Cytopathology and More | The Pap test under fire

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August 2014—The humble Pap test is perhaps one of the most lauded and disdained laboratory tests, lauded because it is the lab test with the best track record of preventing cancer and disdained because the test is labor-intensive, the results are operator dependent, and the regulations are burdensome. Recently the Pap test has come under fire, threatened to be replaced with HPV tests and maligned by patients and physicians for its sometimes unexpected high cost.

Dr. Crothers

When used appropriately, the Pap test is a remarkable laboratory test. In 1917, George Papanicolaou, MD, PhD, a research physician, was studying sex determination through cervical cell changes in guinea pigs. At that time, cervical cancer in women was a leading cause of cancer death, and Dr. Papanicolaou realized his method of cervicovaginal cell collection could be applied to human subjects to identify abnormal cells associated with cancer. In collaboration with Herbert Traut, MD, Dr. Papanicolaou discovered the Pap smear, and the newly formed American Cancer Society in 1948 encouraged its use in detecting and preventing cervical cancer.[1] Now, the Pap test is used primarily to detect the presence of preneoplastic cells (high-grade squamous intraepithelial lesion [HSIL] or cervical intraepithelial neoplasia 2–3 [CIN2–3]) so that the precursor can be eradicated before it becomes invasive carcinoma. Since the Pap test's inception, it has been beneficial in many other ways, including in evaluating hormonal status and in detecting infectious agents such as herpes simplex virus, Candida spp., Trichomonas vaginalis, Actinomyces spp., and bacteria associated with vaginosis. It was through observation of cellular changes in the Pap test that investigators, including Alexander Meisels, MD, first hypothesized the presence of a virus, human papillomavirus, as a possible etiologic agent inducing preneoplastic changes in squamous cells that define "dysplasia."[2] The Pap test has serendipitously been able to detect glandular neoplasms such as endometrial and endocervical carcinoma, and its precursor lesion, endocervical adenocarcinoma in situ—lesions that are often silent and difficult to detect clinically. It may also detect metastatic and recurrent carcinomas in the cervix or vagina of women with known cancer.

One reason for the Pap test's tremendous success was the simple, oft-repeated message that all women should have the test yearly along with a pelvic examination. HPV tests have changed this paradigm, and current practice guidelines[3,4] are more complex, incorporating a woman's age, screening history, and HPV result into interval screening recommendations. Although the Pap test has an irreducible false-negative rate and is subject to interpretive error, its success is due in part to frequent opportunities to detect and eradicate cervical cancer precursors through annual screening. It can be a diagnostic test, but it is designed as a screening test to be used

frequently to sample a large area of the cervix. How it might perform as a triage or diagnostic test is uncertain.

Dr. Tambouret

Despite significant changes in Pap testing technology since the mid-to-late 1990s, including liquid-based collection media, automated processing/computer screening, and improved staining techniques, and rigid government regulation, the advances have only marginally reduced the overall incidence of cervical cancer in the United States.5 New technologies are the reason for the "Pap smear" becoming the "Pap test." Most Pap tests in the United States are now processed from a liquid-based medium, as opposed to having the cells smeared directly from collection onto a glass slide. The annual number of newly discovered cases of cervical cancer leveled off between 2007 and 2011.[5] The main reason the incidence of cervical cancer has not declined further, despite new technologies and the advent of HPV reflex testing for specific abnormal interpretations and HPV cotesting in conjunction with the Pap test in women over age 30, is the number of women who are not screened or not screened frequently enough. The reasons for this are complex; among them are economic factors, lack of access to care, and confusion about screening guidelines. There is evidence that new algorithms have resulted in less-timely cervical cancer screening. The percent of women in the United States 18 years or older who had a Pap test in the past three years dropped in 2010 to 74 percent from the highest screening rate, 81 percent, reported in 2000. The Centers for Disease Control and Prevention's 2020 Healthy People target for cervical cancer screening is to ensure that 93 percent of women ages 21 to 65 have a Pap test within three-year intervals.[6] Women of lower socioeconomic status, who are older, poor, or less educated, are less likely to be screened. Minorities, especially Hispanics and blacks, are at higher risk for cervical cancer and have lower screening rates.[6]

One of the Pap test's advantages has been its low cost. In its heyday, the Pap test was a loss leader, provided at a price below its market cost in return for other contracted, more profitable services, such as surgical biopsy cases. This resulted in an undervaluing of the Pap test and reimbursement as low as $5 per test in some laboratories, not enough to cover the cost of the product. A basic Pap test now costs $25 to $40, depending on how it is processed, except when additional tests are added to it. Additional testing "off-the-vial" has become a marketing mantra because of the convenience of performing additional tests from the remaining liquid medium after a single patient visit for a Pap test. In a New England Journal of Medicine commentary, Cheryl Bettigole, MD, MPH, a family practice physician, lamented the "thousand-dollar Pap smear."[7] Her patients had complained that their routine Pap test was resulting in unexpected out-of-pocket costs as high as hundreds of dollars. This was the result of additional tests that were bundled or added on to the basic interpretation of the Pap test and included molecular tests such as evaluation for HPV. Unfortunately, many other unordered tests were added, such as molecular tests for type-specific fungi and sexually transmitted diseases (Chlamydia, Trichomonas, and gonococcus), that may not have been clinically indicated for the patient. These tests can be ordered inadvertently by checking a laboratory requisition box that allows for reflex testing, and health care providers may be unaware what tests are included in the mix. The shotgun laboratory testing approach to clinical diagnosis is well known to pathologists, who generally strongly discourage its use but who may not be involved directly in the marketing arm of laboratory services. (See Dr. Bettigole's commentary, page 60.)

