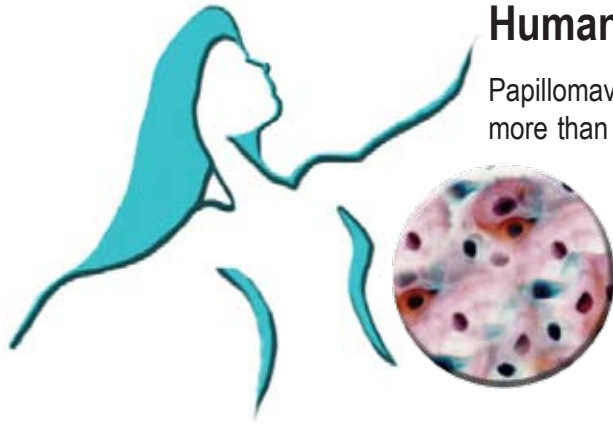


HPV Type-Detect



Human Papillomavirus (HPV)

Papillomaviruses are a diverse group of viruses that have been found in more than 20 different mammalian species, as well as birds and reptiles.

In the 1980s, the newly developed techniques of molecular biology enabled the detection of many human papillomaviruses (HPVs) in benign and malignant lesions. To date, there are more than 200 HPV subtypes. An HPV type is defined as a complete genome whose L1 gene sequence is at least 10% dissimilar to that of any other HPV type.

EPIDEMIOLOGY

- Over 40 HPV types infect the anogenital tract, 15 of them have been classified as high-risk for development of cervical cancer, 3 have been classified as probable high-risk, 12 have been classified as low-risk and 3 are considered as undetermined-risk.
- HPV types associated with an increased risk of malignancy vary by geographic location. For example, HPV-16 is found in 77% of cervical cancers in Germany, 71% in South America, 59% in the United States, but only 33% to 39% in Japan.
- HPV infection shows a peak prevalence in women below age 25, a decrease among women aged 35-54, and a second peak after age 55.
- HPV infection in men: A recent Danish study has found 33.8% HPV prevalence (n=374 male conscripts 18-29 years) by PCR.
- HPV infection was detected in 70% of the male partners of HPV infected women.
- Patients with penile lesions are found 99.5% to be HPV positive.
- In family: The most common HPV profile was high-risk (HR) HPV in all family members (29%), followed by HPV-positive mother-infant pairs (26%). HPV-positive father-infant pairs were less frequent (11%), and in 8% of the families, only the infant was HR HPV positive.
- In infants: HPV DNA was detected in 15% of the genital and 10% of the oral samples at birth, reaching peaks of 18% and 21%, respectively, at 6 months, and declining to 10% by 24 months.
- In adolescents: Cumulative HPV incident rates have been approximately 40% and prevalence rates as high as 80%.

- Despite the rate of anogenital infection, very few HPV infections result in cellular changes, genital warts, intraepithelial neoplasia, or cancer. Most infections (70% – 90%) are contained by the host immune system and become undetectable within 6 to 10 months.
- Infections with multiple HPV subtypes are common and the median duration of infection persistence of a specific HPV type is 168 days.

HPV IN ANOGENITAL CANCERS

- Cervical cancer is the second most common cancer among women world wide, with a mean age standardized incidence rate varying from 11.3 per 100,000 women in more developed countries to 18.7 per 100,000 women in less developed countries. The link between high-risk HPV and the development of cervical cancer is statistically greater than that between smoking and lung cancer.
- The life-time risk of ever contracting HPV is estimated to be 80%. Despite the high prevalence of HPV in cervical cancer, it's a rare event occurring after a long period of viral persistence which reflects the multi-step nature of HPV-induced cervical cancer.
- HR-HPV has been detected in up to 99.7% in cervical squamous-cell carcinomas and 94%–100% of cervical adeno- and the adenosquamous carcinomas.
- HPV type distribution in cervical cancer: Of the more than 40 HPV types found in the genital tract, HPV-16 accounts for some 50% to 60% of the cervical cancer cases in most countries, followed by HPV 18: 10% to 20%, HPV 45: 4%-8%, and HPV 31: 1%-5%. The five most common HR HPVs (16, 18, 45, 31 and 33) account for 80% of the distribution in squamous-cell cancers and of 94% of adenocarcinomas.

