

Third Faculty of Medicine

Prevention of Cervical Cancer

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SUMMARY

Cervical cancer is one of the few cancers where incidence and mortality have been falling within the last decades. It is natural to look at this in combination with the simple screening tool used for detection. A simple test can prove the early stages of cancer, and by this detection, early treatment can be initiated and death prevented.

National screening programs have been started in Norway and many other countries, which both inform women about risk factors for cervical cancer and symptoms of the disease and screen them for early detection of the cancer at regular intervals. Combined, these preventive measures have been proven to reduce both the incidence and mortality of cervical cancer in the countries where they have been introduced.

Many studies have shown that the most probable cause of cervical cancer is the presence of Human Papilloma Virus (HPV). Recently, pharmaceutical companies have developed vaccines against the most common causative agents of cervical cancer – HPV-16 and HPV-18. The introduction of this vaccine, mainly to young girls before their sexual debut (9-12 years), is expected to reduce the incidence even further. Some countries have already included this vaccine in their national vaccination program, whereas in other countries the vaccine has to be purchased by the individual girl/woman. The vaccine has to be taken in three doses and the cost is still quite high. So, in countries where the vaccine is not included in the vaccination program or covered by the insurance, the availability of this vaccine is limited.

INTRODUCTION

Cervical cancer is still one of the leading causes of cancer mortality among women globally, predominantly in less developed countries. In Norway, 300 women are diagnosed with cervical cancer every year, and 100 women die every year from this type of cancer. In the US, cervical cancer strikes about 10,000 women a year and causes up to 4,000 deaths. According to the World Health Organization, there were 500 000 new cases of cervical cancer in 2005.

In our time, the evolution of cervical cancer management is an exciting part of medicine where we have seen great improvement. After the introduction of cytological Papanicolaou (Pap) testing the cervical cancer deaths have been considerably reduced over the last 5 decades. It has for a long time been accepted that cervical cancer generally progresses through a series of stages prior to its development. Through careful monitoring of at-risk women, clinicians can identify and remove dysplastic cells before they progress to cervical cancer.

Identifying and removal of dysplastic lesions has been very successful but is far from perfect. The examination can be uncomfortable for the woman, it brings with it a slight elevated risk for birth-related complications, and the woman herself must be compliant. Even in countries with highly organized screening programs, lack of attendance remains a major cause of cervical cancer incidence. Ironically, another major problem related to this form of prevention is over-treatment. Some studies have shown that whilst Pap screening is effective, far too many dysplastic lesions are removed without any obvious benefit; meaning that many of the precursor lesions would not have progressed into cervical cancer. This of course relates to our lack of understanding as to how cervical cancer actually develops.

Up until 1995, it was up to the individual woman in Norway to get tested for cervical cancer. The number of tests taken in 1994 was 542,666 (Norway has about 5 million inhabitants), but the percentage of women taking the test at regular intervals was only about 70%. So, in 1995 the government started a large screening program for all women nationwide in the ages between 25 and 69. The results were registered in a cytology register and all women were reminded every three years to go for a new screening test. The goal was not to increase the number of tests taken, but to increase the percentage of women taking the screening test to at least 80%.

In the 10-year period since the start of the nationwide screening program of cervical cancer, both the incidence and the mortality of the disease have decreased. By comparison, the incidence of cervical cancer in the period 2000-04 was about half of the incidence in 1970-74. We also assume that the underlying risk is bigger today than in the 1970s, which indicates that the absolute effect of the program is bigger than 50%.

Most researchers support the theory that cervical cancer is fundamentally a symptom of viral infection. The virus in question – *Human Papillomavirus* (HPV) – resides in the cervical epithelia and usually only cause nothing more than occasional benign lesions. HPV is one of the most common sexually transmitted infections and up to 80% of women will contract this infection over their lifetime. Yet, very few women will develop cervical cancer. This shows that HPV is not a sufficient cause but it does

appear to be a necessary cause. So what characteristics of this usually harmless virus allow it to become deadly?

