Prevalence of Genital Human Papilloma Virus Infection and Genotypes among Young Women in Sicily, South Italy

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Abstract

Infection with oncogenic human papilloma virus (HPV) types is a necessary cause of cervical cancer. This study assessed the prevalence of HPV infection and genotypes among 1,006 randomly selected women, ages 18 to 24 years, living in Sicily (south Italy). The overall HPV rate was 24.1% (95% confidence interval, 21.5-26.9). The most frequent types were HPV-16 (4.5%), HPV-53 (2.7%), and HPV-84 (2.6%). The prevalence of vaccine types HPV-6, HPV-11, and HPV-18 was 1.4%, 0.1%, and 1.3%, respectively. Cytologic abnormalities were uncommon (3.1%) and associated with HPV detection ($P < 0.0001$). The only risk factor for HPV infection was the number of sexual partners (women with 2-3 partners versus women with 1 partner: odds ratio, 3.86; 95% confidence interval, 2.45-6.09). Genital HPV infection is relatively high in young Italian women. The high prevalence of viral types other than vaccine types should be taken into account to ensure accurate postvaccine surveillance and early detection of a possible genotype replacement. (Cancer Epidemiol Biomarkers Prev 2008; 17(8):2002–6)

Introduction

Infection with human papilloma virus (HPV) is a necessary cause of cervical cancer (1). About 60 distinct HPV genotypes can infect the genital tract. On the basis of their association with cervical carcinogenesis, they are grouped as low–oncogenic risk HPV (e.g., HPV-6 and HPV-11) or high–oncogenic risk HPV (e.g., HPV-16 and HPV-18).

Among women in the general population, the prevalence of HPV infection ranges from 2% to 44%, with the highest prevalence (44.8%) in women ages 20 to 24 years (2, 3). Variation is also evident in the regional distribution of HPV types (1, 4). Within the framework of vaccination with type-specific bivalent (for HPV-16 and HPV-18) or quadrivalent (for HPV-6, HPV-11, HPV-16, and HPV-18) prophylactic vaccines, (5), population-based data for HPV-type distribution and determinants of infection are essential to planning adequate preventive measures.

In Italy, the prevalence of HPV infection has been determined in women ages 25 to 70 years (6), whereas data for women ages 18 to ≤25 years are still lacking. Younger women make up a different segment of population, and they are considered to be more susceptible to HPV infection (2). Generalizing about the findings obtained from older women, therefore, may be questionable (7).

The primary objective of this study was to assess the prevaccine prevalence of cervical HPV infection and type distribution among the female population ages 18 to 24 years living in Sicily (south Italy). A secondary objective was to examine HPV infection in light of the presence of cytologically abnormal cervical findings and some selected sociobehavioral characteristics.

Materials and Methods

Study Population and Sample Collection. The study was conducted by the Department of Hygiene and Microbiology and the Mother and Child Institute of the University of Palermo, and the Department for Regional Health Inspection; it was approved by the Ethics Committee of the University of Palermo. The sample size calculation was based on data from another Italian study (6), which showed a 13% prevalence of HPV infection in women ages 25 to 29 y. Assuming a slightly higher prevalence in women ages 18 to 25 y (3, 7), it was anticipated that examining 1,225 women would allow observation to a point estimate of 15% HPV rate [95% confidence interval (95% CI), 12.8-17.2 ± 0.02]. Assuming a participation rate of about 25%, between June 2006 and February 2007, letters of invitation were mailed to a population-based, age-stratified sample of 4,600 women, who were identified from randomized lists produced from population registers.

A second letter was sent to women who failed to contact their screening center 15 d after the first letter had been posted. The women called to replace those not accepting the invitation were from the same age groups.

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Table 1. HPV prevalence by sociobehavioral characteristics: univariate analysis

