From causality to Prevention: The case of cervical cancer

Nubia Muñoz

Worldwide, cervical cancer is the second most common cancer in women, with about half of million cases being diagnosed every year and over 80% of these cases occurring in developing countries. It is the second most common cause of death from cancer among young women, accounting for nearly 300,000 deaths annually (1). Its main public health importance lies in the fact that it affects relatively young poor women, devastating their families and being an important cause of lost years of life in the developing world. This cancer reflects more than any other cancer the substantial inequities that exist in health. A major discovery in human cancer etiology has been the recognition that cervical cancer is a rare consequence of an infection by some mucosatropic types of Human Papillomavirus (HPV). In Public Health terms, the importance of this finding is comparable to the unveiling of the association between cigarette smoking and lung cancer, or between chronic infections with Hepatitis B or Hepatitis C viruses and the risk of liver cancer. This discovery has led to great advances in the prevention of this disease on two fronts: (i) primary prevention by the use of prophylactic HPV vaccines; and (ii) secondary prevention by increasing the accuracy of cervical cancer screening.

Although already 166 years ago Rigoni Stern thought that a sexually transmitted agent could be linked to cervical cancer, only during the last 25 years the human papillomavirus (HPV) has been identified as main cause of this cancer.

I have had the privilege of being one of the scientists that participated in this discovery. My first observation goes back to 1974 when I tried to link the high prevalence of giant condyloma with the high incidence of cancer of the cervix and of the penis in Recife, Brazil. In collaboration with Gerard Orth from the Pasteur Institute in Paris we looked for HPV particles in biopsies from giant condylomas, from cervical cancer and from cancer of the penis. Unfortunately, since HPV can not be grown in vitro, at that time, electron microscope was the only technology available to look for the virus in tissues. A few particles were seen in the condylomas but not in the genital cancers. Today we know that once the cancer is established, HPV viral particles are not longer present in the malignant cells, but fragments from its genes. In the same samples we looked for HSV-2 DNA in collaboration with Harald zur Hausen, with negative results. In the late 1970s Harald zur Hausen proposed that HPV may be one of the initiators of the carcinogenic process in the cervical epithelium (2). He, Lutz Gissmann and other scientist from his group, subsequently conducted groundbreaking research that led to the molecular characterization of HPV DNA isolated from cervical cancer samples (3-4). The demonstration of this series of molecular events was essential for the scientific community to accept that HPV infection was the likely cause of cervical cancer. Reasons for scepticism at the time came from observations in his own laboratory and in others that HPV infection was quite ubiquitous and, as such, it could not plausibly be a cause of disease, since a large proportion of asymptomatic women harboured HPV DNA in their cervices. In addition, formal epidemiological evidence of an association between HPV and cervical cancer was lacking at that time (5). HPV natural history studies have now revealed that HPVs are the commonest of the sexually transmitted infections in most populations. Most HPV exposures result in spontaneous clearance without clinical manifestations and only a small fraction of the infected persons, known as chronic or persistent carriers, will retain the virus and progress to precancer and cancer. Molecular characterization and cloning of the first HPV types in the early 1980s (3), made possible the development of hybridization assays to look for HPV gene fragments in human tissue.

Using PCR-based hybridization assays at my former Unit at the International Agency for Research on Cancer (IARC) we undertook the following epidemiological studies to investigate the role of HPV in cervical cancer:

Case- control studies

In 12 countries around the world we studied a total of 2,500 women with cervical cancer and 2,500 control women without cancer. These women were interviewed using a standardized questionnaire to elicit information on risk factors for cervical cancer and underwent a gynecological examination to collect cervical cells from the tumours and normal cervices for the detection of HPV DNA of 30 HPV types that infect the genital tract. The prevalence of HPV DNA was over 95% in the tumor cells of women with cervical cancer and it ranged from 5 to 20% in normal cervical cells of control women.

These prevalences correspond to Odds Ratios (ORs) of over 100 indicating a very strong association between HPV and cervical cancer. The magnitude of the ORs allowed an epidemiological classification of 15 HPV types as carcinogenic or high-risk types, 12 as low-risk types and 3 types as probably carcinogenic (6-7). This epidemiological classification correlates quite well with the phylogenetic classification based on sequencing of L1 gene.