Certain factors determine the HPV's ability to cause cervical cancer. These appear to be HPV type, HPV persistence, HPV load, physical state of HPV, and possibly the existence of multiple HPV infections. Other co-factors have also been implicated, such as smoking, HLA type, parity, and sexual behavior, although many of these (and other) co-factors are likely to work in combination with one or more of the 'main' HPV-related determinants.

This knowledge has led us to believe that if we can control HPV, we can control cervical cancer. Based on this, pharmaceutical companies have developed vaccines against two of the main HPV types associated with cervical cancer (HPV-16 and HPV-18). The two vaccines that are now on the market are Gardasil, which protects against HPV-16 and HPV-18 and also includes protection against the two main causes of genital warts, HPV-6 and HPV-11, and Cervarix, which protects against HPV-16 and HPV-18. Gardasil was approved for sale in EU countries in the fall of 2006 and in Norway in September the same year.

In 2007, the health ministry of Norway decided to include the vaccine in the states vaccination program for children. This will probably not eradicate cervical cancer, but will undoubtedly have a considerable impact on cervical cancer development and related mortality.

REVIEW OF CURRENT KNOWLEDGE

The Cervix

The narrow part of the uterus is called the cervix and extends to the top of the vagina. (Figure 1) The name is derived from the Latin word for 'neck', referring to the neck of the womb.

The cervix is predisposed to the formation of cancer because of the high cell turnover that occurs in an area referred to as the transformation zone. In this area of constant epithelial shedding, differentiation of basal and parabasal cells must occur in order to replenish the epithelial layer. It is within these totipotent cells that the path towards cervical cancer begins.

Figure 1: The Cervix.

Cervical Intraepithelial Neoplasia

By using Pap smear testing, cervical squamous cell carcinoma can be detected and managed at an early stage because it exhibits a well-defined pre-malignant phase. This pre-malignant phase is characterized by various stages through which cells proceed prior to the onset of invasive disease. These stages are accompanied by numerous measures of cancer-related changes in the proliferating primitive cells, such as nuclear abnormalities; including nuclear size, number of mitoses, and presence of abnormal mitotic forms.

Basal and parabasal cells form on the basement membrane of the dermis and develop, through a process of differentiation, into flat squamous cells which comprise the squamous epithelium. It is during this process that the carcinogenic process occurs and subsequently is the basis for the cervical intraepithelial neoplasia (CIN) staging system (Figure 2). The first stage of CIN, namely CIN 1, applies when the proliferation of parabasal-like cells is confined to the lower one-third of the epithelium. If the proliferating cells extend into the middle third of the epithelium, they are marked as CIN 2. Once these cells involve the upper third of the epithelium, they are considered to be severe dysplastic lesions (CIN 3). If they replace the entire thickness of the epithelium, they are termed carcinoma *in situ*. Cervical squamous cell carcinoma occurs when the proliferating cells gain the ability to invade the stroma.

Figure 2: Cervical Intraepithelial Neoplasia.

Cervical Cancer Globally

Cervical cancer is decreasing in developing countries, but is still widespread in developing countries (Figures 3 and 4). This makes cervical cancer the second most common cancer amongst women globally. Age-standardized incidence rates (ASR) per 100,000 women range from 5.8 in Western Asia, up to 42.7 in Eastern Africa.

These correspond to observed mortality rates (age- standardized) of 2.9 per 100,000 in Western Asia and 34.6 per 100,000 in Eastern Africa. In Northern Europe the age-standardized incidence is relatively low (9.0 per 100,000), as is the mortality rate (3.6 per 100,000).

Figure 3: Worldwide Cervical Cancer Incidence Rates.

Figure 4: Worldwide Cervical Cancer Mortality Rates.