Characteristics	No.	HPV positive n (%)	OR (95% CI)	*P*
Age (y)				0.29
18	75	14 (18.6)	1	
19	98	16 (16.3)	0.85 (0.39-1.87)	
20	110	27 (24.5)	1.42 (0.69-2.93)	
21	167	48 (28.7)	1.76 (0.90-3.44)	
22	159	42 (26.4)	1.56 (0.79-3.09)	
23	148	33 (22.3)	1.25 (0.62-2.51)	
24	249	63 (25.3)	1.48 (0.77-2.82)	
Age at first sexual intercourse (y)				0.01
≤17	400	103 (25.8)	1	
18-19	220	46 (20.9)	0.76 (0.51-1.13)	
≥20	117	14 (12.0)	0.39 (0.21-0.72)	
Time since first sexual intercourse (y)				0.00
≤1	91	13 (14.3)	1	
2-3	182	28 (15.4)	1.09 (0.54-2.22)	
≥4	464	122 (26.3)	2.14 (1.15-3.99)	
No. of lifetime partners				0.00
1	479	57 (11.9)	1	
2-3	334	115 (34.4)	3.89 (2.72-5.56)	
≥4	97	45 (46.4)	6.41 (3.94-10.41)	
Pregnancies				0.40
0	697	163 (23.4)	1	
1-2	109	24 (22.0)	0.93 (0.57-1.50)	
≥3	13	1 (7.7)	0.27 (0.04-2.12)	
Education				0.46
Primary school*	7	1 (14.3)	1	
Middle school	257	49 (19.0)	1.41 (0.16-12.01)	
High school	589	134 (22.8)	1.77 (0.21-14.81)	
University	39	11 (28.2)	2.36 (0.25-21.90)	
Occupation				0.36
Housewife*	139	27 (19.4)	1	
Unemployed	152	28 (18.4)	1.34 (0.83-2.17)	
Working	218	52 (23.8)	0.93 (0.52-1.68)	
Student	385	94 (24.4)	1.29 (0.77-2.19)	
Oral contraceptives				0.03
No*	637	133 (20.9)	1	
Yes	171	49 (28.6)	1.52 (1.03-2.23)	
Smoking				0.00
No*	520	102 (19.6)	1	
Yes	356	98 (27.5)	1.55 (1.13-2.14)	

*Reference level.

The analyses were restricted to those women who reported being sexually active. The women were invited to sign an informed-consent form and answer a questionnaire that included information about age, education, occupation, number of pregnancies, age at first sexual activity, lifetime number of partners, smoking habits, and current use of oral contraceptives. The participants received a standardized pelvic examination by a gynecologist and cervical sampling for HPV typing and cervical cytology. Neither the vulva nor the vagina was examined in this study, nor were vulvovaginal samples collected. All the women received a written response of their cytology and HPV result.

HPV Testing and Cytology. The DNA was extracted with the use of the QIAamp Mini Kit (Qiagen). HPV detection was done with the Linear Array HPV Genotyping Test (Roche Diagnostics), which allows the identification of 16 types considered low–oncogenic risk HPV (HPV-6, HPV-11, HPV-40, HPV-42, HPV-54, HPV-55, HPV-61, HPV-62, HPV-64, HPV-70, HPV-71, HPV-72, HPV-81, HPV-83, HPV-84, and HPV-89) and 21 types (HPV-16, HPV-18, HPV-26, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-53, HPV-56, HPV-58, HPV-59, HPV-66, HPV-67, HPV-68, HPV-69, HPV-73, HPV-82, and HPV-IS39) considered definitive or probable high–oncogenic risk HPV types (1, 8). All the women were screened for the presence of abnormal cervical cytology with the use of thin-layer Pap smears. The cytologic test results were classified as atypical squamous cell of undetermined significance, low-grade squamous intraepithelial lesion, and high-grade lesions, according to the Bethesda classification (9). Any abnormalities detected were dealt with in compliance with the guidelines updated by the Italian Association of Colposcopy and Cervical/Vaginal Pathology (10).

Statistical Analysis. The association between HPV infection and behavioral/clinical variables was assessed through the χ^2 test or Fisher's exact test ($P \leq 0.05$ significant) and crude odds ratio (OR). Multivariate logistic regression was applied to obtain the adjusted OR. To deal with missing data, a multiple imputation model with fully conditional specification (11) was used.

Results

A total of 1,013 (22.5%) of the women agreed to participate. Of them, 7 (0.7%) were excluded due to samples inadequate for HPV testing, resulting in a final sample of 1,006 women.

A total of 243 (24.1%; 95% CI, 21.5-26.9) women were HPV positive. On the whole, 18 high–oncogenic risk and 14 low–oncogenic risk HPV types were identified (Fig. 1). The high–oncogenic risk HPV types were present in 175 women (17.4% of all women and 72.0% of those HPV positive); multiple infections were evident in 104 women (10.3% of all women and 42.8% of those HPV positive). The most frequent type was HPV-16 (45 infections; 4.5% of all women), followed by HPV-53 (27 infections; 2.7%), HPV-84 (2.6%), HPV-42 (2.5%), HPV-62 (2.4%), HPV-66 (2.2%), and HPV-89 (2.2%). The prevalence of vaccine types HPV-6, HPV-11, and HPV-18 was 1.4%, 0.1% and 1.3%, respectively. The cumulative rate of HPV-16 and HPV-18 infections was 5.8%; that of HPV-6, HPV-11, HPV-16, and HPV-18 infections was 7.3%. Two (0.5%) women had concomitant HPV-16 and HPV-18 infection; no concomitant HPV-6, HPV-11, HPV-16, and HPV-18 infections were observed.

