

Review Article

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HPV & HPV vaccination: Issues in developing countries

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Cervical cancer is the second-most common cancer in women worldwide causing most cancer related deaths in women in developing countries including India. The most predominant etiological factor for cervical cancer is persistent infection of certain high-risk types of human papillomaviruses (HR-HPVs), while low-risk types are associated with benign cervical lesions and genital warts. In India, the most common (98%) oncogenic types are HPV types 16 and 18 with HPV 16 exclusively (80-90%) prevalent. Two recently developed virus-like particle (VLP) based prophylactic HPV vaccines, quadrivalent Gardasil (HPV 16/18/6/11) and Cervarix (HPV 16/18) offer great promise. Several other therapeutic vaccines are also in clinical trials and are yet to establish their efficacy. The use of already developed VLP vaccines in resource-poor regions is limited by several factors, most importantly the high cost of the vaccine. Therefore efforts are being made in India to develop cost-effective second-generation vaccines. Besides cost, there are several socio-cultural and ethical issues involved with the implementation of already developed vaccines including the acceptability of HPV vaccination by preadolescent girls and their parents in India.

Key words Cervical cancer - human papillomavirus (HPV) - issues in India - prophylactic vaccines - therapeutic vaccines

Introduction

Recently, the two new HPV vaccines “Gardasil” and “Cervarix” have been shown to be highly immunogenic and effective in preventing infection with high-risk HPV types 16 and 18, the two most common oncogenic types associated with the development of cervical cancer in women. Though, the prospect of preventing cervical cancer is promising, the implementation of HPV vaccination programme in India’s national approach to cervical cancer control is complex due to several socio-cultural and economic issues. There is a need to look

at issues of implementation and effectiveness of these vaccines in developing and resource-poor regions of the world along with the future prospects of second generation HPV vaccines in relation to worldwide HPV vaccination programmes.

Cervical cancer and papillomaviruses

Cervical cancer is the most common gynecological cancer among women worldwide. There are estimated 493,000 new cases and 274,000 deaths due to cervical cancer in 2002 globally but more than 80 per cent cases

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are occurring from developing countries¹. In India, the annual incidence of cervical cancer is about 130,000 cases with 75-80,000 deaths². Thus India shares about one fourth of the global cervical cancer burden. A large number of risk factors are known to contribute to high incidence of this disease but most important of them are early age of marriage, multiple sexual partners, multiple pregnancies, poor genital hygiene, smoking, use of oral contraceptives and multiparity³. But the most important factor has been considered to be the infection of human papillomaviruses (HPVs). Several lines of clinico-epidemiological studies have demonstrated worldwide that 99 per cent of cervical tumors are having the presence of HPV and cervical cancer develops due to the persistent infection of oncogenic HPV types. In addition, there are several cofactors including the role of host cellular and genetic factors, immunodeficiency, HPV variants, viral load and viral integration are also considered to be important during progression and development of cervical cancer⁴.

Till date, more than 111 genotypes of HPV have been described, but only about 30 of them are associated with anogenital cancer. Mainly HPV types 16 and 18 are considered as most prevalent "high risk" types for cervical cancer while HPV types 6 and 11 are considered to be the most prevalent low risk-types associated with benign lesions and genital warts. There are at least thirteen more high-risk HPV types (31,33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73 and 82) and three probable high-risk types (26, 53, 66)⁵. HPV 16/18 is estimated to account for about 70 per cent of all cervical cancers and altogether HPV 16, 18, 45, 31, 33, 35, 52, 58 are responsible for about 90 per cent of all cervical cancers worldwide⁶. In addition to the most prevalent low-risk HPV types HPV 6 and 11, other types are 40, 42, 43, 44, 54, 61, 70, 72 and 81⁷. The data from all over world showed that HPV types 16 and 18 are present in more than 70 per cent in cervical cancer cases⁸, but the prevalence of HPV type 16 in India is found to be exclusively very high (~90%)⁹⁻¹¹ while occurrence of HPV type 18 varies from 3-20 per cent, followed by other high risk types such as HPV 45, 33, 35, 52, 58, 59 and 73^{11,12}. HPV 16/18 is estimated to account for more than 80 per cent of invasive cervical cancer including CIN 3, vulvar intraepithelial neoplasia (VIN) 2/3 and for 50 per cent of CIN 2 lesions. In India, 85-90 per cent cervical cancer cases are squamous cell carcinoma but only 10-15 per cent cases are adenocarcinoma. Interestingly, in India HPV 16 is the most prevalent type both in squamous cell

