57.96

[a] Rate per 1000 women-years.

[b] Group 1 are women who are human papillomavirus type 16 (HPV16) DNA positive (prevalent or incident). Total n = 460; number with redetection = 33.

[c] Group 2 seropositive are women who were HPV16 DNA negative at baseline and HPV16 seropositive. Total n = 247; number with redetection = 34.

[d] Group 2 seronegative are women who were HPV16 DNA negative and HPV16 seronegative at baseline. Total n = 640; number with redetection = 117.

lost to follow-up at the time of their second HPV16 DNA detection, 8 cleared with no further detection, and 7 continued to be persistently or intermittently positive. Two of these women developed HPV16-associated CIN 2/3, 1 within 4 years after the second detection; the other women had been negative for HPV16 for 8 years and then developed CIN 2/3 within 2 years of the third detection—10 years after the second detection. Four women had normal cytology as of 5–12 years of follow-up. One case continued to be HPV16 persistent at the time she was lost to follow-up and had developed abnormal cytology, but because she was pregnant, no biopsy was obtained.

Rate of HPV16 DNA Detection Among Group 2 Women

Of the 247 women who were HPV seropositive at baseline but cervical HPV16 DNA negative, 3.3% (95% CI, 1.6%–6.8%) had cervical HPV16 detected by 1 year, 5.8% (95% CI, 3.0%–9.3%) by year 2, 7.5% (95% CI, 6.6%–12.2%) by year 3, and 15.3% (95% CI, 10.7%–21.6%) by year 5 (Figure 2A). In comparison, time to HPV16 detection was faster among the 640 women who were initially HPV16 DNA negative and HPV seronegative—2.4% (95% CI, 1.4%–4.0%) had an incident HPV16 by year 1, 8.5% (95% CI, 6.4%–11.3%) by year 2, 14.7% (95% CI, 11.9%–18.1%) by year 3, and 22.3% (95% CI, 18.7%–26.4%) by year 5 (Figure 2A; P = .03). Detection of cervical HPV16 DNA among the seropositive group was 17.76 per 1000 women-years compared to 31.44 per 1000 women-years among the HPV seronegative group (Table 2).

Among the 34 cervical HPV16 DNA detections in the seropositive group 2 women, time to clearance was similar to the 117 seronegative group 2 women, with 52.1% (95% CI, 43.7%–61.1%) clearing by year 1 and 86.0% (95% CI, 77.9%–92.3%) by year 3 (Figure 2B; P = .36). Of the 27 seropositive and 78 seronegative women who showed clearance, 0% had HPV16 redetected once more by year 1 and 9.4% (95% CI, 4.7%–17.9%) by year 3 with no difference noted by serostatus (Figure 2C; P = .41). Of the 13 women who had a second HPV16 DNA redetection, 76.6% (95% CI, 47.8%–96.1%) cleared within 1 year, and 100% cleared within 3.5 years (Figure 2D). The small sample size in this last group of women prohibits any statistical comparison.

CIN 2/3 Development Among Seropositive Women

Among the 34 seropositive women who acquired HPV16 (Figure 2A), none developed HPV16-associated CIN 2/3. In comparison, among the 117 HPV16 infections in the seronegative women, 4 developed HPV16-associated CIN 2/3.

### Risks for Redetection of Cervical HPV16

Table 3 describes the unadjusted and adjusted hazard ratio (HR) estimates for factors associated with risk of redetection of cervical HPV16 among women who had documented HPV16 DNA infection and clearance (group 1). Adjusted analysis found younger age of first intercourse (P = .07), history of douching since the last visit (P = .05), current use of medroxyprogesterone (P = .02), having >1 sex partner (P = .04), having a new sex partner (P = .02), and having a documented STI (P = .03) associated with HPV16 redetection.

Table 4 provides results for factors associated with cervical HPV16 DNA detection in women HPV16 negative at baseline but who were HPV seropositive (group 2). Adjusting for age and condom use, currently smoking cigarettes (P = .02) and current use of medroxyprogesterone (P = .002) remained significant with a trend for having >1 sexual partner in the past 8 months (P = .069). Condom use and having >1 sex partner were highly correlated (data not shown). When we adjusted for age only, having >1 sex partner remained significant (HR = 2.13 [95% CI, 1.03–4.42]; P = .04].

