

Sexual Behavior and Evidence for an Infectious Cause of Prostate Cancer

Howard D. Strickler¹ and James J. Goedert²

BACKGROUND: ENVIRONMENTAL FACTORS

Prostate cancer incidence and mortality rates vary widely according to geography and race with more than 90-fold differences in incidence between the highest and lowest (1, 2). The world's highest prostate cancer incidence is among African-Americans who have annual age-standardized rates of 185 per 100,000 persons (world standard population) (3). The lowest prostate cancer incidence rates are reported in Asian countries such as Japan (9 per 100,000 population) and China (2 per 100,000 population) (4). Genetic factors could explain some of these differences. However, studies of migrants have consistently shown that risk increased following immigration from a low-incidence to a high-incidence country. For example, Japanese immigrants to America experienced a greater than fourfold increase in prostate cancer rates following immigration (5), and cross-sectionally, prostate cancer rates among Asians in America are much higher than in their native countries (6). Similar patterns have been observed for Asian immigrants to Australia (7). Overall, the geographic/racial heterogeneity of prostate cancer rates and the effects of migration have been interpreted as evidence that environmental factors may have important effects on the risk of prostate tumorigenesis (8, 9).

PROSTATE CANCER AND SEXUAL BEHAVIOR

Quisenberry (10) in 1960, speculated that some of the ethnic differences observed in prostate cancer rates might be due to cultural variations in male sexual behavior. Subsequent evidence supporting this hypothesis was reported by epidemiologic studies conducted during the early 1970s. In a case-control study involving just 39 "recent" prostate cancer patients, Steele et al. (11) in 1971, found self-reported history of extramarital sexual intercourse was significantly associated with prostate cancer. Cancer cases were also substantially (albeit, not significantly) more likely to report having had a sexually transmit-

ted disease, premarital sex, more than six sexual partners, and use of condoms. The investigators further observed that more prostate cancer cases than controls wished they had experienced sexual intercourse with greater frequency. The data, they concluded, were consistent with an infectious, sexually transmitted etiologic agent or an association of prostate cancer with "sexual drive", possibly due to a mutual association with androgenic hormone activity. Soon thereafter, in 1974, a larger and more formal case-control study by Krain (12) found that sequential prostate cancer patients ($n = 221$) had significantly more sexually transmitted diseases, sexual partners, condom use, and frequency of sexual intercourse than did age- and race-matched patients with non-genitourinary, non-cancerous conditions from the same hospitals.

A number of subsequent epidemiologic case-control investigations have studied the association of prostate cancer with sexual behavior. Most studies through the 1990s found an association with self-reported history of having had a sexually transmitted disease (13-17). This effect, though, was seldom statistically significant (15), and findings regarding individual types of sexually transmitted diseases, mainly gonorrhea and syphilis, were varied (15, 16, 18-26). Age at first intercourse was associated with prostate cancer in several (16, 20, 27, 28) but not all (24, 25, 29) studies, whereas lifetime number of sexual partners was not found to be associated with prostate cancer (14-16, 20, 25, 27, 30) except in a small number of studies (24, 28). Few investigations involved serologic assays for sexually transmitted diseases (13, 14, 31, 32) and these gave conflicting results. Additional factors, such as circumcision (14, 15, 17), which might protect against development of sexually transmitted diseases (33), a history of homosexuality (14), and a history of sexually transmitted disease in sexual partners (20, 21) were occasionally found to be associated with prostate cancer.

Among these studies, few involved more than 150 cases. Hsieh et al. (26) was among the largest, with 320 hospitalized cases and 246 individually matched hospital controls (26). Self-reported history of sexually transmitted diseases was more common in that study population than in most other investigations, 14 percent overall. Nonetheless, no association was found between history of a sexually transmitted disease and prostate cancer. Statistical power somewhat limited the interpretation of these results, however. The sample size was reportedly adequate for detecting an odds ratio ≥ 2.0 , assuming 15 percent prevalence, just slightly higher than the overall prevalence of sexually transmitted diseases. Thus, even in that large study with a high

Received for publication September 29, 2000, and accepted for publication February 15, 2001.

Abbreviations: CI, confidence interval; OR, odds ratio.

¹Department of Epidemiology and Social Medicine, Albert Einstein College of Medicine, Bronx, NY.

²Viral Epidemiology Branch, National Cancer Institute, Bethesda, MD.

Reprint requests to Dr. Howard Strickler, Department of Epidemiology and Social Medicine, Albert Einstein College of Medicine, 1300 Morris Park Avenue, Belfer Bldg., Room 1308-B, Bronx, NY 10461 (e-mail: strickle@aecom.yu.edu).

prevalence of exposure, statistical power was moderate. In addition, it is unclear from their report whether Hsieh et al. accounted for their individually matched design in their analyses. Assuming they did not, this would have biased the results toward the null (34).