carcinoma as well as adenocarcinoma while global reports indicate preferential occurrence of HPV 18 in adenocarcinoma^{13,14}.

Region specific prevalence of HPV varies in India and the most consistent variation has been observed in the prevalence of HPV 16 rather than other types. This regional variation may be due to genetic, cultural and ethnic diversity as well as heterogeneity between studies¹⁵. Occurrence of an exclusive high prevalence of HPV16 puts India in an advantageous position because both the new vaccines are against HPV 16 and 18. These will have maximum impact in India, as they would be able to take care of about 90 per cent of cases.

A comparative study of age related incidence of high risk-HPV infection and cervical cancer in Indian women. Das *et al*¹⁵ showed that the peak of HPV infection appears at the later age as compared to that of western countries. This information is very important for HPV vaccine programme in India for selection of age of target population¹⁵.

HPV vaccines

Prophylactic vaccines

Recently developed two successful prophylactic HPV vaccines - quadrivalent 'Gardasil' (HPV 16/18/6/11) by Merck and bivalent 'Cervarix' (HPV 16/18) by Glaxo SmithKline (GSK) are recommended by US Food and Drug Administration (FDA) for vaccinating young adolescent girls at or before onset of puberty. These viral capsid proteins based vaccines are produced in the form of spontaneously reassembled virus-like-particles (VLPs) expressed either in yeast for 'Gardasil' or in baculovirus for 'Cervarix'. These two vaccines give protection from infection with two most common cancer-causing HPV types 16 and 18 which are associated with more than 70 per cent of cervical cancer cases. Clinical trials have shown the vaccines to be highly immunogenic, well tolerated and safe, highly effective in preventing incident and persistent HPV infections including development of precancerous lesions¹⁶⁻²⁰.

Each 0.5 ml recommended dose of Gardasil contains 20 µg each of HPV 6 and HPV 18 and 40 µg each of HPV 11 and HPV 16 with amorphous aluminium hydroxyphosphate sulphate as adjuvant. This vaccine is given in a series of three 0.5 ml intramuscular injections over six months with 0, 2 and 6-month schedule²⁰. Available data^{16,17,20} indicate that the three dose regime

can induce a high level of antibody formation a level to prevent infection that persists for more than 48 months post-vaccination. The overall efficacy of this tetravalent vaccine was found to be 96 per cent in preventing persistent infection with the HPV genotypes included in the vaccination¹⁷. Gardasil was also found to be 96 per cent effective in preventing genital wart formation and served 100 per cent protection against vulvar and vaginal intraepithelial neoplasia (grade 2-3)²¹⁻²⁴. Five years follow-up studies showed persistence of neutralizing antibodies but beyond that an approximate decline of 10 folds over the first 2 years occur after the first peak and can prevent type-specific persistence of HPV infection. Certainly a longer follow up and monitoring is required to defend that antibody titer²⁵ is sufficient to prevent infection. This vaccine is now approved by the US-FDA for human vaccination and is being introduced for adolescent vaccination in several schools in the US^{26,27}.

GSK's vaccine 'Cervarix' given intramuscularly in 0.5 ml dose at 0, 1 and 6 month schedule contains 20 µg each of HPV 16 and 18. It has a proprietary adjuvant ASO4 comprising of 500 µg of aluminium hydroxide and 50 µg of 3-deacylated monophosphoryl lipid A which induced a stronger and sustained immune response¹⁸. Extended follow up studies have showed that more than 98 per cent seropositivity was maintained for HPV 16/18 antibodies¹⁸. Randomized control trial data of 4.5 years showed an efficacy of about 91.6 per cent against incident infection¹⁹. This vaccine found to control not only the HPV types contained in it, but also infections by other HPV types especially genetically closely related types such as HPV 45 (vaccine efficacy 60%) and to a lesser extent, HPV types 31 and 52 (efficacy 32-36%)²⁸.