Our case-control studies also allowed the identification of the following cofactors that acting together with HPV increase the risk of progression from HPV persistent infection to cervical cancer: tobacco, high parity, long term use of oral contraceptives and past infections with herpes simplex virus type 2 and Chlamydia trachomatis. (8-9). In addition, this studies contributed to establish the important role of male sexual behavior in the risk of developing cervical cancer (10-11).

Survey of HPV types in invasive cervical cancers

Over 1,000 women with invasive cervical cancer from 22 countries around the world were included in this study. HPV DNA detection with PCR-based assays revealed that 99.7% of the cases were HPV-positive. This finding led us to propose for the first time that HPV was not only the main cause of cervical cancer, but also a necessary cause (12). No other cancer has been shown to have a necessary cause.

The above two studies made possible the estimation of the proportion of cervical cancer cases attributable to the main HPV types in the various geographical regions. These estimates are being used to estimate the impact of preventive strategies based on HPV (13).

Implications

The demonstration that infection with certain types of human papillomavirus (HPV) is not only the main cause but also a necessary cause of cervical cancer has led to great advances in the prevention of this disease on two fronts:

(i) In primary prevention by the use of prophylactic HPV vaccines.

Two safe and efficacious prophylactic HPV vaccines have been developed using viral like particles (VLPs); the quadrivalent vaccine (Gardasil) contains VLPs of HPV 16 and 18, responsible for about 70% of cervical cancers, and VLPs of HPV6 and 11 that cause about 90% of genital warts. The bivalent vaccine (Cervarix) contains only VLPs of HPV16 and 18. Both vaccines have been shown to have a high efficacy for the prevention of high-grade precancerous lesions of the cervix (CIN2/3) and this protection has been shown to last at least 5 years (14-16). The quadrivalent vaccine has been shown in addition to prevent high-grade precancerous lesions of the vulva and vagina caused by HPV16 and 18 and genital warts caused by HPV 6 and 11 (14, 16). Universal vaccination of adolescent girls offers a great potential for the prevention of cervical cancer. Both vaccines have been licensed in over 100 countries but their high price limits their accessibility in the countries that need them most; it is hoped that a special price for developing countries could be negotiated with the pharmaceutics companies (17).

(ii) In secondary prevention by increasing the accuracy of cervical cancer screening.

Several studies have shown that HPV DNA detection assays are more sensitive than cytology for detection of high grade precursor lesions of the cervix (CIN2/3) and suggest that they should be used as primary screening test followed by triage with cytology or visual inspection (18). Evidence suggests that if the current HPV vaccines were introduced into developing countries and combined effectively with appropriate secondary cervical screening strategies, the lifetime risk of developing cervical cancer could be reduced as much as 60%. Mathematical models have shown that if the cost per vaccinated girl is less than $25, HPV16/18 vaccination would be very cost-effective in all 33 Latin American countries (19). The current price of the commercially available HPV tests is also the main barrier for their widespread introduction in developing countries. It is hoped that a fast and inexpensive HPV test developed with funds from the Gates foundation, will shortly be commercially available (20).

References

1. Ferlay J, Bray F, Pisani P, Parkin DM. GLOBOCAN 2002: Cancer Incidence, Mortality and Prevalence Worldwide IARC CancerBase No. 5. version 2.0, IARCPress, Lyon, 2004.

2. zur Hausen H. Condylomata acuminate and human genital cancer. Cancer Res 1976 Feb;36(2 pt 2):794

3. Gissmann L zur Hausen H. Human papillomavirus DNA: physical characterization and genetic heterogeneity. Proc Natl Acad Sci USA 1976 Apr;73(4): 1310-3

4. Durst, M Gissmann L and zur Hausen H. A papillomavirus DNA from a cervical carcinoma and prevalence in cancer biopsy samples from diferent geographical regions. Proc Natl Acad Sci USA 1983 Jun80(12):3812-15

5. Muñoz N, Bosch X, Kaldor JM. Does human papillomavirus cause cervical cancer? The state of the epidemiological evidence. Br J Cancer. 1988 Jan;57(1):1-5.

6. Muñoz, N., Bosch, F.X., de Sanjosé, S., Tafur, L., Izarzugaza, et al. (1992) The causal link between human papillomavirus and invasive cervical cancer: a population based case control study in Colombia and Spain. Int. J. Cancer, 52, 743 49.