An adequate cytologic diagnosis was obtained for 923 (91.7%) women. Abnormalities were detected in 29 (3.1%) women (22 atypical squamous cell of undetermined significance, 6 low-grade squamous intraepithelial lesion, and 1 high-grade lesions). Of the women with abnormal cytology, 24 (82.7%) were HPV positive; of those with normal cytology, 200 of 894 (22.4%) were HPV positive ($P < 0.0001$). However, among the 224 HPV-infected women with adequate cytology, only 24 (10.7%) had an abnormal finding.

The characteristics of the study women in relation to HPV infection are reported in Table 1. The mean age was 21.6 ± 1.9 y, the mean age of first sexual intercourse was 17.4 ± 2.0 y (range, 13-24 y), the mean time since the first sexual intercourse was 4.33 ± 2.3 y (range, 0-11 y), the mean number of lifetime partners was 2.22 ± 1.66 (range, 1-10), and the mean number of pregnancies was 0.21 ± 0.61 (range, 0-5). At different age strata, HPV prevalence was 18.6% (95% CI, 10.2-29.4) for 18 y, 16.3% (95% CI, 9.8-25.6) for 19 y, 24.5% (95% CI, 16.3-33.4) for 20 y, 28.7% (95% CI, 22.6-36.4) for 21 y, 26.4% (95% CI, 19.2-33.6) for 22 y, 22.3% (95% CI, 15.4-29.1) for 23 y, and 25.3% (95% CI, 20.6-31.3) for 24 y. In multivariate analysis (Table 2), only the number of lifetime partners was a significant risk factor for HPV infection. Additionally, the number of sexual partners was found to be a risk factor for infection with high–oncogenic risk HPV types (OR of women with 2-3 sexual partners, 4.0; 95% CI, 2.58-6.28; OR of women with ≥4 sexual partners, 7.3; 95% CI, 4.49-13.92) and infection with multiple HPV types (OR of women with 2-3 sexual partners, 4.0; 95% CI, 2.32-7.18; OR of women with ≥4 sexual partners, 4.63; 95% CI, 2.18-9.65) (data not shown).

Table 2. HPV infection by sociobehavioral characteristics: multivariate analysis

Characteristics	OR (95%CI)	*P*
Age at first intercourse (y)		
18-19	1.00 (0.62-1.62)	0.99
≥20	0.79 (0.40-1.56)	0.50
Time since first sexual intercourse (y)		
2-3	0.69 (0.21-2.26)	0.55
≥4	0.84 (0.37-1.88)	0.66
No. of lifetime partners		
2-3	3.86 (2.45-6.09)	<0.0001
≥4	5.67 (3.21-10.03)	<0.0001
Oral contraceptives		
Yes	1.53 (0.82-2.87)	0.19
Smoking		
Yes	1.21 (0.70-2.07)	0.50

NOTE: Only variables significant in the univariate analysis are included.

Discussion

The present study showed a 24.1% prevalence of HPV infection among young, sexually active women <25 years of age living in Italy. This rate was in the range of the 19.7% to 39.0% reported worldwide for women of this age group (7, 12), and it was higher than that (8.8%) found by a north Italy study (6) in women ages 25 to 70 years. This latter finding confirms that the prevalence of HPV infection is highest among young women and drops with increasing age (2, 13).

Infection with oncogenic HPV types was predominant (72.0% of HPV-infected women), and this was consistent with what has been reported in sexually active young adults (13, 14). Similar to other international (2, 4, 15) and Italian (6, 16) findings, HPV-16 was the most prevalent type (4.5%). After HPV-16, the most common types were HPV-53 (2.7%), HPV-84 (2.6%), HPV-42 (2.5%), HPV-62 (2.4%), HPV-66 (2.2%), and HPV-89 (2.2%). Of these results, HPV-66 and HPV-42 were also the two most common types (after HPV-16) in north Italy (6). However, the following most frequent types in that study were HPV-45 and HPV-31, which, in Sicily (south Italy), were more rarely detected (0.6% and 1.5%, respectively). On the other hand, HPV-53, which was the second most frequent type in Sicily, was only rarely (0.2%) found in north Italy. These differences may be attributable to the type of HPV test used or the age of the study population; alternatively, they could indicate a different pattern of the distribution of HPV types that might reflect the geographic origins of samples (4, 17).