The adoption of molecular platforms for primary cervical cancer screening, a screening strategy now under investigation as an alternative to the Pap test, may encourage additional, inappropriate molecular tests from the sample vial in what could become a marketing strategy for some laboratories and clinics and drive up the cost of

cervical cancer screening. Clinical practices in 31 states are permitted to "client bill" for laboratory services, which means the practice directly bills the patient for laboratory tests and then pays the reference laboratory only the customary or discounted cost, thereby allowing the practice to keep a percent of the profit.[8] The inappropriate use of some molecular tests might be deterred if patients receive a bill directly from the laboratory and question the use of those tests, as Dr. Bettigole's patients did.

Does this mean that molecular tests should not be considered in cervical cancer screening? The Roche Cobas 4800 became in April the first HPV test to gain Food and Drug Administration approval to replace the Pap test for primary screening for cervical cancer. There are many advantages to using HPV tests in cervical cancer, primarily improved sensitivity for the detection of high-grade lesions. FDA approval allows for a proposed algorithm for screening in which women age 25 and older are tested for HPV DNA and referred to colposcopy if the test is positive for HPV 16/18, and reflexed to a Pap test if HPV positive but HPV 16/18 negative. Although this may be a reasonable approach, there is no peer-reviewed, U.S.-population–based study that documents the performance of the Pap test as a reflex test. Most of the concerns about primary HPV screening center on its implementation in the United States, where cervical cancer screening is opportunistic. Successful outcomes for programs using HPV screening tests have been reported in European countries with national cervical cancer screening programs,[9–11] in which providers are monitored on their use of testing and patients are recalled and triaged appropriately. Of the United Kingdom's ARTISTIC randomized trial using HPV for primary cervical cancer screening, the authors wrote, "Compliance with surveillance and optimal management of HPV-positive/cytology-negative women after primary HPV screening is of key importance."[10]

Women in the U.S. may forego screening for economic reasons, especially if the cost of the test is unpredictable. Studies have shown that health care providers in the U.S. do not follow consensus practice guidelines when ordering HPV tests. In a 2006 cross-sectional study by Lee, Berkowitz, and Saraiya,[12] only 75 percent of 376 health care providers in U.S. office-based practices ordered HPV tests, and 28.5 percent ordered low-risk HPV tests that are specifically discouraged. Most of the providers (59.6 percent) used cotesting in women under age 30 in contradiction to practice guidelines. Seventy-one percent ordered an HPV test for a Pap test interpretation of atypical squamous cells, cannot exclude high-grade squamous intraepithelial lesion (ASC-H), and 50.7 percent ordered it for high-grade intraepithelial lesion. Both of these interpretations warrant colposcopic referral regardless of the HPV test result. Inappropriate use of HPV tests, and overuse of HPV tests (such as ordering both a Pap test and HPV test on a too-frequent, annual basis), have the potential to increase health care costs significantly and discourage women from returning for follow-up visits.

In an Italian study assessing HPV DNA primary screening for cervical cancer and its precursors, the authors determined the following: "HPV-based screening should not start before 30–35 years. There is evidence that below 30 years HPV-based screening leads to an increased over-diagnosis of CIN2 that would regress spontaneously, with consequent over-treatment. Some increase in over-diagnosis is plausible also between 30 and 34 years. Below such ages, cytological screening is the recommended test."[13] Cervical cancer screening and Pap test follow-up guidelines in the U.S. were revised recently because the risk of potential harm from overtreatment of low-grade squamous intraepithelial lesions (LSIL) in women 30 and younger had outweighed the risk of developing cervical cancer. If primary HPV screening directs younger women to colposcopy, will that increase patient harm due to over-interpretation of colposcopic lesions as a significant cervical abnormality, and is colposcopy sufficiently sensitive and specific to detect and select all high-grade cervical lesions?

The use of HPV tests for primary cervical cancer screening is promising and the technology used to perform these tests continues to improve, which should reduce overall costs and increase efficiency for large-scale screening. To replace the Pap test in primary cervical screening, a proposed test should be highly sensitive and specific and perform as well as or better than the Pap test. Most current-generation HPV tests detect viral DNA. Next-generation HPV tests that detect abnormal methylation patterns of human or HPV DNA, or viral DNA integration into human DNA (in most cases the inciting event in the cascade to carcinoma) through detection of specific products of the integrated genome such as mRNA or protein product sequences, may provide further specificity. This might reduce the potential for unintended harm to women with lower-grade HPV lesions that do not have a propensity to

progress to cervical carcinoma. Molecular tests are expensive, and it would be prudent to evaluate the cost-benefit ratio for patient care before implementing new practice guidelines promoting their use, especially if existing tests that are less expensive will meet clinical needs.

What can pathologists and laboratories do to encourage appropriate test utilization? Pathologists can be involved in laboratory marketing efforts to ensure that laboratory representatives do not misrepresent appropriate laboratory testing. When pathologists serve as consultants to health care providers, providing advice on appropriate screening and follow-up tests, the patient is the beneficiary. Laboratories can design requisitions that make it clear to health care providers what tests are available and what their indications are, and make it easy to order ancillary tests separately, such as reflex testing, after initial screening tests have been performed. Prices of laboratory tests can be printed clearly on requisitions, which can serve as a reminder of the costs for health care providers who are concerned about their patients' ability to pay. As patients become more involved in their health care decisions, pathologists, patients, and health care providers should work as a team toward improved outcomes and appropriate test utilization.

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