Innovation and a New Role for HPV Testing in Cervical Cancer Screening

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Cervical Cancer

- #14 (cancer death in women)
- 12,000 cases, 4,200 deaths, (50% unscreened)
- Goal: detection of preinvasive disease
- Early detection: 5-year survival rate >90%
- Persistent High-Risk HPV infection
  - Almost 100% of cervical cancers HR HPV+
  - HPV16 (55-60%), HPV18 (10-15%)
- Cause all common/most rare histologic types
  - Squamous cell carcinoma (80-90%)

HPV Genome

HPV Genome

More than 100 HPV types
- High-risk types: HR16, HR18, etc.
- Low-risk types: LR11, LR12, etc.

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13 HR HPV Types:
- 16/18/31/33/35/39
- 45/51/52/56/58/59/68

Cervical Cancer

The Three Stages of Cervical Cancer Progression

The steps can be conceptualized as infection with specific high-risk types of human papillomavirus (HPV), progression to a precancerous lesion, and invasion. HPV infections are usually transient and are often associated with mild histologic abnormalities. Persistent infection with high-risk types of HPV is uncommon and is required for progression.
Role of E6 and E7 in Cervical Cancer

E6: HR-HPV types E6 protein causes degradation of p53 and activates telomerase

E7: Interaction with pRB and transactivation of E2F dependent promoters

RXRα deletion and E6E7 oncogene expression are sufficient to induce cervical malignant lesions in vivo

Cancer Letters - 28 April 2012 (Vol. 317, Issue 2, Pages 226-236)

Here we introduce a mouse model that develops spontaneously malignant cervical lesions allowing the study of the cooperative effect between HPV16E6E7 expression and the lack of RORα in cervical cancer development.

What are ASCUS and LSIL?

- ASCUS and LSIL are acronyms for two mild abnormalities detected by Pap tests.
  - ASCUS stands for atypical squamous cells of undetermined significance.
  - LSIL for low-grade squamous intraepithelial lesion.
- A diagnosis of ASCUS means that the nature of the abnormality is uncertain or equivocal.
- A diagnosis of LSIL means that there is a more definite, but still mild, abnormality.

Summary of 2012 ACOG, ASCP, ACS and ASCCP Cervical Cancer Screening Guidelines

<table>
<thead>
<tr>
<th>Population Age</th>
<th>Screening Recommendation</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Less than 21 years</td>
<td>No cervical cancer screening of any kind</td>
<td>HPV testing should not be used for screening or ASC-US reflex in this age group</td>
</tr>
<tr>
<td>21 - 25 years</td>
<td>Cytology alone primary screening every 3 years (acceptable)</td>
<td>HPV testing is recommended in cases of ASC-US cytology</td>
</tr>
<tr>
<td>26 - 30 years</td>
<td>HPV and cytology “co-testing” every 3 years (preferred)</td>
<td>Routine HPV “co-testing” is not recommended in this age group</td>
</tr>
<tr>
<td>30 - 65 years</td>
<td>No screening following adequate history of negative prior screening</td>
<td>Screening by HPV testing alone is not recommended for most clinical settings</td>
</tr>
<tr>
<td>Over 65 years</td>
<td>No screening if no previous history of CIN 2</td>
<td>Women with history of ≥CIN 2 should continue screening for at least 20 years</td>
</tr>
<tr>
<td>After hysterectomy</td>
<td>No screening if no previous history of CIN 2</td>
<td>Continue screening (cytology) if there is history of ≥CIN 2 in the past 20 years or cervical cancer ever</td>
</tr>
<tr>
<td>HPV vaccinated</td>
<td>Follow age-specific recommendations (same as unvaccinated women)</td>
<td></td>
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</tbody>
</table>

Primary Screening Women ≥30

Interpretation

For women aged 30 years and older in routine clinical practice who are negative by co-testing (both HPV and cytology), 3-year screening intervals were safe because a single negative test for HPV was sufficient to reassure against cervical cancer over 5 years. Incorporating HPV testing with cytology also resulted in earlier identification of women at high risk of cervical cancer, especially adenocarcinoma. Testing for HPV without adjunctive cytology might be sufficiently sensitive for primary screening for cervical cancer.