Vaccine types other than HPV-16 (i.e., HPV-6, HPV-11, and HPV-18) were detected in 1.4%, 0.1%, and 1.3% of participants, respectively. This low prevalence of each type, together with the low rate (0.5%) of concomitant HPV-16 and HPV-18 infections and the absence of concomitant HPV-6, HPV-11, HPV-16, and HPV-18 infections detected, would suggest that the young population being studied is an appropriate target for the prophylactic vaccination.

Cytologic abnormalities were uncommon (3.1%) in this population, and nearly all were low-grade changes. The low prevalence of cervical squamous intraepithelial lesions corroborated previous findings that young women with a high prevalence of HPV infection were detected with lesions in only 1.1% to 7.0% of cases (18, 19). A possible explanation for such discordance may be that certain biological risks/conditions among young women are relevant to the acquisition of HPV infection but not to the development of squamous intraepithelial lesions (18, 19).

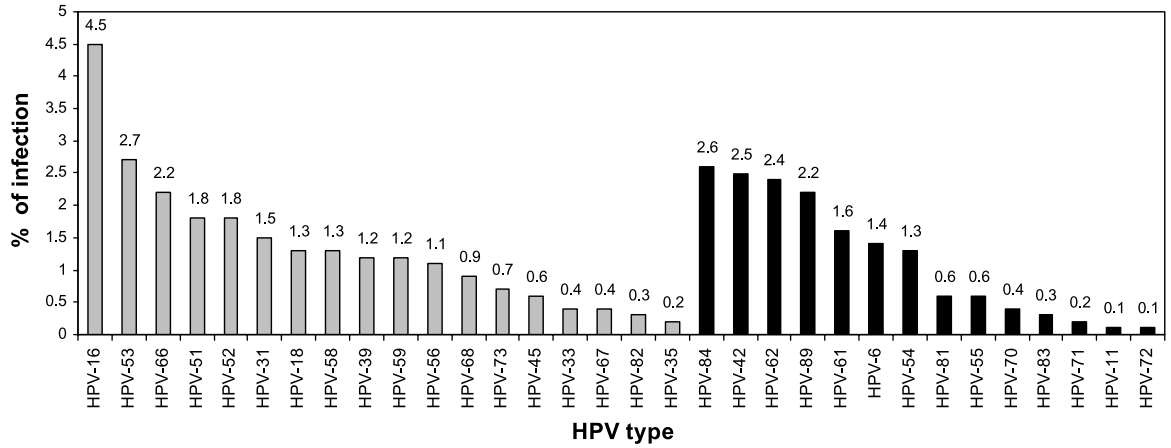

Figure 1. Type-specific distribution of high-risk (*light gray*) and low-risk (*dark gray*) HPV among 1,006 young women ages 18 to 24 y in Sicily, Italy, 2006-2007.

No association between HPV infection and increasing age was found, probably due to the limited age range examined, and this was also observed in other studies (7, 15). The only important determinant of HPV detection in multivariate analysis was the reporting of multiple lifetime sex partners. This finding is congruent with that of previous studies (13, 20-22), in which the prevalence of HPV among women who had had two or more sexual partners was double that in women who had had only one partner. Moreover, and consistent with other reports (13, 23), the high number of sexual partners was also found to confer an increased risk of infection with high–oncogenic risk HPV types and multiple HPV types. The age at first sexual intercourse, time since first sexual intercourse, use of oral contraceptives, and a smoking habit revealed an association with HPV infection in a crude analysis but not in a multivariate analysis. Across various studies, the results of the association between these variables and HPV infection have been inconsistent (15, 21, 23, 24). A possible explanation may be that they could be a marker for other risky sexual behavior (e.g., greater numbers of partners; ref. 24).

A major strength of this study is the fact that it provides the first estimates of the prevalence of HPV among young women from the general population in Italy. However, these data cannot be considered as nationally representative because the study analyzed women from a population living in Sicily, south Italy, who may differ from other Italian populations (17). Another potential limitation is the extent of missing data (ranging from 9% to 26%) that required an imputation scheme; however, the results of the analysis with imputed missing data were found to be consistent with those obtained by analyzing complete records only (data not shown).

In conclusion, this study showed a high prevalence of genital HPV infection in young Italian women. The high prevalence of viral types other than vaccine types should be taken into account to ensure accurate postvaccine surveillance and early detection of a possible genotype replacement. This information will contribute to elucidating the epidemiology of HPV infection across subpopulations, and it will also be helpful in the implementation of future prevention strategies.

Disclosure of Potential Conflicts of Interest

Prof. Antonio Perino has research contracts with Sanofi-Pasteur MSD, and GlaxoSmithKline. The other authors do not have any conflict of interest.