We also examined factors associated with acquisition of HPV16 among seronegative women for comparison. Factors associated with acquisition were similar to those found among women with documented HPV16 DNA infection and clearance (group 1) and are shown in Table 4. The risk of acquiring HPV16 was enhanced if other HPV types were also found at the visit with HPV16 detection. This was true for both groups. For group 1, the HR was 2.19 (95% CI, 1.08–4.45) and for group 2, 4.08 (95% CI, 2.95–5.63) if multiple types were present. HPV detection at the preceding visit did not increase the risk. Last, we examined factors associated with clearance of the second HPV16 detection in groups 1 and 2, which are summarized in Table 5. Factors that were associated with clearance with both groups included condom use and any sexual contact.

DISCUSSION

Among women with evidence of prior HPV16 infection and clearance defined via HPV DNA tests, only 4% were observed to have a second detection within 2 years. Subsequently, this
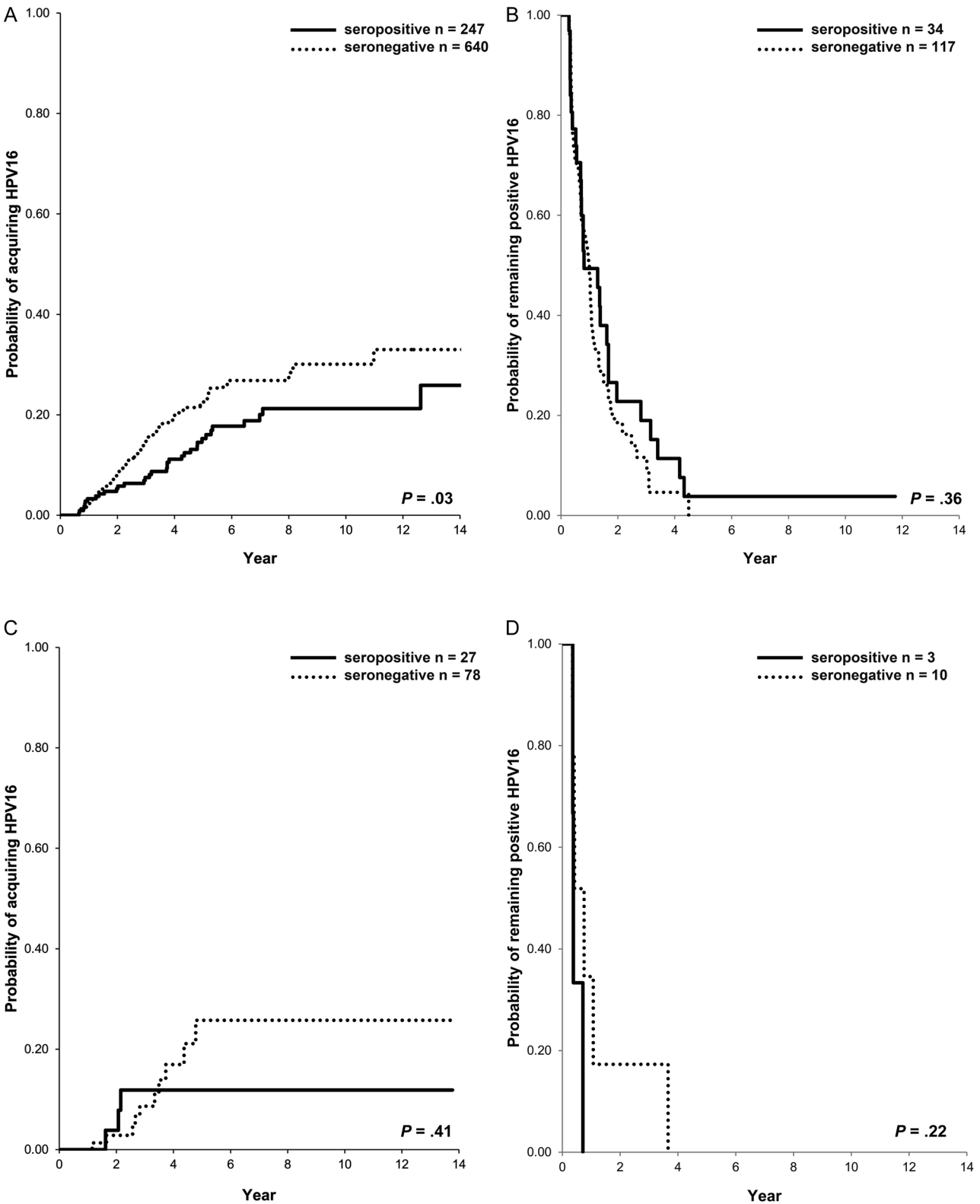
Figure 2. Among women who were human papillomavirus type 16 (HPV16) DNA negative at baseline but have evidence of a prior HPV16 infection by serology (seropositive), time to detection of HPV16 DNA (*A*) and time to clearance of these HPV16 infections (*B*) are shown. Among the women who cleared these incident HPV16 infections, time to another HPV16 infection (*C*) and time to clearance of these infections (*D*) are shown. For comparison, time to clearance and acquisition are shown in each panel for women who were HPV16 negative and HPV16 seronegative at baseline. Women HPV16 seropositive at baseline are noted by a solid line and seronegative by a dashed line. *P* values are based on log-rank test. Abbreviation: HPV, human papillomavirus type 16.