A study involving 250 individually matched hospitalized cases, as well as 238 hospitalized and 240 neighborhood controls, in contrast, found a significant association (odds ratio (OR) = 1.9; 95 percent confidence interval (CI): 1.3, 2.5) of prostate cancer with history of sexually transmitted disease, after accounting for individual matching and other variables (14). The number of homosexual, but not heterosexual, partners was also significantly related to prostate cancer. However, an important concern based on the report is whether or not this study was entirely new or involved data from subjects reported in an earlier positive study (13). Interestingly, the few additional studies of moderate size gave mixed results, with some tendency for positive studies to be those with higher sexually transmitted disease prevalence in the study population (15, 16) than negative studies (23, 25).

Thus, the findings until recently were suggestive but far from conclusive, with the most consistent limitations being small sample size and frequent dependence on self-reported sexual history. Moreover, the paucity of prospective studies (e.g., nested case-control investigations) is striking. An observational cohort study of approximately 10,000 male syphilis patients, with diagnosis and follow-up between 1972 and 1987, found no association with prostate cancer (22). However, the population was probably too young and the period of observation too short to have had reasonable expectation of detecting such a relation. In a paper by Key (35), summary odds ratios across published studies were calculated, using the variance of each result to determine a weighted average. The summary results for age at intercourse, number of sexual partners, and history of sexually transmitted disease were each significant. The effect of number of sexual partners was mostly accounted for by the early studies of Steele et al. (11) and Krain (12), though, and the summary statistics by Key (35) do not represent a formal meta-analysis.

Recently, a large population-based case-control investigation was reported by Hayes et al. (36). The study involved 981 cases (479 African-Americans and 502 Caucasian-Americans) and 1,315 controls. The lifetime number of sexual partners was not associated with case-control status. However, cases were significantly more likely to report a history of syphilis (OR = 2.6; 95 percent CI: 1.3, 5.1), to have *Treponema pallidum* serum antibodies (MHA-TP) (OR = 1.8; 95 percent CI: 1.0, 3.5), and a higher number of episodes of gonorrhea ($p_{\text{trend}} = 0.0005$) after adjusting for age and race. Cases were also more likely to report sex with prostitutes (OR = 2.3; 95 percent CI: 1.3, 4.2) and less frequent use of condoms ($p_{\text{trend}} = 0.009$), opposite the findings for contraceptive use in studies by Steele et al. (11) and Krain (12) but consistent with the existence of a sexually transmitted prostate cancer agent. Interestingly, the prevalence of these risk factors was considerably greater among African-Americans.

Thus, the largest case-control study to date suggests that prostate cancer is associated with sexual history and, in particular, history of sexually transmitted disease. The study was strengthened by the inclusion of serologic data, which are invulnerable to recall or response biases. In addition, the high rate of sexually transmitted diseases among African-American males observed by the researchers is consistent with US national data (37, 38), suggesting that the study population was reasonably representative. The high rate of sexually transmitted diseases among African-Americans is, as well, ecologic data consistent with the possibility that a sexually transmitted infectious etiologic agent could help explain high rates of prostate cancer among African-Americans. It may, furthermore, help explain some of the disparities in earlier case-control studies, many of which studied ethnic groups that have lower rates of sexually transmitted diseases, making their estimation of relative risk imprecise, especially given their limited sample sizes. Because of the modest effects (i.e., odds ratios) observed by Hayes et al. (36), issues of sample size and rates of exposure must be carefully considered in all future studies.

Another ecologic observation, increased rates of prostate cancer among ever married men, has also been used by some investigators to support the sexual association of prostate cancer. The controversial relation of prostate cancer with marriage has been confirmed by large registry-based studies in the United States and Europe (39, 40). One hypothesis is that marital status and sexual drive are related, that married men have more sexual intercourse, and that both may be indicators of higher androgenic activity. Against this theory, testosterone levels in the physiologic range have not been clearly associated with sex drive or marital status (41). No consistent association between prostate cancer and the frequency of intercourse or ejaculations has been found (11, 12, 14–16, 25, 27, 30), and no such association was observed in the large case-control study by Hayes et al. (36). Additionally, because sexually transmitted diseases currently are more common in unmarried men, the greater frequency of prostate cancer among married men at first glance appears to be inconsistent with an infectious, sexually transmitted etiologic agent (38, 42). It is unclear, however, how sexually transmitted disease history differs by marital status in the elderly birth cohorts from which most prostate cancer cases arise.

VIROLOGIC STUDIES OF PROSTATE CANCER

Direct evidence of an infectious agent involved in prostate tumorigenesis has been sought for decades. Initial studies primarily focused on herpesviruses, since at the time it was thought that these agents, mainly herpes simplex virus type 2, might play a major role in cervical cancer. Herpesviruses, including herpes simplex virus type 1, herpes simplex virus type 2, and cytomegalovirus can infect anogenital tissues, and in vitro can immortalize human cells (43). Moreover, Kaposi's sarcoma herpes virus appears to be the cause of Kaposi's sarcoma (44), and Epstein-Barr virus may play a role in lymphomas, as well as in nasopharyngeal carcinoma (45). Centifanto and colleagues (46–49) and oth-

TABLE 1. Summary of studies investigating human papillomavirus (HPV) in the prostate