The Effectiveness of Papanicolaou Testing

The lack of effective Pap smear screening strategies and adequate access to medical care generally attributable to the large disparity between cervical cancer incidence in 'Developed' and 'Developing' countries. Indeed, when women migrate from regions of high cervical cancer incidence (with well organized cytological screening) to those with low incidence rates, they may acquire rates, which more closely resemble those of the new host country. This highlights the effectiveness of Pap smear screening. Despite the unequivocal benefits associated with the use of Pap smear screening, there are however many controversial issues regarding its specific implementation.

Probably foremost in the debate on Pap smear screening are the benefits of organized versus opportunistic screening, and frequency of testing. It is generally held true that organized screening reduces the incidence and mortality of cervical cancer, although this has been disputed. The frequency of testing required to maintain effective prevention of cervical cancer is a little less obvious. In a review on this topic, it was noted that many European countries conducting organized screening at 3-yearly intervals possessed similar cervical cancer related mortality to those countries employing 5-yearly screening. In some countries (Austria and Germany), which performed annual testing, the mortality rate was even higher than in some neighboring countries performing 3- or 5-yearly testing (Belgium, Italy, Netherlands and France). Obviously it is difficult to directly compare mortality from different countries in this way since they potentially have underlying differences in risk for this disease. Nevertheless, it suggests that the use of 5-yearly screening does not in itself appear to decrease the effectiveness of screening.

A second issue of importance in the use of Pap smear screening to prevent cervical cancer is its lack of specificity. The percentage of severe dysplasia/cervical cancer *in situ* lesions that are thought to progress towards cervical cancer has been estimated at more than 12%. The corollary of this is that the majority of lesions ordinarily removed as a matter course in cervical cancer prevention, would not have progressed towards an invasive endpoint. Whilst the benefits attributable to Pap smear testing have been considered to outweigh its distinct lack of specificity, identification of an intermediate (and more specific) predictor of progression would be immensely useful. HPV-related factors such as HPV persistence, HPV load, physical state of HPV, and multiple HPV infections, may prove to be useful in this regard, although specifically which one (and how they can be utilized) continues to be a matter for discussion.

Human Papillomavirus – A Necessary Cause of Cervical Cancer

The role of Human Papillomavirus (HPV) in cervical cancer development has been studied in depth, culminating in the conclusion that the presence of certain HPV types is necessary for the development of cervical cancer.

HPV is not considered to be a sufficient cause of cervical cancer. During their lifetime, more than 50% of women worldwide will acquire a genital HPV infection. Clearly, this far exceeds the likelihood for women developing cervical cancer, which suggests that HPV in itself is not a sufficient cause. A possible reason for this is that one or more co- factors are also necessary to trigger cervical cancer formation. This may be through alteration of the host environment (e.g., immune system), direct carcinogenic potential (e.g., creation of genotoxic adducts), or interaction with HPV in another way. It could also be that the use of blunt measures for HPV exposure may be inadequate for identifying a sufficient cause. For example, different HPV types exhibit differing oncogenic potentials. The use of an oncogenic HPV exposure would be deemed more sufficient than a non-oncogenic type. Likewise, perhaps a particular strain of an oncogenic HPV type might later be found to exhibit properties, which make it a sufficient cause.

More recently, researchers have attempted to refine their knowledge by trying to determine exactly what conditions are necessary for HPV to cause cervical cancer. Most studies now focus upon particular characteristics of specific HPV infections that predispose individuals towards cervical cancer development.

Human Papillomavirus Type

HPV typing was one of the first discriminating tools used to gain a more accurate portrayal of HPV's involvement as a causal factor in cervical cancer development. This was an important step since, of the 100 or more HPV types identified this far, 40 different HPV types are known to infect the genital tract. A large number of studies have investigated the prevalence of different HPV types in various countries. Others have quantified the risks they impose upon cervical cancer incidence. These (and other) studies have been vital in assigning carcinogenic potential to different HPV types. Of the 40 HPV types inhabiting the genital tract, at least 14 are considered to be significantly associated with the progression to cervical cancer and are referred to generally as high risk, or oncogenic, HPV types.

