1. INTRODUCTION

Cervical cytological screening, as first proposed by Papanicolaou, is one of the most successful cancer prevention strategies ever devised (1). Nevertheless, cervical cancer is the second or third most frequent cancer and cause of cancer deaths in women (2). This paradox has arisen because many of the world’s women either do not have access to an effective screening program or do not properly comply with an existing program (1). The problem of access is particularly acute in developing countries, where the high cost of screening limits access to only the wealthiest subset of women (3). Consequently, cervical cancer is the leading cause of cancer death in many developing countries (2). Countries with effective screening programs spend a substantial amount of health care financial resources on them. The cost of screening and follow-up of abnormal Papanicolaou (Pap) smears was recently estimated to cost more than $5 billion annually in the United States alone (National HPV & Cervical Cancer Prevention Resource Center; http://www.ashastd.org/hpvccrc/background.html). Thus cervical cancer prevention strategies that are both more accessible and acceptable to women need to be developed.

Infection with certain types of sexually transmitted human papillomaviruses (HPVs) is the central cause of cervical cancer, providing an opportunity to implement new approaches to cervical cancer prevention (4). One approach is to screen directly for high-risk HPV infections of the cervix (5,6). This strategy is based on the finding that 99% of cervical cancers are HPV DNA positive (7), and that persistent cervical HPV DNA is the major risk factor for progression to cervical neoplasia (8–10). Detection of high-risk HPV DNA has been extensively evaluated as a substitute for or adjunct to cervical cytologic screening. HPV DNA-based screening strategies appear to have higher sensitivity and greater reproducibility relative to Pap screening, although they generally have lower specificity for neoplastic disease, especially in younger women (6,11,12). Because of the effort needed to train competent cytologists and the limited number of samples that a cytologist can process daily, it also may be easier to introduce an effective HPV DNA-based program in settings that currently lack comprehensive screening programs. However, it is unclear whether HPV DNA screening can be made affordable to women in the lowest resource settings that are at highest risk of cervical cancer (3). In addition, since HPV DNA testing would require collection of a cervical specimen, it is unlikely to increase compliance among women who choose not to participate in Pap screening programs.

The causal link between HPV infection and cervical cancer also raises the possibility that cervical cancer could be prevented with an antiviral vaccine. Vaccines against other viral diseases are among the most effective and inexpensive public health intervention measures (13), leading to eradication of smallpox and control of polio, rubella, and several other viral infections. It is widely believed that vaccines against the major HPV
types that cause cervical cancer offer the best hope for cervical cancer prevention in developing countries (see http://www.who.int/vaccines-documents/DocsPDF99/www9914.pdf for a World Health Organization report). Effective HPV vaccines could reduce incidence, cost and morbidity associated with prevention programs in developed countries as well.

Two distinct classes of vaccines could be used to prevent cervical cancer (14). The first type would prevent HPVs from infecting the cervix. Antibodies are believed to be the main immune effectors of immunoprophylaxis for other viral vaccines (15). Vaccine-induced antibodies to virion surface epitopes bind the virions and prevent them from infecting target cells in a process called virus neutralization. Since it takes years to decades for primary HPV infection to progress to cancer, it should be possible to prevent cervical cancer by inducing regression of established HPV-induced lesions before they undergo malignant progression (16). Immune effector mechanisms for therapeutic vaccines are largely cell-mediated responses rather than antibody-dependent. It is very unlikely that viral antibodies will induce regression of HPV lesions, since HPVs are nonenveloped viruses that assemble in the nucleus, and HPV infection does not appear to expose sufficient numbers of intact viral proteins on the cell surface to serve as targets for antibody-mediated effector mechanisms. Both prophylactic and therapeutic HPV vaccines are under active development for cervical cancer prevention (17). In addition, some strategies attempt to generate both neutralizing antibodies and cell-mediated responses to non-virion HPV proteins, thereby generating combined prophylactic/therapeutic vaccines.

2. PROPHYLACTIC VACCINES

Most currently licensed viral vaccines are based on live attenuated virus strains or inactivated virus preparations (13). These approaches are not reasonable for developing prophylactic HPV vaccines. As papillomaviruses cannot be efficiently grown in cultured cells (18), the viruses cannot be mass-produced, even if they could be attenuated. Also, HPVs that cause cervical cancer encode oncoproteins in their genomes (19). The theoretical possibility of vaccine-induced carcinogenesis would preclude using a live or inactivated virus vaccine that contained the viral genome in healthy young people, the probable target group for a prophylactic vaccine.

2.1. Preclinical Studies

Early studies in animal models made it clear that neutralizing antibodies to papillomaviruses primarily recognized conformation-dependent epitopes on the virion surface. Although intact virions were excellent inducers of neutralizing antibodies, subunit vaccines based on denatured forms of the L1 major virion protein, or peptides thereof, were not (20,21). However, approx 10 yr ago, papillomavirus L1s were found to assemble into virus-like particles (VLPs) if they were expressed from a strong heterologous promoter in the absence of all other viral gene products (22). VLPs retain the structural features and high immunogenicity of authentic virions, but are noninfectious since they are composed of a single viral protein and contain no viral genes. HPV VLPs have been produced in mammalian and insect cells, yeast, and even to a limited extent in bacteria (23–27). Prophylactic vaccine candidates now in clinical trials are based on L1 VLPs.

Papillomavirus infections are species-restricted, and experimental inoculation of HPVs does not induce productive infection or disease in any animal model (28). Therefore, prophylactic vaccine studies have involved VLPs from animal papillomaviruses and their corresponding host species. Unfortunately, no domestic mouse papillomavirus has been isolated. This has limited using the best-characterized immune response model in prophylactic papillomavirus vaccine studies. Prevention of experimental papillomavirus infection after VLP vaccination has been demonstrated in rabbit, dog, and cow models using L1 VLPs and challenge virus of cottontail rabbit papillomavirus, canine oral papillomavirus, and bovine papillomavirus type 4, respectively (29–32). Low-microgram doses of VLPs produced excellent protection against high-dose challenge with the homologous virus type in each experimental model, and adjuvant was not required. However, no protection was seen after vaccination with a heterologous VLP type. Protection could be passively transferred to naive animals via serum, indicating that neutralizing antibodies alone can prevent infection.

A number of studies have addressed the question of whether antibodies (raised in mice or rabbits) against VLPs of one HPV type will neutralize other HPV types in in vitro infectivity assays. The question of cross-protection is important because more than a dozen HPV types have been found repeatedly in cervical cancers. Cross-protective responses could greatly simplify vaccine development and manufacture. Unfortunately, cross-neutralizing antibodies were infrequently detected. Limited cross-neutralization was detected for some closely related types (e.g., HPV16/HPV31 and HPV18/HPV45), but the cross-neutralizing titers were always much lower than titers