Study (reference no.) and date	HPV detection	Collection (storage)	No.	Subjects Type	HPV (%)	HPV types*			
						6	16	18	X
McNicol and Dodd (61), 1990	Southern blot for HPV 16, 18	Mostly TURP†, also SPT† (frozen)	4	Prostate cancer cases	75				
McNicol and Dodd (63), 1990	E6 polymerase chain reaction for HPV 16, 18	Mostly TURP, also SPP and autopsy (frozen)	12	Benign prostatic hypertrophy controls	33				
Masood et al. (68), 1991	In situ hybridization for HPV 6, 11, 16, 18, 31, 33, 35	Biopsies/TURP (paraffin)	4	Prostatic cancer cases	100	4			
McNicol and Dodd (64), 1990	E6 polymerase chain reaction for HPV 16, 18	TURP/SPP (frozen)	15	Benign prostatic hypertrophy controls	93	14	3		
Anwar et al. (62), 1992	E6 polymerase chain reaction for HPV 16, 18, 33	TURP/SPP and autopsy (paraffin)	5	Normal autopsies	20	1			
Rotola et al. (63), 1992	E6 polymerase chain reaction for HPV 6/11, 16	Collection not specified (frozen)	20	Prostate cancer cases	None				
Effert et al. (69), 1992	"Differential" polymerase chain reaction for HPV 16, 18	Collection not specified (frozen)	20	Benign prostatic hypertrophy controls	None				
Serfling et al. (53), 1992	L1 consensus primer polymerase chain reaction	Collection not specified (frozen)	27	Prostate cancer cases	52	14	1		
Ibrahim et al. (65), 1992	L1 polymerase chain reaction and in situ hybridization	Biopsies/TURP/SPP (paraffin and frozen)	56	Benign prostatic hypertrophy controls	63	34	3		
Sarkar et al. (84), 1993	E6/E7 polymerase chain reaction for 6/11, 16, 18, also Southern blot	Surgical not TURP (paraffin-microdissection)	68	Prostate cancer cases	41	11	17	5	
Dodd et al. (66), 1993	Reverse transcription polymerase chain reaction for E6/E7 mRNA of HPV 16	Collection not specified (frozen)	10	Benign prostatic hypertrophy controls	None				
Tu et al. (85), 1994	L1 consensus primer polymerase chain reaction	Surgical not TURP (tumors-paraffin; metastasis-frozen)	10	Prostate cancer and prostatic intraepithelial neoplasia cases	13	3			
Moyret-Lalle et al. (86), 1995	E6 polymerase chain reaction for HPV 16, 18	Collection not specified (frozen)	7	Prostate cancer cases	43	3			
Wideroff et al. (67), 1966	L1 consensus primer polymerase chain reaction, and E6 polymerase chain reaction for HPV 6, 11, 16, 18, 31, 33, 45	TURP and SPP (paraffin)	10	Benign prostatic hypertrophy controls	50	5			
			43	Prostate cancer cases	2	1			
			17	Metastases	6				
			1	Normal	None				1
			17	Prostate cancer cases	53	9			
			22	Benign prostatic hypertrophy controls	32	7			
			56	Prostate cancer cases	L1, 13				
			42	Benign prostatic hypertrophy controls	E6, 0				
					L1, 10				
					E6, 0				

Author(s)	Methodology	Prostate cancer cases	Prostate cancer cases	HPV types
Suzuki et al. (87), 1996	L1 consensus primer polymerase chain reaction	51	16	None
Anderson et al. (88), 1997	E2, E6, and E1 consensus primer polymerase chain reaction	14 10	None None	None
Terris and Peehl (71), 1997	E6 (two separate but overlapping regions) and L1 consensus primer polymerase chain reaction	53 21	Prostate cancer cases Benign prostatic hypertrophy controls	E6 ^a , 4 E6 ^b , 19 L1, 0 E6 ^a , 10 E6 ^b , 33 L1, 0 E6 ^a , 3 E6 ^b , 14 L1, 0
Gherdovich et al. (89), 1997	L1 consensus primer polymerase chain reaction	37	Normal	None
Strickler et al. (70), 1998	E6 and L1 (MY09/11 and GP5+/6+) consensus primer polymerase chain reaction	5 60 63 61	Prostate cancer cases Benign prostatic hypertrophy controls Prostate cancer cases Benign prostatic hypertrophy controls	None None None None
Noda et al. (72), 1998	Nested consensus primer polymerase chain reaction	38 71	Prostate cancer cases Benign prostatic hypertrophy controls	None 4
Serth et al. (73), 1999	Quantitative HPV 16 E6 polymerase chain reaction	47 37	Prostate cancer cases Benign prostatic hypertrophy controls	Most Most

* The HPV types were reported in the table if specified by the investigators.

† Abbreviations: TURP, transurethral resection of the prostate; SPP, suprapubic resection of the prostate; NS, not specified.

ers (32, 50–52) during the 1970s and early 1980s found evidence of herpes simplex virus and cytomegalovirus in prostate tissue specimens. Increased herpes simplex virus and cytomegalovirus seroprevalence among prostate cancer cases also was reported (13, 31, 32), but not by all studies (14), and subsequent virologic studies generally failed to provide support (32). In particular, a small study utilizing modern virologic methods (i.e., polymerase chain reaction) failed to detect herpes simplex virus in benign prostatic hypertrophy or prostate cancer (53).