One of the key findings gained from studies on individual HPV types was the considerable role of HPV-16 and HPV-18 in cervical cancer development. These types are generally considered to confer the greatest risk for cervical cancer development, responsible for an estimated 70% of cervical cancer. This knowledge paved the way for development of vaccines against HPV-16/18 to combat cervical cancer.

Human Papillomavirus Load

As a further step towards the isolation of characteristics of HPV infections that might predict carcinogenic potential more accurately, a number of researchers have

investigated the potential role of HPV load. One landmark study in this field utilized a population-based case-control study in Sweden to assess the association between HPV-16 load and CIS. These researchers found that high HPV load, present in cervical smears taken within a year prior to diagnosis of CIS, placed women at a 43-fold increased risk for CIS compared to HPV negative women. This was in distinct contrast to the 3.1-fold increased risk (compared to HPV negative women) seen in women whose last smear within a year prior to diagnosis contained low HPV-16 load.

Human Papillomavirus Persistence

Most HPV infections are considered transient and will be cleared or suppressed by cell-mediated immune mechanisms within one or two years following initial exposure. As one might expect, there is a positive relationship between the lack of clearance (persistence) of an HPV infection and subsequent development of cervical lesions. Persistence of HPV infections has gained great acceptance among many in the HPV research community as a marker for high cervical cancer risk. As with HPV load, persistence of HPV infections appears to signify a much greater risk for cervical cancer development than HPV presence alone.

Human Papillomavirus Integration

One particularly interesting marker for increased cervical cancer risk is the physical state of HPV in the host. HPV DNA may exist in episomal (extra-chromosomal) or integrated (within the host cell genome) forms. During a normal life cycle, high-risk HPV genomes replicate as episomal molecules. However, integration of HPV viral DNA into the host cell genome is frequently observed in cervical carcinomas and in a subset of CIN3 lesions. This is not entirely unexpected since the current understanding of HPV's carcinogenic potential revolves around the disruption of a regulatory HPV gene (E2), thought to occur mainly as a result of integration into the host cell genome.

HPV Pathogenesis

The natural life cycle of HPV involves infection of basal or para-basal cells (presumably accessed through micro-abrasions of the epidermis), replication of its own DNA during basal cell differentiation and subsequent amplification of its DNA to high copy numbers, along with capsid protein synthesis and viral assembly, in the differentiated keratinocytes.

The HPV life cycle is controlled by relatively few viral proteins and the virus must utilize host cell factors to regulate viral transcription and replication. Of particular interest are the HPV E6, E7 and E2 proteins. The E6 and E7 proteins are coded for in a region referred to as the long control region (LCR). These two proteins are produced early in the viral cycle and are used to deregulate the host cell growth cycle. They do this by binding with certain tumour suppressor proteins, cell cyclins, and cyclin-dependant kinases, effectively inactivating them. Two of these inactivated proteins – the tumour suppressor protein p53 and the retinoblastoma gene product pRB – are major players in cell growth. HPV E6 proteins bind to p53 and ensure its degradation through ubiquitination, whereas HPV E7 proteins bind to hypophosphorylated pRB forms and abrogates their ability to form a complex with

the cellular transcription factor E2F-1. Once liberated, E2F-1 initiates transcription of genes required for the cell to enter S-phase of the cell cycle. The outcome of these (and other) activities by E6 and E7 proteins is continuous cell proliferation and stimulated DNA synthesis. Subsequently, after a period where E5 proteins encourage continued proliferation and delayed differentiation of the host cells, DNA binding proteins (E2) are translated, which block transcription of E6 and E7 proteins. At this point, E1 proteins are able to bind to the viral origin of replication, which enables extrachromosomal replication. Furthermore, due to E2's antagonism of E6 and E7, regulatory elements of the host's cell cycle (such as p53 and pRB) return to normal and allow the differentiation process to continue. Viral particles are then assembled in the host's nucleus and mature HPV virions are later released when the cells are shed.