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References

1. Munoz N, Castellsague X, de Gonzalez AB, Gissmann L. Chapter 1: HPV in the etiology of human cancer. Vaccine 2006;24S3:S1–10.
2. Trottier H, Franco EL. The epidemiology of genital human papillomavirus infection. Vaccine 2006;24 Suppl 1:S1–15.
3. Dunne EF, Unger ER, Sternberg M, et al. Prevalence of HPV infection among females in the United States. JAMA 2007;297:813–9.
4. Clifford GM, Gallus S, Herrero R, et al. Worldwide distribution of human papillomavirus types in cytologically normal women in the International Agency for Research on Cancer HPV prevalence surveys: a pooled analysis. Lancet 2005;366:991–8.
5. Ames A, Gravitt P. Human papillomavirus vaccine update. Curr Infect Dis Rep 2007;9:151–8.
6. Ronco G, Ghisetti V, Segnan N, et al. Prevalence of human papillomavirus infection in women in Turin, Italy. Eur J Cancer 2005;41:297–305.
7. Manhart LE, Holmes KK, Koutsky LA, et al. Human papillomavirus infection among sexually active young women in the United States: implications for developing a vaccination strategy. Sex Transm Dis 2006;33:502–8.
8. de Villiers EM, Fauquet C, Broker TR, Bernard HU, zur Hausen H. Classification of papillomaviruses. Virology 2004;324:17–27.

9. Solomon D, Davey D, Kurman R, et al. The 2001 Bethesda System: terminology for reporting results of cervical cytology. JAMA 2002; 287:2114–9.
10. Gestione della paziente con il Pap-test anormale. Linee Guida Edizione 2006. La Colposcopia in Italia, Società Italiana di Colposcopia e Patologia Cervico Vaginale 2006;Anno XXI.
11. van Buuren S, Brand JPL, Groothuis-Oudshoorn K, Rubin DB. Fully conditional specification in multivariate imputation. J Stat Comput Simul 2006;76:1049–64.
12. Winer RL, Lee SK, Hughes JP, Adam DE, Kiviat NB, Koutsky LA. Genital human papillomavirus infection: incidence and risk factors in a cohort of female university students. Am J Epidemiol 2003;157: 218–26.
13. Ho GY, Bierman R, Beardsley L, Chang CJ, Burk RD. Natural history of cervicovaginal papillomavirus infection in young women. N Engl J Med 1998;338:423–8.
14. Giuliano AR, Harris R, Sedjo RL, et al. Incidence, prevalence, and clearance of type-specific human papillomavirus infections: The Young Women's Health Study. J Infect Dis 2002;186:462–9.
15. Shin HR, Franceschi S, Vaccarella S, et al. Prevalence and determinants of genital infection with papillomavirus, in female and male university students in Busan, South Korea. J Infect Dis 2004;190:468–76.
16. De Francesco MA, Gargiulo F, Schreiber C, Ciravolo G, Salinaro F, Manca N. Detection and genotyping of human papillomavirus in cervical samples from Italian patients. J Med Virol 2005;75:588–92.
17. Walter H, Matsumoto H, Danker-Hopfe H, De Stefano GF, Rickards O. GM and KM allotypes in nine population samples of Sicily. Ann Hum Biol 1997;24:419–26.
18. Moscicki AB, Hills N, Shiboski S, et al. Risks for incident human papillomavirus infection and low-grade squamous intraepithelial lesion development in young females. JAMA 2001;285:2995–3002.
19. Moscicki AB, Palefsky J, Gonzales J, Schoolnik GK. Human papillomavirus infection in sexually active adolescent females: prevalence and risk factors. Pediatr Res 1990;28:507–13.
20. Moscicki AB, Schiffman M, Kjaer S, Villa LL. Chapter 5: Updating the natural history of HPV and anogenital cancer. Vaccine 2006;24 Suppl 3:S42–51.
21. Karlsson R, Jonsson M, Edlund K, et al. Lifetime number of partners as the only independent risk factor for human papillomavirus infection: a population-based study. Sex Transm Dis 1995;22:119–27.
22. Vaccarella S, Herrero R, Dai M, et al. Reproductive factors, oral contraceptive use, and human papillomavirus infection: pooled analysis of the IARC HPV prevalence surveys. Cancer Epidemiol Biomarkers Prev 2006;15:2148–53.
23. Silins I, Kallings I, Dillner J. Correlates of the spread of human papillomavirus infection. Cancer Epidemiol Biomarkers Prev 2000;9: 953–9.
24. Kahn JA, Rosenthal SL, Succop PA, Ho GY, Burk RD. Mediators of the association between age of first sexual intercourse and subsequent human papillomavirus infection. Pediatrics 2002;109:E5.