More recently, Kaposi's sarcoma herpes virus was detected in five of eight benign prostatic hypertrophy and two of eight prostate cancer specimens in Italy where this virus is endemic (54). As reviewed by Blackburn and Levy (55), however, infected infiltrating lymphocytes or laboratory artifacts could explain these findings. Expression of Kaposi's sarcoma herpes virus-related RNA was reported in 12 of 16 prostate tissues, including four with prostate cancer (56), but this observation has yet to be corroborated. In a large serosurvey of cancer patients in South Africa, Kaposi's sarcoma herpes virus seroprevalence among 202 prostate cancer patients was not elevated (57). In summary, although the herpesviruses, Epstein-Barr virus, and Kaposi's sarcoma herpes virus are human cancer viruses, and herpesviruses can infect anogenital tissues, there is little evidence that herpesviruses affect the risk of prostate cancer.

The possible etiologic role of human papillomavirus in prostate cancer is an active focus of research. Human papillomavirus appears to cause most cervical cancers, and the virus is commonly detected in cancers of the anus and penis as well as in a subset of vaginal and vulvar tumors. Human papillomavirus is sexually transmitted, and human papillomavirus E6 and E7 proteins can immortalize human prostate cells in vitro through their effects on the cellular tumor suppressor gene products p53 and Rb, respectively (58). In addition, men who develop anal cancer (a human papillomavirus-associated tumor) have an increased risk of prostate cancer (59), and the incidence of cervical and prostate cancer in African-Americans (both high) and Jewish-Americans (both low) are consistent with an analogous role of human papillomavirus in prostate cancer (3, 60). Thus, there are biologic and epidemiologic reasons to consider human papillomavirus as a possible cause of prostate cancer.

Studies of the detection of human papillomavirus in prostate tissues have given mixed results (table 1). Some found a clear cancer association with human papillomavirus (61, 62), but others reported that human papillomavirus was equally prevalent in benign prostatic hypertrophy and even in normal prostate tissue (63–67). Additional studies did not detect human papillomavirus in any prostate tissues (53, 68, 69). Complicating matters further, most of these investigations have been small, have used varied approaches, and there is no obvious pattern in the methods of specimen collection, preservation, DNA testing, or patient populations to explain the different rates of human papillomavirus detection.

In a recent investigation, we attempted to better understand this issue by addressing some of the concerns raised by earlier studies (70). To optimally preserve DNA, we froze prostate cancer and benign prostatic hypertrophy tis-

sues in vapor phase liquid nitrogen immediately after collection. To minimize concerns that loss of selected viral sequences during tumorigenesis could cause negative findings (71), we used two primer sets (MY09/MY11 and GP5+/GP6+) that amplify different regions of L1, and a third set (WD66,67,154/WD72,76) targeted to E6. To minimize the possibility that patient characteristics could affect the findings, cases were 49 African-Americans (men at high risk of prostate cancer) and 14 Italians (intermediate risk) with prostate cancer, and a similar number of benign prostatic hypertrophy controls from each country. The sensitivity of the two L1 polymerase chain reaction assays was shown to be one human papillomavirus DNA genome per 100 cells, and the adequacy of tissue extracts for polymerase chain reaction was demonstrated by amplification of human β -globin DNA in all specimens except three cancers. Despite this, no human papillomavirus DNA was detected in any case or control specimens by MY09/MY11 or E6 polymerase chain reaction. Microdissection of 27 cancer specimens was conducted to minimize non-tumor DNA, but results remained negative by MY09/MY11 and GP5+/GP6+ polymerase chain reaction. Thus, our findings suggested that human papillomavirus DNA is uncommon in the prostates of older men and is not associated with prostate cancer.

Two additional polymerase chain reaction studies of human papillomavirus and prostate cancer have been reported since our investigation. Noda et al. (72) used a nested polymerase chain reaction assay designed to maximize test sensitivity for a wide range of human papillomavirus types. The results were positive in only three of 71 benign prostatic hypertrophy and zero of 38 prostate cancer specimens, generally consistent with our negative findings. In strong contrast, Serth et al. (73), using a novel quantitative human papillomavirus type 16 E6 polymerase chain reaction assay, found human papillomavirus type 16 DNA in most benign prostatic hypertrophy and prostate cancer tissues tested. Prostate cancer specimens were more likely to contain a high viral copy number. Interpretation of this study is limited by several factors. The sensitivity and specificity of the assay used by Serth et al. has never been demonstrated in human tissue specimens (e.g., cervix or other accepted reservoirs of human papillomavirus). Additionally, it seems improbable that a single human papillomavirus type, even an important type such as human papillomavirus type 16, could be present in essentially all male prostates, as only a subset of men have probably been exposed through an infected sexual partner. Human papillomavirus type 16 prevalence in the cervix is less than 5 percent among healthy women.