Table 3. Factors Associated With Redetection of Human Papillomavirus Type 16 (HPV16) DNA Among Women Who Had Evidence of HPV16 DNA Infection and Clearance (Group 1; n = 460)

Factor Associated With Redetection	Unadjusted HR (95% CI)	Adjusted[a] HR (95% CI)
Age of first sexual intercourse, per y	0.84 (.69–1.01)[b]	.82 (.66–1.02)[b]
Menarcheal age, per y	0.93 (.71–1.23)	
Age, per y	0.94 (.83–1.07)	
Weekly alcohol use, vs less than weekly	0.59 (.27–1.29)	
Weekly marijuana use, vs less than weekly	0.66 (.23–1.88)	
Condom use, always, vs less than always	1.08 (.49–2.39)	
Douched[c]	2.17 (1.00–4.72)[b]	2.35 (1.02–5.41)[d]
Anal sex[c]	1.66 (.64–4.33)	
Warts[c]	2.83 (.67–12.01)	
Yeast infection[c]	1.45 (.62–3.38)	
Pregnancy[c]	0.88 (.12–6.46)	
Currently smokes cigarettes	1.37 (.61–3.06)	
Current use of medroxyprogesterone[e]	3.95 (1.14–13.63)[d]	4.46 (1.23–16.17)[d]
Current use of combined hormonal contraception[e]	1.14 (.53–2.45)	
Reports polygamy in past 8 mo	2.23 (1.05–4.76)[d]	2.90 (1.22–6.90)[d]
STI[f] in past 8 mo	3.78 (1.13–12.64)[d]	4.03 (1.15–14.10)[d]
New sex partner in past 8 mo, per partner	1.1 (1.02–1.19)[d]	1.1 (1.02–1.19)[d]
Prevalent infection, vs incident	0.44 (.21–.92)	

Empty cells reflect nonsignificant hazard ratios in the model adjusting for age, condom use, and prevalent infection. Thirty-three women had redetection of human papillomavirus type 16 DNA.

Abbreviations: CI, confidence interval; HR, hazard ratio; STI, sexually transmitted infection.

[a] Variables were adjusted for age, condom use, and prevalent infection.

[b] *P* < .1.

[c] Reported since last visit: yes vs no.

[d] *P* < .05.

[e] Versus no hormonal contraception.

[f] Sexually transmitted infections include laboratory-documented *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and *Trichomonas vaginalis* infection.

gradually narrowed over time. This observation may be due to the fact that the cohort was aging and becoming more monogamous, so potential exposures in both groups were lessening, or that the seronegative group was becoming seropositive over time due to HPV16 exposures. Repeat serology was not performed. It is also possible that antibody response is a measure of a previous established infection but irrelevant to redetection. The rapid clearance of the observed redetections suggests that a cell-mediated immune response is most likely accountable for clearance in these second infections [9]. One of the striking findings was the difference in clearance for the second detection between the women with prevalent and incident HPV16 infections. Among women with incident infections, almost all cleared and more importantly, none developed HPV16-associated CIN 2/3. In contrast, those women who entered the study with a prevalent infection were more likely to persist or have recurrent detection than women with an incident infection, and 2 developed HPV16-associated CIN 2/3. This observation suggests that some of the women with prevalent infections are a different group and reflect persistent infections and had either missed detections or had intermittent shedding. Rodriguez et al [20] also found that prevalent infections were much more likely to result in CIN 3 than incident infections.

