Colposcopy vs Hybrid Capture II Assay in detection of cervical human papilloma virus infection

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Summary

Purpose of investigation: Considering the relationship between high-risk human papillomavirus types and the presence or subsequent development of cervical high-grade preinvasive lesions, the aim of the study was to determine if the Hybrid Capture II test can be used to triage women with atypical colposcopic findings.

Methods: The study was carried out on 100 patients with suspicious colposcopy findings (suggestive of human papillomavirus infection) who underwent a cervical smear for human papillomavirus testing DNA Hybrid Capture II and direct biopsies for histopathological analysis.

Results: Sixteen patients were negative for human papillomavirus. Of the eight patients positive for high-risk HPV type, seven presented an abnormal transformation zone grade 2 (high-grade squamous intraepithelial lesion of the cervix at histopathology). There was a significant positivity of medium-high risk virus types in the cases with more abnormal colposcopy ($\chi^2 = 7.44$; $p < 0.005$). Histopathological findings of high-grade squamous intraepithelial lesions were registered in the patients positive for medium-high risk human papillomavirus types ($\chi^2 = 7.66$; $p < 0.025$).

Conclusions: Based on these results it can be concluded that if a diagnosis of a high-grade squamous intraepithelial lesion has been made on the basis of colposcopic and histopathological findings, there is a high probability that the infection was due to one or more types of human papillomavirus. There are necessary further studies to interpretate both the advantages and disadvantages of intermediate triage procedures, like Hybrid Capture II testing, compared with immediate colposcopy.

Key words: Human Papillomavirus; Atypical colposcopic findings; DNA Hybrid Capture II Assay.

Introduction

Human papillomavirus (HPV) infection in the lower female genital tract is now recognised as the principal cause of virtually all cases of cervical cancer worldwide (99.7%) [1]. A number of studies have also demonstrated the relationship between high-risk (HR) HPV types and the presence or subsequent development of high-grade preinvasive lesions (CIN) [2, 3]. The classic model of cervical cancer prevention - primary screening with cytology, followed by diagnostic colposcopically directed biopsy, is undergoing a dynamic change; infact the introduction of HPV DNA testing provides more options but increases the complexity in the sequence of screening, triage and diagnosis [4]. The aim of the study was to determine if the Hybrid Capture II (HC II) test can be used to triage women with atypical colposcopic findings and to compare the detection rate of a biopsy-proven squamous intraepithelial lesion (SIL) using immediate colposcopy and the HC II test.

Materials and Methods

Between June 2003 and November 2004 100 women, who attended the Outpatient Clinic of cervical cancer prevention and colposcopy of our Department because of a cytologic screening result of ASCUS or higher grade of abnormality, were included in the study. Based on a suspicious colposcopic lesion highly suggestive of HPV infection, after verbal informed consent was obtained, a cervical smear for HPV DNA testing using the Hybrid Capture II test (Digene Corp., Beltsville, MD) and directed biopsies on the cervical abnormal transformation zone (ANTZ) were performed. The colposcopic and histopathological lesion was correlated with the HPV type isolated: at low risk (6, 11, 42, 43, 44) and at medium-high risk (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59 and 68). Patients were aged between 18 and 46 years with a mean age of 32.5 years. The cervical specimens were placed into specimen transport medium for HPV HC II testing and centrifuged for 5 min at 1,500 rpm; the pellets were resuspended in cervical specimen buffer of the Bio-Pap HPV typing (kit cod. En 20211) and processed according to the commercially available (second generation microtiter) HC II assay protocol. The colposcopic and histopathological characteristics were then compared with the HPV types using the χ^2 test.