It would appear that HPV has no need (or desire) to transform cells. On the contrary, evidence suggests that HPV generally has a vested interest in ensuring the host cells resume their normal course. For HPV, it is important that the host cells continue to differentiate since some of its late-stage development can only occur in differentiating cells. So what happens? It would appear that the problem lies in disruption of the E2 protein. Integration of HPV DNA into the host's DNA is often accompanied by disruption of the E1 or E2 genes. Loss of expression of the E2 gene leads to a loss of E2 protein function, which corresponds to an increased expression of the E6 and E7 proteins. Effectively, this results in increased proliferation of the host cell and genomic instability. Consequently, the host cell accumulates a greater amount of damaged DNA and has a reduced ability to repair it. This process may finally result in the accumulation of mutations, which allow fully transformed cancer cells to form.

HPV Transmission

It is common knowledge that the main route for transmission of HPV is through sexual intercourse. However, reports of unusually high incidence rates of HPV infection in children raise the issue of alternative methods for transfer of the virus. Numerous studies have reported the presence of HPV in oral cavities and genitalia of children and infants, as described recently in a review by Cason and Mant. The likely mode of HPV transmission in these cases is cause for debate however. Certainly, a proportion of these cases could be attributable to sexual abuse, although presumably this is less likely in infants than older children. Another possibility is the acquisition of HPV in utero, intrapartum or postpartum. Evidence has accumulated in favour of this 'vertical' form of transmission although some studies have been unable to confirm this.

The route of transmission raises important issues, which may influence the effectiveness of HPV vaccination programs. The currently held dogma is that vaccination should be introduced to females prior to sexual debut since efficacy is likely to be compromised if an HPV infection is current. Furthermore, it is not known how *previous* infections may influence vaccine effectiveness.

Risk factors

Presence of HPV infection cannot be considered a sufficient causative agent due to the numbers of HPV infected women who do not develop cervical cancer. The roles of other potential risk factors in cervical carcinogenesis therefore need to be considered.

- Smoking - An increased risk of cervical cancer associated with tobacco smoking has been established on the basis of a number of epidemiological studies since the 1980's. Whether this link is related to genotoxic DNA adducts of smoking in the cervix epithelium, its effect on malignant transformation of HPV infected cells, or its influence on HPV infections via localized immunosuppression, has been discussed. The risk is also increased in women with exposure to environmental tobacco smoke. The incidence of cervical cancer in women who smoke is 3 times higher than in non-smoking women.
- Old age – women aged 60 and above are at increased risk of cervical cancer as they are less willing or able to seek medical care for screening of this disease and participate in treatment.
- Early age at first intercourse (16 years and younger)
- A history of many sexual partners
- Current or past partner who has had many sexual partners
- A history of genital human papillomavirus (HPV) infection or other sexually transmitted diseases
- The presence of other genital tract neoplasia
- Prior squamous intraepithelial lesion (SIL)
- Long-term use of oral contraceptives
- Immunodeficiency or HIV positivity
- Poor nutrition

Symptoms of cervical cancer

Early cervical cancer may not cause noticeable signs or symptoms. For this reason, all women should have regular check-ups, including a Pap smear to check for abnormal cells in the cervix. Symptoms include pelvic pain, bleeding, unusual vaginal discharge and pain during intercourse.

Pap testing

The Pap test is a screening tool that identifies women likely to have premalignant disease and at high risk for cervical cancer. The recommendation for how often a woman should have a Pap test varies from country to country, but the most commonly recommended interval between tests is 3 years, the first test taken 3 years after the woman first becomes sexually active and no later than the age of 21. To make sure that the test comes back as accurate as possible, women are asked to take a few precautions. 24 hours prior, do not douche, insert a tampon, have intercourse or bath in a tub. The examination should also be done in between menstrual periods.