Similar to incident HPV16 infection in HPV16-naive women and our previous studies of HPV acquisition [13], sexual risk behavior was the predominant risk for HPV16 redetection in women with evidence of prior infection. The association with >1 male partner and new sexual partners with redetection demonstrates that most of the redetections were a consequence of a new exposure, similar to the conclusions reached by Trottier and colleagues [4]. A recent study by Theiler et al [21] showed that redetection of the same type in healthy women who reported abstinence was extremely rare with a detection rate of <0.1 per 100 women-years. Notably, having an STI also increased the risk of HPV16 redetection. STIs are certainly markers of partner risk behavior; however, STIs also create inflammation and decrease epithelial barriers, potentially exposing basal epithelial cells to infection with HPV [14]. The association with medroxyprogesterone is interesting as its use has been associated with the development of cervical cancer [22]. Potential mechanisms may include progesterone's ability to enhance E6 and E7 transcription and induce cellular proliferation [23]. Behaviors associated with birth control choice not measured in this study may have also influenced our finding.

The differences observed in risk for redetection between the seropositive group from group 2 and group 1 (ie, those with documented HPV16 DNA acquisition and clearance) is also worth noting. It has been demonstrated that not all women who acquire HPV16 infection seroconvert, and those with transient infections are least likely to seroconvert or develop memory immune responses [6, 24–27]. It is plausible to surmise that seroconversion is more likely to occur in women

rate remained relatively stable over time, with approximately one-fifth of women experiencing redetection within 8.5 years of follow-up. When we examined redetection rates in women with serologic evidence of prior infection, very similar rates were found. In contrast, in women with no evidence of prior HPV infection either by DNA tests or serology, rates of HPV16 detection were much higher, specifically within the first 2 years, suggesting that women with a previous infection had some type of immune protection [4, 6, 8]. On the other hand, the difference between the seropositive and seronegative groups

Table 4. Risk Factors for Acquisition of Human Papillomavirus Type 16 (HPV16) by HPV 16 Serostatus Among Women Who Were HPV 16 DNA Negative at Baseline (Group 2)

Risk Factor	Seropositive Unadjusted HR (95% CI), n = 247	Seropositive Adjusted[a] HR (95% CI)	Seronegative Unadjusted HR (95% CI), n = 640	Seronegative Adjusted[a] HR (95% CI)
Age of first sexual intercourse	1.2 (1.01–1.37)[b]		0.92 (.84–1.02)	
Menarcheal age	1.07 (.83–1.37)		1.13 (.98–1.29)[c]	
Age	1.14 (.98–1.35)		0.94 (.89–1.05)	
Weekly alcohol use	1.40 (.70–2.79)		1.50 (1.04–2.17)[b]	1.71 (1.08–2.72)[b]
Weekly marijuana use	NE[d]		1.94 (1.30–2.90)[b]	
Condom use, always[e]	1.48 (.70–3.11)		0.58 (.35–0.97)[b]	0.58 (.35–.97)[b]
Douched[e]	0.68 (.26–1.78)		0.72 (.35–1.48)	
Anal sex[e]	NE[d]		0.87 (.47–1.62)	
Warts[e]	1.30 (.17–9.65)		3.21 (1.40–7.34)[b]	3.75 (1.51–9.37)[b]
Yeast infection[e]	1.58 (.73–3.41)		1.28 (.80–2.07)	
Pregnancy[e]	1.71 (.40–7.28)		1.04 (.38–2.82)	
Currently smokes cigarettes	1.95 (.91–4.21)[c]	2.57 (1.15–5.74)[b]	1.36 (.92–2.02)	
Current medroxyprogesterone use[f]	4.53 (1.30–15.85)[b]	7.52 (2.05–27.56)[b]	1.39 (.51–3.82)	
Current use of combined hormonal contraception[f]	0.96 (.46–1.98)		0.89 (.59–1.36)	
New sex partner in past 8 mo	1.07 (.80–1.27)		1.07 (1.03–1.1)[b]	1.07 (1.03–1.1)[b]
STI[g] diagnosis in past 8 mo	NE[d]		2.81 (1.25–6.31)[b]	3.37 (1.23–9.20)[b]
Polygamy reported in past 8 mo	2.05 (.99–4.24)[c]	1.97 (.95–4.10)[c]	2.62 (1.77–3.88)[b]	2.64 (1.61–4.31)[b]

Empty cells reflect nonsignificant hazard ratios in the model adjusting for age and condom use. Thirty-four human papillomavirus type 16 (HPV16)–seropositive and 117 HPV16-seronegative women acquired HPV 16 DNA.

Abbreviations: CI, confidence interval; HR, hazard ratio.

[a] Variables were adjusted for age and condom use.

[b] $P < .05$.

[c] $P < .1$.

[d] Hazard ratio is not estimable due to very few events.

[e] Reported since last visit: yes vs no.

[f] Versus no hormonal contraception.

