Title

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Journal

Archives of Pathology & Laboratory Medicine, 140(1)

ISSN

0003-9985

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Publication Date

2016

DOI

10.5858/arpa.2015-0004-cp

Peer reviewed

Trends in Cervical Cytology Screening and Reporting Practices

Results From the College of American Pathologists 2011 PAP Education Supplemental Questionnaire

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• ***Context.***—**The College of American Pathologists periodically surveys laboratories to determine changes in cytopathology practices. We report the results of a 2011 gynecologic cytology survey.**

Objective.—**To provide a cross-sectional survey of gynecologic cytology practices in 2010.**

Design.—**In 2011, a survey was sent to 1604 laboratories participating in the College of American Pathologists gynecologic cytology interlaboratory comparison education program and proficiency testing programs requesting data from 2010 on the following topics: terminology/reporting, cytotechnologist workload, quality assurance, reagents, and ancillary testing.**

Results.—**Six hundred and twenty-five laboratories (39%) replied to the survey. The nonstandard use of "low-grade squamous intraepithelial lesion cannot exclude high-grade squamous intraepithelial lesion" is used by most laboratories to report the presence of low-grade squamous intraepithelial lesion with possibility of high-grade squamous intraepithelial lesion. Most laboratories also report the presence or absence of cells from the transformation zone. Most respondents do not limit cytotechnologist screening workload during the work shift. Only about one-third of laboratories (188 of 582; 32%) use image-assisted screening devices. Rapid prescreening as a quality assurance measure is used by only 3.5% (21 of 594) of the laboratories. When used for screening, most laboratories use the imager for retrospective review of slides to detect human locator and interpretive errors. Most laboratories receive both liquid-based cytology samples (mainly ThinPrep, Hologic, Marlborough, Massachusetts) and conventional Papanicolaou tests. Expiration dates of liquid-based cytology test vials are not usually recorded.**

Conclusions.—**The field of gynecologic cytology is evolving rapidly. These survey results offer a snapshot of national gynecologic cytology practices in 2010.**

(*Arch Pathol Lab Med*. 2016;140:13–21; doi: 10.5858/arpa.2015-0004-CP)

Cervical cytology has been the focal point for cytology innovation for the past 20 years, beginning with the adoption of the 1991 Bethesda System (TBS) for reporting cervicovaginal cytology. Since then, the practice of processing, screening, interpreting, and reporting Papanicolaou (Pap) tests has evolved to include adoption of liquid-based cervical sample collection with automated processing systems, implementation of automated screening devices, reflex testing of residual samples for human papillomavirus (HPV), and updated TBS reporting terminology to augment our improved understanding of HPV pathogenesis in cervical cancer and to complement clinical practice guidelines. There are few opportunities to assess and monitor national laboratory practices in cytology as they change over time, but the College of American Pathologists (CAP) interlaboratory comparison programs provide a unique opportunity to investigate changes in practices among participating laboratories by creating

Accepted for publication April 30, 2015.

Published as an Early Online Release June 5, 2015.

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The authors have no relevant financial interest in the products or companies described in this article.

The opinions or assertions contained herein are the private views of the authors and do not reflect the official policy of the Department of the Army, the Department of Defense, or the US government.

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Table 1. Responses to Reporting Practices in Gynecologic Cytology

Question	Response No. (%)
How do you report Papanicolaou tests with obvious LSIL and cells suspicious for HSIL?[a] (n = 576)	
LSIL, cannot exclude HSIL	466 (80.9)
LSIL and ASC-H	56 (9.7)
HSIL	55 (9.5)
LSIL	49 (8.5)
ASC-H	33 (5.7)
Does your laboratory report endocervical cells/transformation zone sampling? (n = 591)	
Report presence and absence	445 (75.3)
Report only absence	111 (18.8)
Do not report	28 (4.7)
Report only presence	7 (1.2)

Annual Tests With Absence of Endocervical Cells/Transformation Zone Sampling								
No.	Mean	Minimum	Maximum	10th Percentile	25th Percentile	50th Percentile	75th Percentile	90th Percentile
336	15.5	0	92	4	6	13	20	30

Abbreviations: ASC-H, atypical squamous cells, cannot exclude high-grade squamous intraepithelial lesion; HSIL, high-grade squamous intraepithelial lesion; LSIL, low-grade squamous intraepithelial lesion.

