Cytopathology + More | Primary HPV screening, Pap-HPV cotesting: interim guidance and a retrospective study

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August 2015—The Food and Drug Administration in 2001 approved the use of high-risk HPV testing to triage ASCUS Pap test results (reflex testing). Two years later the FDA expanded the indications for hrHPV testing to include its use as an adjunct to cytology in women over age 30 (cotesting). The rationale for age 30 as a cotesting cutoff point was that hrHPV is common in sexually active young women and most infections are transient and clear without medical intervention. Cotesting has since been widely adopted and multiple FDA-approved testing platforms are being used in the United States. The American Cancer Society, American Society for Colposcopy and Cervical Pathology, and American Society for Clinical Pathology in 2011 released updated screening guidelines that advocated cytology and HPV cotesting as the preferred screening option in women 30 and older. A CAP survey distributed to laboratories in 2013 found that about 60 percent of U.S. laboratories were performing cotesting in 2012.[1]

The Roche Cobas HPV test was approved by the FDA on April 24, 2014 as a primary screening test,[2] as detailed in the August 2014 issue of CAP TODAY.[3] The 2011 screening guidelines addressed the issue of primary HPV screening, stating that "in most clinical settings, women aged 30 years–65 years should not be screened with hrHPV testing alone as an alternative to cotesting at 5-year intervals or cytology alone at 3-year intervals" because there were insufficient data to recommend primary HPV screening at that time. To reconcile the various testing options, 13 experts representing multiple professional societies convened to develop an interim guidance document to address primary HPV screening. The panel sought expert opinion and conducted a literature review, which included review of data from European randomized controlled screening trials, the ATHENA (Addressing THE Need for Advanced HPV Diagnostics) trial, and a Medline query.[4]

The panel agreed to several guiding assumptions: 1) No screening test will detect all prevalent or incipient cervical cancer cases; 2) A desirable sensitive screening test will detect more cervical cancer and precursor lesions (CIN3+) in baseline screening, with reduced detection during subsequent screening rounds; and 3) An increased number of colposcopy procedures is a surrogate measure for negative aspects of screening.[5]

The panel addressed two main and four additional questions. Its members concluded: 1) A negative hrHPV test provides greater reassurance of a low risk of CIN3+ than a negative cytology result; 2) Primary hrHPV screening can be considered to be an alternative to cytology alone and cotesting; 3) For women who are hrHPV positive, a combination of HPV 16/18 genotyping and reflex cytology (for women positive for at least one of the 12 HPV genotypes other than 16/18) is a reasonable approach to managing women and determining which women should receive colposcopy; 4) Following a negative primary hrHPV test, women should be rescreened no sooner than three years; 5) Primary hrHPV screening should not be initiated in women younger than age 25; and 6) Maximum screening benefit can be achieved only by identifying women who are unscreened or under-screened.

The panel reemphasized that of the four FDA-approved hrHPV assays, only one is FDA approved for primary HPV screening and that clinicians should not use an FDA-approved test without a specific primary hrHPV screening

indication. The guidance also discusses the harms and benefits of primary screening of women ages 25–29 and factors that may affect analytic sensitivity, including specimen adequacy, controls, and interfering substances.[5] The panel identified areas of future research, including comparative lifetime effectiveness studies, direct cost comparisons to five-year cotesting methods, and the need for cancer risk data to include several rounds of screening extending five years or longer for ATHENA and similar studies.

A second significant article, colloquially known as the Quest Diagnostics Health Trends study, aimed to provide a real-world retrospective comparison between three screening approaches for cervical cancer.[6] The study evaluated 256,648 samples from women 30 to 65 years of age who had a cotest and a cervical biopsy within one year of each other at Quest Diagnostics from January 2005 to September 2011. The cases were retrieved using a search for CPT terminology for cotest and cervical biopsy. The majority of Pap slides were liquid based—ThinPrep Pap test and ThinPrep Imaging System (Hologic, Bedford, Mass.) and SurePath Pap test and BD FocalPoint Imaging (Becton Dickinson, Burlington, NC)—and a small percentage of slides were prepared using conventional Pap smear. Testing for hrHPV was performed using Digene Hybrid Capture HPV DNA test (HC2) (Qiagen, Gaithersburg, Md.).

Quest Diagnostics validated assay modifications to the HC2 for the detection of HPV in SurePath specimens. A positive Pap test was defined as an interpretation of ASC-US or greater (≥ASC-US). In this population, 74.7 percent of samples (191,776 of 256,648 specimens) were hrHPV positive, 73.8 percent (189,304 of 256,648 specimens) were Pap test positive (regardless of HPV result), and 89.2 percent (229,020 of 256,648 specimens) were positive for at least one test (positive cotest). Biopsy yielded ≥CIN3 in 1.6 percent (4,090 of 256,648 specimens). Of the ≥CIN3 specimens, 63.3 percent (2,589 of 4,090 specimens) were submitted in ThinPrep medium, 33.7 percent (1,377 of 4,090 specimens) in SurePath medium, and three percent (124 of 4,090 specimens) were conventionally prepared. Of the HPV-negative cancers, 15.2 percent (50 of 329) were submitted in ThinPrep medium, and 18.3 percent (36 of 197 specimens) were submitted in SurePath medium. The calculated sensitivities for detection of ≥CIN3 were as follows: positive cotest, 98.8 percent (4,040 of 4,090); positive HPV-only test, 94 percent (3,845/4,090); ≥ASC-US cytology, 91.3 percent (3,734/4,090). The calculated specificities for the three options were cytology only, 26.3 percent; HPV only, 25.6 percent; positive cotest, 10.9 percent. Positive predictive values for cytology only, HPV only, and cotest were 1.97 percent, 2.0 percent and 1.76 percent, respectively, while negative predictive values were 99.5 percent, 99.62 percent, and 99.83 percent.

The study yielded 526 cancers, and 18.6 percent (98/526) of cancers were HPV negative, 12.2 percent (64) were Pap test negative, and 5.5 percent (29) were cotest negative. On average, women with HPV-negative cancers were significantly older than women with any HPV-negative results. The average ages of all women with HPV-negative cervical cancer ThinPrep and SurePath specimens were 52.4 and 52.7 years respectively, while the average ages of HPV-negative women with any ThinPrep and SurePath specimens in the study were 43.5 and 44.2 years respectively. Of the malignant cases, there were 169 cervical origin adenocarcinomas, of which 26.6 percent (45 of 169 specimens) were HPV negative, 20.7 percent (35 of 169 specimens) were cytology negative, and 8.3 percent (14 of 169 specimens) were negative by both tests.

The authors disclosed the limitations of the study. The HC2 is not FDA approved for a primary HPV screening indication or for use on SurePath specimens. This study also did not follow the FDA-approved algorithm to include genotyping. About 75 percent of all cotests Quest performed did not have biopsies analyzed by Quest. Clinical information to include colposcopy findings and intervening testing or treatment was not available.

In summary, based on the results, the authors concluded that a greater number of cancers and precursor lesions would be identified by the HPV-Pap cotesting method than either test alone. The American Cancer Society estimates that 12,360 women are diagnosed with cervical cancer each year.[7] In this study, 19 percent of all cervical cancers were HPV-negative, meaning that 2,400 cervical cancers in the U.S. would be missed, including an even higher proportion of adenocarcinomas.

Readers may be interested in a recent point-counterpoint publication[8] written by a proponent of primary HPV screening and a proponent of Pap-HPV cotesting. The cotesting proponent is one of the coauthors of the Quest

Diagnostics study.

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