These VLP-based vaccines are obviously not therapeutic but the vaccine trial data showed that there is an anamnestic response to a single dose of vaccine in previously infected subjects¹⁷. The basis of this immune response is not very clear. It is also suspected that 10-30 per cent of other high-risk HPV types (31, 35, 39, 45, 51, 52, 56, 58) in cervical cancer may take lead because of change in microenvironment, as the two present vaccines are not giving protection for these HPV types¹⁵.

Therapeutic vaccines

Some preventive HPV vaccines showed lot of promise but are ineffective in the elimination of pre-existing infection and HPV-related disease²⁹.

Therefore, the development of therapeutic vaccines for HPV is warranted because there is an estimated 5 million women worldwide already infected with HPV, which will develop into invasive cervical cancer³⁰. Therapeutic vaccines are the bridge for the temporal deficit by attacking already persistent HPV infections and to treat cervical cancer in women. Although prophylactic vaccines appear to be successful, but it would take decades to perceive the benefits because it takes 10-20 years to develop invasive cervical cancer¹⁵. However, major theoretical obstacle to develop such vaccines is that the immunological determinants for viral persistence or regression remain poorly defined, although it is clear that hosts with impaired cellular immunity are at increased risk of persistent HPV infection and carcinogenic progression.

The choice of target antigen is very important for designing therapeutic vaccines. Since the HPV-encoded E6 and E7 are essential for transformation and are co-expressed in HPV-associated lesions, these proteins represent ideal targets for the development of HPV therapeutics. Several attempts are also being made to deliver HPV E6 and E7 oncoproteins or derived peptides as vaccines using recombinant viruses, and fusion constructs with potentially adjuvanting TLR agonists or cytokines³¹⁻³⁴. However, these papillomavirus proteins are not expressed on the cell surface, there is little potential for antibody-dependent cytotoxicity to mediate regression. Instead, potentially effective cytotoxic responses will probably require a vaccine that induces the presentation of small virally encoded peptides to antigen presenting cells. In cells that possess class I molecules, the normal process of partial intracellular degradation of cytoplasmic or nuclear viral proteins can, following the binding of small viral peptides to the class I molecules, lead to the induction of antigen-specific reactivity of CD8-positive cytotoxic T lymphocytes (CTLs).

Chimeric vaccines

An ideal HPV vaccine should be able to generate both humoral and cell-mediated immunity to prevent new infections as well as to eliminate established HPV infection. Therefore, in order to incorporate the therapeutic intervention along with prophylactic vaccination, several attempts were made to develop chimeric vaccines^{35,36}. Clinical trial of chimeric VLPs (L2E7E6 fusion protein vaccine) had shown to enhance HPV type 16, E6 and E7-specific T-cell immunity in healthy volunteers through vaccination with TA-CIN³⁶.

These vaccines may be relevant for a population who do not go for routine screening but already have HPV related cervical disease.

Second generation vaccines / DNA vaccines

DNA vaccines are an attractive approach for development of second-generation HPV vaccine. DNA vaccines generate effective CTL and antibody responses by delivering foreign antigen to antigen presenting cells (APCs) that stimulate CD4+ and CD8+ T cells. Since DNA vaccine is a plasmid-based vaccine, it is cost-effective and simple to produce in large quantities. Robustness and stability of DNA vaccine even in higher temperature provide an advantage over the other vaccines, particularly for distribution in the remote areas in resource poor settings.

