

ABSTRACT

The use of non-radioactive biotinylated probes for the detection of Human papillomavirus type 16 in cervical scrapes and biopsy specimens

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Cancer of the uterine cervix is one of the most common malignant diseases among women worldwide and seems increasingly prevalent throughout the Caribbean. Although morbidity and mortality rates have declined in the last decade as a direct consequence of comprehensive screening programs, the disease remains an important public health problem.

The strong implication of Human papillomavirus (HPV) as a possible cause of the disease has served as rationale for the implementation of large-scale screening of HPV infection worldwide. The public health benefit of changes in the screening practices for cervical cancer can be easily realized with a sensitive, efficient detection mechanism that recognizes specific oncogenic HPV types. In addition, information on the occurrence of HPV cervical infections in asymptomatic women in Trinidad and Tobago is also lacking.

In this thesis, a safe, rapid detection procedure was used to detect the specific oncogenic HPV, type 16. The presence of HPV nucleotide sequences in genital biopsies and cervical scrapes was examined by hybridization using non-isotopic, random primer, biotin labelled probes representing HPV type 16. Biotin labeling of probes was performed using DNA labeling and a commercially provided phototope detection kit. Hybridization was performed under stringent conditions. The hybrids were detected by using Streptavidin and biotinylated alkaline phosphatase and visualized with a catalyzed chemiluminescent detection reaction. Dot blot hybridization with biotinylated probes proved to be a useful technique for the identification of HPV infection.

Using these hybridization techniques between forty-five (45) and fifty (50) percent of the samples tested was seen to be positive for HPV type 16. The techniques employed were also fast and reliable and can be easily incorporated as part of regular clinical screening procedures.

Positive identification of HPV 16 in genital biopsy specimens and cervical scrapes emphasizes the need to screen for other potentially oncogenic types.

Keywords: Hybridization, dot-blot, oncogene.