Seroepidemiologic human papillomavirus studies add an important perspective. Whereas several cross-sectional investigations of human papillomavirus antibodies failed to detect an association with prostate cancer (70, 74), two prospective studies found that human papillomavirus type 16 antibodies predicted development of prostate cancer years later (75, 76). This is similar to the situation for esophageal cancer in which prospective (77, 78) and cross-sectional investigations in the same laboratories have sometimes given conflicting results (74, 79). For esophageal cancer, some

investigators have proposed a "hit and run" model of viral tumorigenesis; that is, the virus contributes to the early phases of tumorigenesis but may later be lost. Such a mechanism may also fit the human papillomavirus/prostate cancer seroepidemiologic data. However, hit and run models are counter to the well-established mechanisms by which human papillomavirus is understood to cause cancer through the actions of E6 and E7. Instead, it is more likely that the prospective human papillomavirus serologic data may reflect the sexual association of prostate cancer, whereas this is not detected in cross-sectional human papillomavirus seroepidemiologic studies because by the time prostate cancer occurs, men have aged sufficiently that some have lost detectable levels of human papillomavirus antibodies. In this connection, human papillomavirus antibodies, when present, are often of low titer, and loss of antibody over time has been reported (80, 81).

CONCLUSIONS

Overall, the association of prostate cancer with sexual history and, particularly, sexually transmitted diseases has been suggested by case-control investigations, but these relations are not firmly established. Although it is not possible to entirely rule out a role for human papillomavirus or other known sexually transmitted infections in a subset of prostate cancers, the failure of studies using sensitive polymerase chain reaction assays to consistently detect viruses in prostate cancer cells, and the marginal strength of these associations when detected, are in marked contrast to the strong, consistent association of human papillomavirus with cervical cancer (ORs = 30–100 or more). Instead, the situation has similarities to that observed for cervical cancer before the association with human papillomavirus was understood: a moderate association was found with sexual behavior and occasional weak associations were found with herpes simplex virus or other sexually transmitted infections, each acting as surrogates for human papillomavirus. By analogy, the findings reviewed above could reflect a yet unrecognized sexually transmitted infection etiologically related to a subset of prostate cancer.

Such an interpretation is speculative. Given its importance, however, we argue that the suggestive findings to date are sufficient to warrant a formal, concerted effort to identify viral sequences in prostate cancer specimens—a search for an infectious cause of prostate cancer. Existing technologies allow testing for known and unknown infectious agents. Kaposi's sarcoma herpes virus, for example, was identified using representational difference analysis (82). The high costs and high risks of failure are obstacles to implementation, though, and the search for an infectious etiology of prostate cancer would require a special commitment by bench researchers.

Epidemiologists will have a similarly difficult role. First, prospective cohort studies will be necessary to provide additional evidence for an association between sexual behavior or sexually transmitted diseases and subsequent risk of prostate cancer. This will be difficult, since the effects being measured appear to be moderate and the prevalence of sexually trans-

mitted diseases will not be high in most populations likely to volunteer for long follow-up studies. It may require the pooled efforts of several research teams in possession of large serum and appropriate databases to conduct such an investigation. Once established, these multi-institutional efforts will, additionally, be the most likely mechanisms for epidemiologic assessment of the causal relation of any laboratory-defined prostate cancer-associated infectious agent(s) that is detected.

Second, epidemiologists and clinicians will need to collaborate with laboratory investigators in the collection of appropriate prostate cancer specimens for virologic testing. Cases with a history of multiple sexually transmitted diseases, selected from populations in which such an agent is most likely to be endemic (i.e., those having high rates of both sexually transmitted diseases and prostate cancer), such as African-Americans, might be the most likely to result in the identification of a novel, sexually transmitted, prostate cancer agent.

It is uncertain whether these several efforts will result in the discovery of an infectious cause of prostate cancer. Nonetheless, the initiatives outlined here are reasonable given the high morbidity and mortality associated with this disease. Identification of an infectious cause of prostate cancer would be a major advance, providing scientists with a new window into prostate tumorigenesis as well as an excellent target for efforts to prevent and treat prostate cancer.