In the last few years, several DNA vaccines have been developed against variety of viral, bacterial as well as parasitic infections in animal models showing long lasting immunity and protection³⁷⁻⁴². Clinical trials of DNA vaccines have been done and/or are underway for various diseases, including cancer, influenza, hepatitis B, HIV, and malaria⁴³⁻⁴⁹. Recently, first DNA vaccine against West Nile virus has been approved by the Department of Agriculture, United States (USDA) for commercial use in horse⁵⁰. Both parenteral and oral immunization with plasmid DNA expressing HPV 16 L1 showed systemic and mucosal antibody production together with cytotoxic T lymphocyte responses in animal models⁵¹. Encouraging data from animal models have led to several therapeutic HPV DNA vaccines in clinical trials⁵²⁻⁵⁴. Since HPV 16 is the type exclusively prevalent in India (~90%) and the other aggressive oncotype is HPV 18 (3-19%), the development of HPV DNA vaccine efforts is being focused on these two high-risk genotypes only.

HPV vaccination associated issues in developing countries

There are a number of limitations and issues for implementation of two successful VLP-based prophylactic HPV vaccines in India. The high cost of the present vaccines is the major concern for mass vaccination program in India as cervical cancer is the major cancer among women mainly from low socio-economic status. So, for low-resource countries, the vaccination with current vaccines will not only possible without substantial reduction in vaccine prices.

Other debatable issue is the selection of target population for vaccination. There are several reports

showing strong immunological response with higher antibody titer during pre-puberty than that in post-puberty⁵⁵ and since HPV infection is acquired through sexual contact particularly those with promiscuous sex life, vaccination of adolescents in India will raise many moral, social, religious and ethical issues because pre-marital sex is not socially acceptable. Preliminary survey of parents of 9-16 yr old school girls suggests that majority of them are unaware of HPV and perceive that their children are not at risk of acquiring HPV as they come from good family (Das *et al*, unpublished). Some parents think that this vaccine will make sex safe leading to freedom to promiscuity and teenage sex, which is not very common in this region of the globe. This will cause social stigma and tarnish their family prestige. They suspect that the vaccine itself may also cause infection to the children. So it is most important to raise general awareness about HPV, de-stigmatizing the HPV infection and gaining acceptance for mass vaccination of pre-adolescent and adolescent girls before it is introduced in India. So the potential strategies may include vaccination of school girls (which may miss the more vulnerable out-of-school girls) or through mother-daughter initiatives or other existing community outreach programs. Although boys do not develop cervical cancer, they can become infected with HPV and can develop other HPV-associated disease such as penile, anal, and oral cancers and genital warts. Some experts believe that vaccinating both males and females would benefit women because women are infected by male sexual partners, but the cost-effectiveness of vaccinating both genders is under investigation. Furthermore, there is still no evidence that vaccinating males reduces the risk of HPV transmission to their female partners^{56,57}.

It is also important to note that the prevalence of cervical cancer and HPV infection in India indicate that the initiation as well as peak of HPV infection occurs at a slightly higher age group (26-35 yr) women mostly in their third decade of sexual activity than that of global scenario (peak in 18-25 yr)¹⁵. And, most cervical cancers in India occur much later by 45-60 yr of age. Therefore, it will be important to test the efficacy of the vaccines in different age groups beyond 26 yr as both Merck and GSK vaccines are equally effective in women between the age 26-55 yr¹⁵. There is, however, no data at present to show effectiveness of these vaccines in the older age group women. So, at this time, such vaccination is not recommended that sexually active older women. Rather, cervical screening is the best approach for this group⁵⁸.

Effective training of health care workers - with clear, realistic and practical goals is crucial in any health program. Health care workers including gynaecologists and pediatricians in many developing countries may not have a clear understanding of biology of HPV infection and its role in development of cervical cancer and its prevention. Health workers need to be educated about how to help patients to understand the enormous advantages offered by both screening and vaccination^{56, 59-61}.

Conclusions

The cervical cancer mortality rates in developing countries including India could conceivably be reduced to the low levels achieved by industrialized countries by combining HPV vaccination with improved screening, diagnosis and treatment. The goal will not be reached without: (i) cooperative efforts of public and private sector-partners together with community leaders; (ii) strengthened health systems, including routine screening for cervical cancer; (iii) availability of HPV vaccine in affordable price through development of effective second generation vaccine; and (iv) a supportative social climate.

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