[g] Sexually transmitted infections include laboratory-documented *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and *Trichomonas vaginalis* infection.

with HPV persistence. One might hypothesize that redetection of HPV DNA in women who seroconvert is more likely to reflect HPV reactivation. The lack of association with new sex partners in the seropositive group may support this premise. On the other hand, the lack of association may have been due to the smaller sample size in this group, which is of older age and in which having a new sexual partner was less frequent than reporting >1 partner. The observed association with smoking is also interesting, as nicotine and its byproducts are known to suppress immune responses in the cervix and smoking is associated with high-risk sexual behavior [25, 28]. This suppression could result in lack of protection from reexposure or allow reactivation. It remains important to underscore that in either situation, the redetected infections were likely to clear rapidly and CIN 2/3 was uncommon. The association between lack of sexual contact and condom use and clearance of the redetected infections underscores the importance of partners in reinfection.

The rate of redetection in our study is difficult to compare to other studies, as length of follow-up and type of cohort influence this rate. In a longitudinal cohort study, Trottier et al [4] found a lower redetection rate for HPV16 of 1.05 per 1000 women-months. Because rates expressed as a function of person-time are dependent on length of follow-up, this difference may not be too surprising. Our total women-months of observation was almost 5 times longer than that reported in the study by Trottier et al [4]. In addition, their cohort was older at entry and may have had overall less exposure to new partners. Rodriguez et al [5] also found a lower recurrence rate than ours of only 3.7% over 7 years for any HPV type (HPV16 was not singled out). This difference is likely due to the nature of their cohort, which was population-based in Guanacaste, Costa Rica. Risk behaviors such as alcohol and marijuana use and multiple sexual partners were likely not as common in their cohort, and the population was also older.

Lack of association with new sex partners in the seropositive group likely underscores the limitations of serology. Wentzensen et al [6] used 2 different serologic methods to define seropositivity in a selected population from the Guanacaste study referred to earlier—one was a standard viruslike particle ELISA

Table 5. Factors Associated With Clearance of Second Detection of Human Papillomavirus Type 16 DNA

Factor	Group 1[a,b] HR (95% CI), n = 33	Group 2[c] HR (95% CI), n = 34
Condom use in past 60 d, always vs less than always	4.71 (1.39–16.29)[d]	2.92 (1.2–7.09)[d]
Any sexual contact, vs none[e]	0.06 (.01–0.3)[d]	0.42 (.18–.96)[d]
Menarcheal age, per year	0.48 (.25–.92)[d]	. . .
Weekly alcohol use, vs less than weekly[e]	0.13 (.10–.96)[d]	. . .
Current use of medroxyprogesterone[f]	0.14 (.02–.86)[d]	. . .
Current use of combined hormonal contraception[f]	0.19 (.05–.72)[d]	. . .
Pregnancy[e]	. . .	16.98 (4.08–70.77)[d]

Empty cells reflect nonsignificant hazard ratios in the model adjusting for age, condom use, and prevalent infection.

Abbreviations: CI, confidence interval; HR, hazard ratio.

[a] Group 1 are women who had evidence of human papillomavirus type 16 (HPV16) DNA infection and cleared and had a second detection.

[b] Variables were adjusted for prevalent infection.

[c] Group 2 are women who had evidence of HPV 16 by serology and had reappearance of HPV16 by DNA detection referred to as second detection.

[d] $P < .05$.

[e] Reported since last visit: yes vs no.

[f] Versus no hormonal contraception.

and the other a competitive Luminex immunoassay. The authors conclude that the competitive Luminex immunoassay measures a subset of the overall polyclonal responses and is more type specific. The differences in serologic tests likely contribute to the conflicting data regarding antibodies and protection and rate of recurrence [4, 26, 27, 29, 30]. We also did not do DNA sequencing of HPV16. However, redetection of the same HPV16 variant does not preclude reinfection. Also, false-negative and -positive DNA test results may have influenced our estimates.

In conclusion, redetection of HPV16 in most healthy young women is likely due to new sexual exposures. To our knowledge, this is the first study to demonstrate the rapid clearance of these redetections and the absence of CIN 2/3 development, supporting the premise that most women who clear HPV infections develop memory immune responses. Interpretation is more complex in women with prevalent infections, as this group appears to include a subset of women with failed immune responses resulting in HPV persistence. The recurrent detection of HPV in these cases may reflect reactivation with subsequent risk for the development of CIN 2/3.

Supplementary Data

Supplementary materials are available at *The Journal of Infectious Diseases* online (http://jid.oxfordjournals.org/). Supplementary materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyedited. The contents of all supplementary data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

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