[a] Multiple responses were allowed for this question.

periodic surveys, referred to as *supplemental questionnaires*, for inclusion with the programs. The CAP also uses responses to those surveys to establish national benchmarks for diagnostic categories and other practice parameters. Completion of these surveys is voluntary. The CAP Cytopathology Committee administers regular gynecologic cytology glass-slide challenges to participating laboratories in the United States, Canada, and other countries through the Interlaboratory Comparison Program in Gynecologic Cytology and the Pap Proficiency Testing Program. The CAP can evaluate emerging trends in cervical cytopathology through analysis of the responses to the survey, including the adoption of new technologies, terminology, and reporting practices of participant laboratories. In 2011, we surveyed laboratories to determine current cervical cytology practices with an emphasis on workload, imaging systems, and the use of molecular tests in cervical cytology. Between 2012 and 2015, we have seen major shifts in screening, management, and prevention options and in the emergence of a US Food and Drug Administration (FDA)–approved HPV platform for primary screening for cervical cancer in the United States. The data collected from 2010 laboratory practices in gynecologic cytology provides a baseline from which to compare results from future practice surveys.

MATERIALS AND METHODS

Members of the Cytopathology Committee formulated a supplemental questionnaire, examining 2010 cytology practices, to accompany the February 2011 mailing of the Pap Interlaboratory Comparison Program in Gynecologic Cytology. The survey questions addressed perceived trends in gynecologic cytology based on newly available technologies, guidelines for cervical cancer screening, and recommendations for calculating Pap test screening workload for semiautomated devices. Questions were divided into the following general categories: demographic information, terminology and reporting, workload, quality assurance using image analysis, Pap test reagents, and testing for HPV or other disease biomarkers. All of the questions were reviewed by a biostatistician (R.J.S.) for statistical soundness. Eight of the 42 questions (19%) related to demographic information, and the remainder covered other topics. Participants were asked to submit data from the 2010 calendar year.

RESULTS

Overall, 1604 laboratories received the survey. Of these, 625 laboratories (39%) responded to the demographic portion of the survey, and 608 laboratories (38%) responded to the supplemental questions. Not every laboratory responded to every question. Multiple responses were allowed for many of the questions. Most respondents (40.9%; 254 of 621) were voluntary, nonprofit hospital laboratories; 16.6% (103 of 621) were regional or local independent laboratories; 8.5% (53 of 621) were proprietary hospitals; 8.4% (52 of 621) were university hospitals; 7.2% (45 of 621) were city, county, or state hospitals; 6.9% (43 of 621) were veteran's hospitals; 5.6% (35 of 621) were national or corporate laboratories; 3.1% (19 of 621) were clinical, group, or doctor's office laboratories; 2.4% (15 of 621) were military hospitals, and 0.3% (2 of 621) were public health or nonhospital laboratories. Laboratories examined an average of approximately 27 000 Pap tests per year with the middle 80% (470 of 621) of respondents reporting 330 to 52 200 tests. The minimum number of Pap tests reported by participants was 330, and the maximum number of Pap tests reported was 1 187 059.

Among 576 responses to the question, "How do you report Pap tests with obvious low-grade squamous intraepithelial lesion (LSIL) and cells suspicious for high-grade squamous intraepithelial lesion (HSIL)?" most respondents (80.9%; 466 of 576) employed the term *LSIL, cannot rule out HSIL (LSIL-H)* (Table 1). Multiple responses were allowed for this question. Less-common interpretations included *LSIL* in conjunction with *atypical squamous cells, cannot exclude high-grade squamous intraepithelial lesion (ASC-H)* (9.7%; 56 of 576), *HSIL* only (9.5%; 55 of 576), *LSIL* only (8.5%; 49 of 576), or *ASC-H* only (5.7%; 33 of 576). Most laboratories reported the presence and absence of endocervical cells/transformation zone (EC/TZ) sampling (75.3%; 445 of 591); 18.8% (111 of 591) reported only the absence of EC/TZ, whereas 1.2% (7 of 591) reported only their presence, and 4.7% (28 of 591) did not mention EC/TZ status. When laboratories were asked what percentage of their Pap tests in 2010 lacked an EC/TZ component, 336 responded that approximately 16% (mean [SD], 15.6% [13.8%]) of Pap tests did not contain transformation zone components with the 10th to 90th percentiles ranging from 4% to 30%. Most laboratories (60%; 189 of 315)