In Norway, the testing is organized on a voluntary basis, which means that the women themselves are responsible for getting tested. They do, however, receive a letter every

three years after turning 25, reminding them to go for a checkup.

Resurge have shown that regular testing reduces the incidence of cervical cancer and hence also the mortality. In developed countries, women have easy access to testing and prompt medical treatment, which has resulted in the reduction of cervical cancer cases in these countries compared to the developing countries where testing and treatment is not so easily accessible.

HPV vaccine

The most commonly used vaccine is Gardasil. The EU and The U.S. Food and Drug Administration, approved this vaccine in 2006, and several countries, including Norway, have now included this vaccine in the national vaccination program.

Gardasil is a recombinant vaccine (contains no live virus) that is given as three injections over a six-month period. The second dose is given two months after the first dose, followed four months later by the third dose.

The cervical cancer vaccine is recommended for girls ages 11 to 12, although it may be used in girls as young as age 9. This allows a girl's immune system to be activated before she's likely to encounter HPV. Vaccination at this age also allows for the highest antibody levels. The higher the antibody levels, the greater the protection. Experts recommend a catch-up immunization for girls and women ages 13 to 26 who haven't been vaccinated or who haven't completed the full vaccine series.

Four studies, one in the United States and three multinational, were conducted in 21 000 women to show how well Gardasil worked in women between the ages 16 and 26 by giving them either the vaccine or placebo. The results showed that in women who had not already been infected, Gardasil was nearly 100 percent effective in preventing precancerous cervical lesions, precancerous vaginal and vulvar lesions, and genital warts caused by infection with the HPV types against which the vaccine is directed. While the study period was not long enough for cervical cancer to develop, the prevention of these cervical precancerous lesions is believed highly likely to result in the prevention of those cancers.

The benefit of the vaccine for sexually active women who have already been exposed to the HPV virus is still under discussion. Some clinical trials have shown the vaccine to have some benefit and so far, no serious adverse effects of the vaccine have been proven. The most common complaint is soreness at the injection site, the upper arm. Low-grade fever or flu-like symptoms are also common, but the effects are usually mild.

Prognosis for cervical cancer

If cervical cancer is detected in early stages, then prognosis for 5-year survival rate is almost 100%. The overall 5-year survival rate average for all stages is 73%.

CONCLUSION

Cervical cancer is still one of the leading causes of cancer death in women worldwide. But by following a few simple preventive measures the numbers of deaths from cervical cancer can, and have already been reduced.

Get a regular Pap smear – The Pap smear can be the greatest defense for cervical cancer. The Pap smear can detect cervical changes early before they turn into cancer.

Limit the amount of sexual partners – Studies have shown women who have many sexual partners increase their risk for cervical cancer. They also are increasing their risk of developing HPV, a known cause of cervical cancer.

Quit smoking or avoid secondhand smoke – Smoking cigarettes increases your risk of developing many cancers, including cervical cancer. Smoking combined with an HPV infection can actually accelerate cervical dysplasia.

If sexually active, use a condom – Having unprotected sex puts one at risk for HIV and other STDs, which can increase the risk for developing cervical cancer.

Follow up on abnormal Pap smears – It is important to follow up with regular Pap smears or colposcopies.

Get the HPV vaccine – All women under 27 may be eligible to receive the HPV vaccine.

Some countries have today large national screening programs for cervical cancer with proven effect of reduction of both incidence and mortality of this cancer type.

Recently, many of the same countries have included the HPV vaccine in their vaccination program, The overall effect of this vaccine is to early to tell, but it is expected to be substantial.

Unfortunately, a vaccine does not solve the global problem of cervical cancer and regular Pap smears, this is a solution, to this date, mostly valid for developed countries. Our biggest challenge is to inform women about what they can do for themselves to prevent the development of cervical cancer and to make medical prevention, testing and treatment cheaper and more available. The prevention measures work, it is just a matter of reaching a larger number of women.

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