Results

The colposcopic findings of the 100 patients with HPV cervical lesions are summarized in Table 1: of the 20 patients positive for medium-high risk HPV types, nine presented an $ANTZ_{G2}$, in ten there was visualized at colposcopy an $ANTZ_{G0\text{-}1}$, associated with flat condylomatosis; in 31 of the 64 patients positive for a low-risk (LR) HPV type, a simple flat condyloma was visualized; in 28 cases of $ANTZ_{G0\text{-}1}$ a condyloma was associated and in the last five cases a papillary condyloma was diagnosed; 16 patients were negative for HPV testing; one showed a papillary condyloma, 12 a flat condyloma and in three

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Table 1. — *The ratio between colposcopic and HPV findings.*

Colposcopic findings	N	HPV findings Negative	Positive LR	Positive MR-HR
Papillary condylomas	6	1	5	
Flat condylomas	44	12	31	1
$ANTZ_{GO-1}$ + condylomas	41	3	28	10
$ANTZ_{G2}$ + condylomas	9			9
Total	100	16	64	20

$X^2 = 7.44$; $p < 0.005$.

cases with $ANTZ_{G0}$ a flat condyloma was associated. The relation among these data using the χ^2-test evaluation shows significant positivity of medium-high risk virus types in the cases with more abnormal colposcopy ($\chi^2 = 7.44$; $p < 0.005$). The histopathological results of biopsy specimens are summarized in Table 2: 50 of the patients showed HPV-correlated cervical inflammation; in 43 cases LGSIL-HPV was present, seven women evidenced HGSIL-HPV, one of these HGSIL was carcinoma in situ. There was a positive correlation between severity of histopathological diagnosis and medium-high-risk HPV type findings ($\chi^2 = 7.66$; $p < 0.025$).

Table 2. — *The ratio between histopathological and HPV findings.*

Histopathological findings	N	HPV findings Negative	Positive LR	Positive MR-HR
HPV correlated cervicitis	50	13	36	1
LGSIL HPV	43	3	28	12
HGSIL HPV	7	7		
Total	100	16	64	20

$X^2 = 7.66$; $p < 0.025$.

Discussion

HPV-DNA testing is highly sensitive for identifying the subset of women who harbored underlying HGSIL among those with ASCUS cervical cytology [3] and can improve the detection rates of CIN2-3 when added to cervical cytology in routine screening [5]. The HC II commercial HPV detection test was more sensitive for the detection of high-grade cervical lesions than classic cytological screening (98.1% versus 85.3% for cytology, with a specificity of 85.2%) [6]. Our study, using the HC-II assay on a series of selected women on the basis of colposcopic suspicious or abnormal lesions, shows a positive correlation between medium-high risk HPV type, high-grade transformation zone, and high-grade histological lesion. Only in one case positive for high medium-risk HPV type were cytology, colposcopy, and histology negative for atypia. Sixteen patients were negative for the test. These results are in agreement with those of other studies showing that histopathological findings do not correlate with HPV types in cases of low-grade SIL, but there is a greater correlation in high-grade SIL [1, 3, 7, 8]. Only 9% of CIN 3, but 38% of CIN 2 would be missed: lost opportunities to diagnose advanced cases of dysplasia, non transient HPV infections, might be averted if patients are routinely referred for colposcopy after an ASCUS smear result. Adding direct colposcopic visualization to cytologic testing offers the ability to detect lesions in which cytologic and cytologically based viral DNA testing may be falsely negative [9]. When compared to the Hybrid Capture Tube (HCT), HC II was also more often positive in women with negative colposcopic evaluations (29% vs 20%) [8]. The probability of the occurrence of HPV infection is greater if colposcopy findings are chronic cervicitis or papillary condyloma and the most frequent is mosaic [10]; cervical inflammation may be associated with high-grade lesions in women infected with oncogenic HPV [11]. The HC II test would have been a useful adjunct at colposcopy because 29% of women assigned at a negative final diagnosis on the basis of colposcopic evaluation, had positive HC II [8]. On the other hand, in women with negative HC II tests, it is possible that lack of shedding from HPV infected dysplastic epithelium can be overcome by direct visual inspection and lead to the discovery of high-grade lesions [9]. There are necessary further studies to interpret both the advantages and disadvantages of intermediate triage procedures, like Hybrid Capture II testing, compared with immediate colposcopy. In our country, it is less expensive to perform colposcopy than the Hybrid Capture II test. However the HC II test can be used to triage woman with atypical colposcopic findings and to follow viral persistence, because only persistent HPV infection is associated with high-grade CIN [12-14].

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