14 HR HPV Types:
16, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68

<table>
<thead>
<tr>
<th>TABLE 1. Comparison of Aptima and Real-time PCR for detection of cervical intraepithelial lesions in cervical population by histological gradea</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diagnostic index</td>
</tr>
<tr>
<td>------------------</td>
</tr>
<tr>
<td>CIN 2 or ≥ 40</td>
</tr>
<tr>
<td>CIN 1 or &gt; 19</td>
</tr>
<tr>
<td>Positive predictive value</td>
</tr>
<tr>
<td>Negative predictive value</td>
</tr>
</tbody>
</table>

a n = 1,118.

b Specified for detection of CIN 2 > between Aptima and Real-time PCR significance at P = 0.05.
In conclusion, our data indicate that the Aptima test is as sensitive as HC2 but more specific for detecting CIN 2 and has the potential to serve as a reliable test for both primary cervical cancer screening and the triage of borderline cytological abnormalities.

A meta analysis consisting of 8 studies. 1,839 ASC-US and 1,887 LSIL cases
False-negative results found in HPV testing

No oversight for HPV tests
Bob Ortega, The Arizona Republic
12:36 a.m. EST January 14, 2013

Is Your HPV Test FDA Approved?

Two LBC preservatives SurePath and ThinPrep

Table 1: Testing times allowed for of samples collected for Hybrid Capture 2 human papillomavirus testing as indicated on national laboratory analytic.

<table>
<thead>
<tr>
<th>Laboratory</th>
<th>RTM</th>
<th>ThinPrep™</th>
<th>SurePath™</th>
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<tbody>
<tr>
<td>A</td>
<td>14 days</td>
<td>1 month</td>
<td>1 month</td>
</tr>
<tr>
<td>B</td>
<td>21 days</td>
<td>1 month</td>
<td>1 month</td>
</tr>
<tr>
<td>C</td>
<td>28 days</td>
<td>Not used</td>
<td>Not used</td>
</tr>
<tr>
<td>D</td>
<td>42 days</td>
<td>30 days</td>
<td>30 days</td>
</tr>
</tbody>
</table>

Additional: 39% cases transport medium.

Conclusion: Recently updated cervical screening guidelines in the US recommended against the use of Hybrid Capture 2 (HC2) testing in cervical screening, including widely used HC2 testing from the SurePath vial. The manufacturer recently issued a technical bulletin specifically warning that the use of SurePath samples with the HC2 HPV test may provide false negative results and potentially compromise patient safety. Co-collection using a Food and Drug Administration-approved Hybrid Capture 2 HPV test medium is recommended for HPV testing of patients undergoing cervical screening using SurePath vials.

Drug, healthcare, and patient safety

Limitations of widely used high-risk human papillomavirus laboratory-developed testing in cervical cancer screening

Objectives: To increase awareness of the limitations of high-risk human papillomavirus (hrHPV) laboratory-developed testing (LDT) widely used in US cervical cancer screening. Methods and results: Written summaries about the FDA’s development and approval of the hrHPV LDT were developed and shared with 24 hrHPV laboratories.

Table 1: Recovery of human papillomavirus nucleic acids from liquid-based cytology media.

Fig. 1: Recovery of nucleic acids by Qiagen extraction following storage in 50% glycerol solution at -80°C for 7 days. The recovery of HR HPV 16 DNA was significantly decreased (p < 0.05) relative to fresh media. For the Qiagen Lysis Buffer, the shape indicates RNA and the solid line indicates DNA.

Fig. 2: Data collected at the company’s headquarters, 2013-2014, Qiagen Inc. All data points were collected using the Qiagen Lysis Buffer. The company’s data shows that the Qiagen Lysis Buffer does not contain DNA, as DNA is not detected in the presence of the glycine buffer at pH 9.5.