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The utility of HPV DNA triage in the management of cytological AGC

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Objective: Given the demonstrated utility of human papilloma virus DNA triage in the management of atypical squamous cells of undetermined significance, this study sought to evaluate the potential role of human papilloma virus DNA testing in the evaluation and management of cytological atypical glandular cells.

Study design: Following institutional review board approval, 28 women presenting with cytological atypical glandular cells underwent repeat thin-prep cytology, Hybrid Capture 2 human papilloma virus DNA testing, colposcopic evaluation, Fisher electrosurgical conization, and endometrial sampling. Human papilloma virus test results in each patient were then correlated with histologic lesions, if present.

Results: Sixteen of the 28 study patients had pathologic lesions (11/28 high-grade squamous intraepithelial lesion, 3/28 low-grade squamous intraepithelial lesion, 1/28 adenocarcinoma in situ, 1/28 simple endometrial hyperplasia). Human papilloma virus DNA testing was available in 24 of 28 subjects (86%). The sensitivity of human papilloma virus positivity to predict the presence of cervical intraepithelial neoplasia was 100% (confidence interval 77% to 100%), specificity 64% (confidence interval 35% to 85%), positive predictive value 76%, and negative predictive value 100%. Women who tested human papilloma virus positive were 12 times more likely to have cervical intraepithelial neoplasia than women who were human papilloma virus negative (Fisher $P < .001$). Human papilloma virus positivity was not predictive of endometrial pathology; women who were human papilloma virus positive were less likely to have endometrial pathology than were women who were human papilloma virus negative (risk ratio 1.6, 95% confidence interval 0.01-1.7).

Conclusion: Atypical glandular cells can represent a variety of lesions. The majority of the lesions will be squamous intraepithelial lesions of the cervix (50%), with high-grade squamous intraepithelial lesion present in 40% of subjects. Human papilloma virus DNA testing is a sensitive test for the presence of squamous intraepithelial lesion, with excellent negative predictive

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value for the absence of squamous intraepithelial lesion. The results of this study suggest human papilloma virus DNA testing might be an effective screening test in the initial evaluation and management of cytological atypical glandular cells.

The 2001 Bethesda system classifies glandular cell abnormalities less severe than adenocarcinoma into 3 separate categories: (1) atypical glandular cells (AGCs) not otherwise specified (NOS); (2) atypical glandular cells favor neoplasia (AGC "favor neoplasia"); and (3) endocervical adenocarcinoma in situ (AIS).[1]

The management of glandular cytological abnormalities can be challenging, given the absence of a clear-cut algorithm or widely accepted evidence-based approach to utilize when faced with this uncommon but potentially serious cytological abnormality. Should all women who present with AGCs undergo colposcopic evaluation, endocervical curettage, and endometrial biopsy at initial presentation? If the workup is negative, should these women, regardless of age or parity, then be subjected to mandatory conization?

In the landmark Atypical Squamous Cell (ASC)-US/Low-Grade Squamous Intraepithelial Lesion (LSIL) Triage Study (ALTS),[2] the incorporation of human papilloma virus (HPV) DNA testing for cancer-associated HPV subtypes was found to be a sensitive triage method for the detection of moderate to severe cervical intraepithelial neoplasia (CIN grades 2,3). HPV DNA testing was found to have a greater sensitivity in detecting CIN 3 or more advanced lesions and showed comparable specificity to a single additional cytological test indicating atypical squamous cells of undetermined significance (ASCUS) or greater. By referring only patients with HPV-positive ASCUS to colposcopy, HPV triage was able to detect an equivalent number of high-grade intraepithelial lesions as did the "immediate colposcopy for all" arm and did so while referring 44% fewer patients to colposcopy.

Given the demonstrated utility of HPV DNA triage in the management of cytological ASCUS, this study sought to evaluate the potential role of HPV DNA triage in the evaluation and management of cytological AGCs.

Material and methods

A prospective study was designed to address the potential utility of HPV DNA triage in the evaluation of cytological AGCs and was approved by the University of Virginia Human Investigation Committee (HIC 11202).

Subjects

With the exception of vulnerable patient populations (incarcerated, pregnant, cognitively impaired), all patients seen and evaluated in the colposcopy clinic at the University of Virginia between April 2002 and May 2004 were screened for study enrollment. Five thousand six hundred women were seen during this period of time, of which 37 presented with an index smear of AGCs. Following pathologic review at the University of Virginia, the cytologic diagnosis of AGCs was confirmed in 28 of the 37 patients. These patients subsequently comprised the study cohort. The mean age of the cohort was 32 years (range 19 to 65 years; Table I).

Review of AGC smears

An independent reviewer reviewed the enrollment AGC Papanicolaou tests sequentially in a blinded fashion. Smears diagnosed as AGCs by the initial study pathologist were entered into a database as a confirmed final diagnosis of AGCs. Otherwise, the smear and all other slides were reviewed by a second blinded independent reviewer in a similar manner, and a final smear diagnosis was derived by majority.

Screening examination

Subjects with confirmed AGCs initially underwent a routine gynecologic examination including collection of an ectocervical and endocervical cytological sample; sample material was rinsed into a liquid medium (PreservCyt, Cytyc Corp, Marlborough, Mass) and transported to the cytology laboratory for preparation as thin-layer cytological slides (Thin Prep, Cytyc Corp) as well as for Hybrid Capture 2 HPV DNA testing.

HPV DNA testing

HPV DNA testing was performed at the University of Virginia in the Department of Pathology on masked PreservCyt specimens obtained at the initial screening examination, as previously described. Samples were tested using the Hybrid Capture 2 microplate method (Diagnostic Technology, Belrose, Australia) for 11 oncogenic subtypes, including HPV 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, and 58. Specimens yielding a chemiluminescence equal to or exceeding that of 3 positive controls containing 1.0 pg/mL HPV DNA were considered positive.

Colposcopic evaluation

After obtaining a cytological sample at the time of the initial, the cervix was rinsed with acetic acid and