REFERENCES

- Parkin DM, Pisani P, Ferlay J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J Cancer* 1999;80:827-41.
- Hsing AW, Tsao L, Devesa SS. International trends and patterns of prostate cancer incidence and mortality. *Int J Cancer* 2000;85:60-7.
- Ries LAG, Kosary CL, Hankey BF, et al. SEER cancer statistics review, 1973-1996. Bethesda, MD: US Department of Health and Human Services, National Institutes of Health, National Cancer Institute, 1999.
- Parkin DM, Muir CS, Whelan SL, et al. Cancer incidence in five continents. Lyon, France: International Agency for Research on Cancer, 1992.
- Haenszel W, Kurihara M. Studies of Japanese migrants. I. Mortality from cancer and other diseases among Japanese in the United States. *J Natl Cancer Inst* 1968;40:43-68.
- Stanford JL, Stephenson RA, Coyle LM, et al. Prostate cancer trends 1973-1995, SEER Program. Bethesda, MD: US Department of Health and Human Services, National Institutes of Health, National Cancer Institute, 1999. (NIH publication no. 99-4543).
- McCredie M, Williams S, Coates M. Cancer mortality in East and Southeast Asian migrants to New South Wales, Australia, 1975-1995. *Br J Cancer* 1999;79:1277-82.
- Pienta KJ, Esper P. Risk factors for prostate cancer. *Ann Intern Med* 1993;118:793-803.
- Nomura AMY, Kolonel LN. Prostate cancer: a current prospective. *Am J Epidemiol* 1991;133:200-27.
- Quisenberry W. Socio-cultural factors in cancer in Hawaii. *Ann N Y Acad Sci* 1960;84:795-806.
- Steele R, Lees REM, Kraus AS, et al. Sexual factors in the epidemiology of cancer of the prostate. *J Chronic Dis* 1971;24:29-37.
- Krain LS. Some epidemiologic variables in prostatic carcinoma in California. *Prev Med* 1974;3:154-9.
- Schuman LM, Mandel J, Blackard C, et al. Epidemiologic study of prostatic cancer: preliminary report. *Cancer Treat Rep* 1977;61:181-6.
- Mandel JS, Schuman LM. Sexual factors and prostatic cancer: results from a case-control study. *J Gerontol* 1987;42:259-64.
- Ross RK, Shimizu H, Paganini-Hill A, et al. Case-control studies of prostate cancer in blacks and whites in Southern California. *J Natl Cancer Inst* 1987;78:869-74.
- Honda GD, Bernstein L, Ross RK, et al. Vasectomy, cigarette smoking, and age at first sexual intercourse as risk factors for prostate cancer in middle-aged men. *Br J Cancer* 1988;57:326-31.
- Ewings P, Bowie C. A case-control study of cancer of the prostate in Somerset and east Devon. *Br J Cancer* 1996;74:661-6.
- Wynder EL, Mabuchi K, Whitmore WF Jr. Epidemiology of cancer of the prostate. *Cancer* 1971;28:344-60.
- Lees RE, Steele R, Wardle D. Arsenic, syphilis, and cancer of the prostate. *J Epidemiol Community Health* 1985;39:227-30.
- Mishina T, Watanabe H, Araki H, et al. Epidemiological study of prostatic cancer by matched-pair analysis. *Prostate* 1985;6:423-36.
- Oishi K, Okada K, Yoshida O, et al. A case-control study of prostatic cancer in Kyoto, Japan: sexual risk factors. *Prostate* 1990;17:269-79.
- Michalek AM, Mahoney MC, McLaughlin CC, et al. Historical and contemporary correlates of syphilis and cancer. *Int J Epidemiol* 1994;23:381-5.
- Hiatt RA, Armstrong MA, Klatsky AL, et al. Alcohol consumption, smoking, and other risk factors and prostate cancer in a large health plan cohort in California (United States). *Cancer Causes Control* 1994;5:66-72.
- Ilic M, Vlainjac H, Marinkovic J. Case-control study of risk factors for prostate cancer. *Br J Cancer* 1996;74:1682-6.
- La Vecchia C, Franceschi S, Talamini R, et al. Marital status, indicators of sexual activity and prostatic cancer. *J Epidemiol Community Health* 1993;47:450-3.
- Hsieh CC, Thanos A, Mitropoulos D, et al. Risk factors for prostate cancer: a case-control study in Greece. *Int J Cancer* 1999;80:699-703.
- Rotkin ID. Studies in the epidemiology of prostatic cancer: expanded sampling. *Cancer Treat Rep* 1977;61:173-80.
- Andersson SO, Baron J, Bergstrom R, et al. Lifestyle factors and prostate cancer risk: a case-control study in Sweden. *Cancer Epidemiol Biomarkers Prev* 1996;5:509-13.
- Fincham SM, Hill GB, Hanson J, et al. Epidemiology of prostatic cancer: a case-control study. *Prostate* 1990;17:189-206.
- Banerjee AK. Carcinoma of prostate and sexual activity. *Urology* 1986;28:159.
- Herbert JT, Birkhoff JD, Feorino PM, et al. Herpes simplex virus type 2 and cancer of the prostate. *J Urol* 1976;116:611-12.
- Baker LH, Mebust WK, Chin TD, et al. The relationship of herpesvirus to carcinoma of the prostate. *J Urol* 1981;125:370-4.
- Laumann EO, Masi CM, Zuckerman EW. Circumcision in the United States: prevalence, prophylactic effects, and sexual practice. *JAMA* 1997;277:1052-7.
- Schlesselman J. Case-control studies. New York, NY: Oxford University Press, 1982.
- Key T. Risk factors for prostate cancer. *Cancer Surv* 1995;23:63-77.
- Hayes RB, Pottner LM, Strickler H, et al. Sexual behaviour, STDs and risks for prostate cancer. *Br J Cancer* 2000;82:718-25.
- Fleming DT, McQuillan GM, Johnson RE, et al. Herpes simplex virus type 2 in the United States, 1976 to 1994. *N Engl J Med* 1997;337:1105-11.
- Rice RJ, Roberts PL, Handsfield HH, et al. Sociodemographic distribution of gonorrhea incidence: implications for prevention and behavioral research. *Am J Public Health*