7. Muñoz, N., Bosch, F.X., de Sanjosé, S., Herrero, R., et al for the International Agency for Research on Cancer Multicenter Cervical Cancer Study Group (2003) Epidemiologic classification of human papillomavirus types associated with cervical cancer. N. Eng. J. Med, 348(6): 518-27.

8. Muñoz, N., Franceschi, S., Bosetti, C., Moreno, V., Herrero,et al., for the International Agency for Research on Cancer (IARC) Multicentric Cervical Cancer Study Group (2002) Role of parity and human papillomavirus in cervical cancer: the IARC multicentric case-control study. Lancet 2002, 359, 1093-101

9. Muñoz, N., Castellsagué, X., de Gonzalez, A.B., Gismann, L. (2006) Chapter 1: HPV in the etiology of human cancer. Vaccine, 24(3), 1-10.

10. Muñoz, N., Castellsagué, X., Bosch, F.X., Tafur, L., de Sanjosé, S., Aristizabal, N, Ghaffari, A.M. & Shah, K.V. (1996) Difficulty in elucidating the male role in Colombia, a high-risk area for cervical cancer. J. Natl Cancer Inst., 88, 1068-75.

11. Castellsagué, X., Bosch, F.X., Muñoz, N., Meijer, C.J.L.M., Shah, K.V., de Sanjosé, S., Eluf-Neto, J., Ngelangel, C., Chichareon, S., Smith, J.S., Herrero, R., Franceschi, S., for the International Agency for Research on Cancer Multicenter Cervical Cancer Study Group. (2002) Male circumcision, penile human papillomavirus infection, and cervical cancer. New Engl. J. Med. 346(15): 1105-1112

12. Walboomers, J.M.M., Jacobs, M.V., Manos, M.M., Bosch, F.X., Kummer, J.A., Shah, K.V., Snijders, P.J.F., Peto, J., Meijer, C.J.L.M. & Muñoz, N. (1999) Human papillomavirus is a necessary cause of invasive cervical cancer worldwide. J. Pathol., 189, 12-19.

13. Muñoz, N., Bosch, F.X., Castellsagué, X., Diaz, M. de Sanjosé, S., Hammouda, D., Shah, K.V. & Meijer, C.J.L.M. (2004) Against which human papillomavirus types shall we vaccinate and screen? The international perspective. Int J Cancer, 111, 278–285

14. Ault KA; Future II Study Group. Effect of prophylactic human papillomavirus L1 virus-like-particle vaccine on risk of cervical intraepithelial neoplasia grade 2, grade 3, and adenocarcinoma in situ: a combined analysis of four randomised clinical trials. Lancet. 2007 Jun 2;369(9576):1861-8.

15. Paavonen J, Jenkins D, Bosch FX, Naud P, Salmeron J, Wheeler CMet al. Efficacy of a prophylactic adjuvanted bivalent L1 virus-like-particle vaccine against infection with human papillomavirus types 16 and 18 in young women: an interim analysis of a phase III double-blind, randomised controlled trial. Lancet 2007;369(9580):2161-70.

16. Villa LL, Costa RLR, Petta CA, Andrade RP, Paavonen J, Iversen O-E, et al. High sustained efficacy of a prophylactic quadrivalent human papillomavirus types 6/11/16/18 L1 virus-like particle vaccine through 5 years of follow-up. Br J Cancer 2006 Nov 21;95(11):1459-66.

17. Muñoz, N., Franco, E.L., Herrero, R., Andrus, J.K., de Quadros, C.A., Bosch, F.X. (2008) Recommendations for Cervical Cancer Prevention in Latin America and the Caribbean. Vaccine 2008;26(11), L96-L107

18. Cuzick J, Clavel C, Petry KU, Meijer CJLM, Hoyer H, Ratnam S, et al. Overview of the European and North American studies on HPV testing in primary cervical cancer screening. Int J Cancer 2006;119:1095–105.

19. Goldie SJ, Diaz M, Constela D, Alvis N, Andrus JK, and Kim SY Mathematical models of cervical cancer prevention in Latin America and the Caribbean. Vaccine 2008;26(11), L59-L72

20. Qiao YL, Sellors J, Eder PS et al . Anew test for cervical cancer screening in developing regions : a cross-sectional study of clinical accuracy in rural China. Lancet Oncology Vol9 Oct 2008:929-36