HYBRID CAPTURE® 2 HIGH-RISK HPV DNA TEST™

- The specimen is denatured in the laboratory, and the liberated single-stranded DNA is hybridized in a solution with an RNA probe mix consisting of five low-risk genotypes (6, 11, 42, 43 and 44), and 13 high-risk HPV types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 68).
- The resulting bound DNA-RNA “hybrids” are reacted with an antibody directed against the hybrids.
- A chemiluminescent substrate, which binds to the antibody, is added and a signal output is recorded.
- Among women which are 30 to 35 years or older, the sensitivity of a single lifetime hc₂ test for detection of high-grade dysplasia ranges from 80% to 90%, and has a specificity of 57% to 89%.
- The hc₂ assay limitations:
 - hc₂ assay can not identify HPV individual subtypes
 - The detection of the hc₂ assay is about 5,000 HPV genome equivalents which makes it less sensitive than PCR.
 - Cross-reactivity of the two probe cocktails may reduce the clinical relevance of a positive result.
 - Patients with cervical cancer may have poor recovery of HPV because of tumor necrosis and bleeding which may alter the test results.
 - Unable to detect HPV-HR Type 66.



OneSwab™

HPV TYPE-DETECT

- The sample is collected from the endocervix and ectocervix with the swab which is then placed in the proprietary transport medium.
- MDL offers the HPV Type-Detect (HPV-TD), with the viral E6/E7 oncogenes as the primer target region.
- HPV-TD is a nested multiplex PCR which involves multiple rounds of PCR reactions consisting of primary PCR with a large amplicon product and a secondary amplification step to generate a smaller amplicon.
- The HPV-TD multiplex-nested PCR based assay includes many broad-based HPV genotypes within the first PCR amplification reaction and the second round nested PCR consists of all type specific primers, including the high and low-risk.
- The HPV-TD was evaluated in 495 cervical scrapes with various histological findings. The HPV prevalence rate was 34.7% in the absence of CIN (CIN 0), 94.2% in the presence of mild to moderate dysplasia (CIN 1/CIN 2), and 97.8% in the presence of CIN 3. In addition, multiple infections were detected in 48% of the patients.

ADVANTAGES OF HPV-TD

- Since 40% of HPV infections are mixed infections, HPV-TD can be used to differentiate between newly acquired HPV subtypes and pre-existing infections when applied over time.
- HPV Type-Detect can detect 19 specific HPV subtypes.
- Although the genetic variation among HPV types is merely 10%, HPV-TD is highly specific without known cross-reaction to other known HPV types.
- HPV-TD has an analytical sensitivity of 10 HPV genome equivalents per reaction.
- In HPV-vaccine recipients, HPV-TD can be used to monitor successful immunization and patient's stratification as well as to determine if patients have been infected with an HPV genotype other than that protected by the vaccine.

HPV VACCINE

- There are two different types of vaccines: prophylactic vaccines that would elicit an antibody response and prevent infection, and therapeutic vaccines that induce a specific and cell mediated response leading to regression of pre-existing lesions.
- Prophylactic multivalent virus like particle (VLP) vaccines comprising the HPV-16 major capsid protein L1 are in phase III clinical trials with preliminary results from phase II clinical trials indicating safety, plus 100% efficacy from persistent infection, as well as from infectious complications such as cervical dysplasia.
- The Prophylactic HPV-16 L1 VLP vaccine (Merck Research Laboratories) was evaluated in a randomized, double-blind, placebo controlled trial. Among 750 placebo recipients, 12 women developed HPV-16 related CIN 2/CIN 3. Among 755 vaccine recipients, there were no cases of CIN.
- The conclusion of the HPV-16 L1 VLP vaccine is that it provides high-level protection against HPV-16 infection and HPV-16 included CIN 2/CIN 3 for at least 3.5 years after immunization.
- Prophylactic HPV vaccines would be given to children before natural exposure and the onset of sexual activity.
- In the Merck Research Study, a subset group of women that received the vaccine were HPV-16 DNA-positive and anti HPV-16 seronegative at the time of enrollment and were less likely to develop subsequent HPV-16-related CIN 2/CIN 3 than those who received placebo injections.
- The Prophylactic HPV vaccines are effective only against the high-risk HPV-16 infection which accounts for over 50% of HPV HR infections in the US.
- It was found the Prophylactic HPV vaccines did not provide any protection against HPV-seropositive individuals.
- Therapeutic vaccines are very much in their infancy compared with prophylactic vaccines. A number of these vaccines have been studied in phase I/II clinical trials including in women with CIN, vulvar intraepithelial neoplasia (VIN) and advanced cervical cancer.