- 1991;81:1252-8.
39. Newell GR, Pollack ES, Spitz MR, et al. Incidence of prostate cancer and marital status. *J Natl Cancer Inst* 1987;79:259-62.
 40. Harvei S, Kravdal O. The importance of marital and socioeconomic status in incidence and survival of prostate cancer: an analysis of complete Norwegian birth cohorts. *Prev Med* 1997;26:623-32.
 41. Tsitouras PD, Martin CE, Harman SM. Relationship of serum testosterone to sexual activity in healthy elderly men. *J Gerontol* 1982;37:288-93.
 42. Johnson RE, Nahmias AJ, Magder LS, et al. A seroepidemiologic survey of the prevalence of herpes simplex virus type 2 infection in the United States. *N Engl J Med* 1989;321:7-12.
 43. Lacey CJ. Assessment of exposure to sexually transmitted agents other than human papillomavirus. In: Muñoz N, Bosch FX, Shah KV, et al., eds. *The epidemiology of cervical cancer and human papillomavirus*. Lyon, France: International Agency for Research on Cancer, 1992:93-105. (IARC scientific publication no. 119).
 44. Neipel F, Fleckenstein B. The role of HHV-8 in Kaposi's sarcoma. *Semin Cancer Biol* 1999;9:151-64.
 45. Anagnostopoulos I, Hummel M. Epstein-Barr virus in tumours. *Histopathology* 1996;29:297-315.
 46. Centifanto YM, Kaufman HE, Zam ZS, et al. Herpesvirus particles in prostatic carcinoma cells. *J Virol* 1973;12:1608-11.
 47. Centifanto YM, Kaufman HE. In vitro transformation by HSV-2 from a human prostatic carcinoma. Lyon, France: International Agency for Research on Cancer, 1975:195-7.
 48. Centifanto YM, Zam ZS, Kaufman HE, et al. In vitro transformation of hamster cells by herpes simplex virus type 2 from human prostatic cancer cells. *Cancer Res* 1975;35:1880-6.
 49. Centifanto YM, Drylie DM, Deardourff SL, et al. Herpesvirus type 2 in the male genitourinary tract. *Science* 1972;178:318-19.
 50. Haid M, Sharon N. Immunofluorescent evidence of prior herpes simplex virus type 2 infection in prostate carcinoma. *Urology* 1984;24:623-5.
 51. Boldogh I, Baskar JF, Mar EC, et al. Human cytomegalovirus and herpes simplex type 2 virus in normal and adenocarcinomatous prostate glands. *J Natl Cancer Inst* 1983;70:819-26.
 52. Sanford EJ, Geder L, Laychock A, et al. Evidence for the association of cytomegalovirus with carcinoma of the prostate. *J Urol* 1977;118:789-92.
 53. Serfling U, Ciancio G, Zhu WY, et al. Human papillomavirus and herpes virus DNA are not detected in benign and malignant prostate tissue using the polymerase chain reaction. *J Urol* 1992;148:192-4.
 54. Monini P, de Lellis L, Fabris M, et al. Kaposi's sarcoma-associated herpesvirus DNA sequences in prostate tissue and human semen. *N Engl J Med* 1996;334:1168-72.
 55. Blackburn DJ, Levy JA. Human herpesvirus 8 in semen and prostate. *AIDS* 1997;11:249-50.
 56. Staskus KA, Zhong W, Gebhard K, et al. Kaposi's sarcoma-associated herpesvirus gene expression in endothelial (spindle) tumor cells. *J Virol* 1997;71:715-19.
 57. Sitas F, Carrara H, Beral V, et al. Antibodies against human herpesvirus 8 in black South African patients with cancer. *N Engl J Med* 1999;340:1863-71.
 58. Choo CK, Ling MT, Chan KW, et al. Immortalization of human prostate epithelial cells by HPV 16 E6/E7 open reading frames. *Prostate* 1999;40:150-8.
 59. Rabkin CS, Biggar RJ, Melbye M, et al. Second primary cancers following anal and cervical carcinoma: evidence of shared etiologic factors. *Am J Epidemiol* 1992;136:54-8.
 60. Rosenwaike I. Causes of death among elderly Jews in New York City, 1979-1981. *Int J Epidemiol* 1994;23:327-32.
 61. McNicol PJ, Dodd JG. Detection of papillomavirus DNA in human prostatic tissue by Southern blot analysis. *Can J Microbiol* 1990;36:359-62.
 62. Anwar K, Nakakuki K, Shiraishi T, et al. Presence of *ras* oncogene mutations and human papillomavirus DNA in human prostate carcinomas. *Cancer Res* 1992;52:5991-6.
 63. McNicol PJ, Dodd JG. Detection of human papillomavirus DNA in prostate gland tissue by using the polymerase chain reaction amplification assay. *J Clin Microbiol* 1990;28:409-13.
 64. McNicol PJ, Dodd JG. High prevalence of human papillomavirus in prostate tissues. *J Urol* 1991;145:850-3.
 65. Ibrahim GK, Gravitt PE, Dittrich KL, et al. Detection of human papillomavirus in the prostate by polymerase chain reaction and in situ hybridization. *J Urol* 1992;148:1822-6.
 66. Dodd JG, Paraskevas M, McNicol PJ. Detection of human papillomavirus 16 transcription in human prostate tissue. *J Urol* 1993;149:400-2.
 67. Wideroff L, Schottenfeld D, Carey TE, et al. Human papillomavirus DNA in malignant and hyperplastic prostate tissue of black and white males. *Prostate* 1996;28:117-23.
 68. Masood S, Rhatigan RM, Powell S, et al. Human papillomavirus in prostatic cancer: no evidence found by in situ DNA hybridization. *South Med J* 1991;84:235-6.
 69. Effert PJ, Frye RA, Neubauer A, et al. Human papillomavirus types 16 and 18 are not involved in human prostate carcinogenesis analysis of archival human prostate cancer specimens by differential polymerase chain reaction. *J Urol* 1992;147:192-6.
 70. Strickler HD, Burk R, Shah K, et al. A multifaceted study of human papillomavirus and prostate cancer. *Cancer* 1998;82:1118-25.
 71. Terris MK, Peehl DM. Human papillomavirus detection by polymerase chain reaction in benign and malignant prostate tissue is dependent on the primer set utilized. *Urology* 1997;50:150-6.
 72. Noda T, Sasagawa T, Dong Y, et al. Detection of human papillomavirus (HPV) DNA in archival specimens of benign prostatic hyperplasia and prostatic cancer using a highly sensitive nested PCR method. *Urol Res* 1998;26:165-9.
 73. Serth J, Panitz F, Paeslack U, et al. Increased levels of human papillomavirus type 16 DNA in a subset of prostate cancers. *Cancer Res* 1999;59:823-5.
 74. Strickler HD, Schiffman MH, Shah KV, et al. A survey of human papillomavirus 16 antibodies in patients with epithelial cancers. *Eur J Cancer Prev* 1998;7:305-13.
 75. Hisada M, Rabkin CS, Strickler HD, et al. Human papillomavirus antibody and risk of prostate cancer. *JAMA* 2000;283:340-1.
 76. Dillner J, Knekt P, Boman J, et al. Sero-epidemiological association between human papillomavirus infection and risk of prostate cancer. *Int J Cancer* 1998;75:564-7.
 77. Dillner J, Knekt P, Schiller JT, et al. Prospective seroepidemiological evidence that human papillomavirus type 16 infection is a risk factor for oesophageal squamous cell carcinoma. *BMJ* 1995;311:1346.
 78. Bjorge T, Hakulinen T, Engeland A, et al. A prospective, seroepidemiological study of the role of human papillomavirus in esophageal cancer in Norway. *Cancer Res* 1997;57:3989-92.
 79. Lagergren J, Wang Z, Bergstrom R, et al. Human papillomavirus infection and esophageal cancer: a nationwide seroepidemiologic case-control study in Sweden. *J Natl Cancer Inst* 1999;91:156-62.
 80. Strickler HD, Kirk GD, Figueroa JP, et al. HPV 16 antibody prevalence in Jamaica and the United States reflects differences in cervical cancer rates. *Int J Cancer* 1999;80:339-44.
 81. Carter JJ, Koutsky LA, Hughes JP, et al. Comparison of human papillomavirus types 16, 18, and 6 capsid antibody responses following incident infection. *J Infect Dis* 2000;181:1911-19.
 82. Chang Y, Cesarman E, Pessin MS, et al. Identification of herpesvirus-like DNA sequences in AIDS-associated Kaposi's sarcoma. *Science* 1994;266:1865-9.
 83. Rotola A, Monini P, DiLuca D, et al. Presence and physical state of HPV DNA in prostate and urinary tract tissues. *Int J Cancer* 1992;52:359-65.
 84. Sarkar FH, Sakr WA, Li YW, et al. Detection of human papillomavirus (HPV) DNA in human prostatic tissues by poly-

- merase chain reaction (PCR). *Prostate* 1993;22:171-80.
85. Tu H, Jacobs SC, Mergner WJ, et al. Rare incidence of human papillomavirus types 16 and 18 in primary and metastatic human prostate cancer. *Urology* 1994;44:726-31.
 86. Moyret-Lalle C, Marçais C, Jacquemier J, et al. *ras*, p53 and HPV status in benign and malignant prostate tumors. *Int J Cancer* 1995;64:124-9.
 87. Suzuki H, Komiya A, Aida S, et al. Detection of human papillomavirus DNA and p53 gene mutations in human prostate cancer. *Prostate* 1996;28:318-24.
 88. Anderson M, Handley J, Hopwood L, et al. Analysis of prostate tissue DNA for the presence of human papillomavirus by polymerase chain reaction, cloning, and automated sequencing. *J Med Virol* 1997;52:8-13.
 89. Gherdovich S, Barbacci P, Mitrone MP, et al. Detection of the human papillomavirus in hyperplastic and cancerous prostatic tissue with PCR. (In Italian). *Minerva Urol Nefrol* 1997;49:73-7.