Table 2. Responses to Gynecologic Cytology Workload Practices

Question	Response, No. (%)
Does your laboratory limit gynecologic cytology screening to: (n = 573)	
We do not limit screening within the work shift	544 (94.9)
First part of the work shift	23 (4.0)
Second part of the work shift	6 (1.0)
Do you keep workload records that distinguish between image-assisted slides and nonimage-assisted slides? (n = 582)	
We do not use an image-assisted screening instrument	394 (67.7)
Yes	117 (20.1)
No	71 (12.2)
For image-assisted slides, do you count slides that require full manual review differently than those not requiring full manual review? (n = 121)	
Yes	108 (89.3)
No	13 (10.7)
For calculating workload, what value do you give image-assisted slides (not requiring full manual review)? (n = 140)	
0.5 (1/2 slide)	104 (74.3)
1.0	30 (21.4)
1.5	1 (0.7)
Other	5 (3.6)
When a full manual review is required for an image-assisted gynecologic cytology slide, the manual review is usually done by: (n = 141)	
The same cytotechnologist who performed the FOV review	132 (93.6)
A different cytotechnologist	4 (2.8)
A supervisory level cytotechnologist (>3 y experience)	4 (2.8)
A pathologist	1 (0.7)
If a full manual review is required for an image-assisted gynecologic cytology slide, when is it reviewed?[a] (n = 158)	
Immediately following the initial FOV with the imaging system microscope	122 (77.2)
Immediately following the FOV with a nonimaging system microscope	44 (27.8)
At a later time with a nonimaging system microscope	26 (16.5)
At a later time on the imaging system microscope	5 (3.2)

Gynecologic Cytology Slides Screened/Hour By Cytotechnologist

No.	Mean	Minimum	Maximum	10th Percentile	25th Percentile	50th Percentile	75th Percentile	90th Percentile
531	8.9	0	68	5	7	9	10	12

Abbreviation: FOV, field of view.

[a] Multiple responses were allowed for this question.

estimated that value rather than recording actual data (40%; 126 of 315).

Table 2 shows the data from questions related to cytotechnologist (CT) Pap test workload limits and how the workload is measured. Few laboratories (5.1%; 29 of 573) limited CT screening to a particular time during the work shift; most (94.9%; 544 of 573) did not. Of the 531 laboratories that reported the number of gynecologic slides screened per hour by CTs, the mean (SD) was 8.9 (3.5) slides (10th–90th percentile range, 5–12 slides, with a median of 9 slides). Most respondents did not use image-assisted screening instruments (67.7%; 394 of 582), but of those who did use image assistance, 62.2% (117 of 188) reported keeping workload records that distinguished between image-assisted slides and nonimage-assisted slides, and 37.8% (71 of 188) of those respondents did not distinguish between those 2 modalities. Of the laboratories that used image assistance, 89.3% (108 of 121) reported counting slides that require full manual review differently than those that do not require full manual review. Only 10.7% (13 of 121) of the respondents did not distinguish the workload limits when full manual review of image-assisted slides was performed. Of the respondents who performed image-assisted Pap tests, 74.3% (104 of 140) counted slides screened by image-assistance only (without full manual review) as 0.5 of a slide when calculating the total workload. Only 21.4% participants (30 of 140) counted image-assisted slides as 1.0 slides, and 4.3% participants (6 of 140) reported using some other value than 0.5 or 1 slide for the image-assisted slides not requiring further manual review. In 93.6% (132 of 141) of the laboratories, when a full manual review was required for an image-assisted Pap test, the CT who performed the original field-of-view (FOV) review also performed the full manual review. It was rarely reported that a different CT (2.8%; 4 of 141), a CT supervisor (2.8%; 4 of 141), or a pathologist (0.7%; 1 of 141) performed the manual review following the image-assisted FOV review. In most cases, as reported by 122 of 158 laboratories (77.2%), a full manual review was performed immediately after the initial FOV review, if required, using the imaging-system microscope. Less commonly, that review was performed immediately using a nonimaging system microscope (27.8%; 44 of 158), at a later time using a nonimaging system microscope (16.5%; 26 of 158), or rarely, at a later time using the

Table 3. Quality Assurance Practices Related to Imaging Systems

No.	Mean	Minimum	Maximum	10th Percentile	25th Percentile	50th Percentile	75th Percentile	90th Percentile
Image-assisted gynecologic cytology slides requiring full manual review, %								
129	33.8	0	100	10	15	25	40	100
ASC/SIL ratio for image-assisted gynecologic cytology slides								
133	2.43	0	44.0	1.0	1.2	1.6	2.0	3.0
ASC/SIL ratio for nonimage-assisted gynecologic cytology slides								
446	2.67	0	60.0	0.6	1.2	1.7	2.5	4.1

Question	Response, No. (%)
For quality assurance purposes, do you retrospectively review slides on the imaging system to detect the following types of errors?[a] (n = 120)	
Imager (cells not in FOV)	107 (89.2)
Human locator (in FOV, not marked)	102 (85.0)
Human interpretative (marked, not significant)	89 (74.2)
For the CLIA mandated 10% negative review, do you include imaged cases that are negative and[a] (n = 170)	
Did not need full manual review?	147 (93.6)
Required full manual review?	145 (92.4)
Do you look at retrospective cases with the imager to determine if there were abnormal cells in the FOV? (n = 170)	
Yes, if applicable	100 (58.8)
No	70 (41.2)

Abbreviations: ASC/SIL, Atypical squamous cells/squamous intraepithelial lesion; CLIA, Clinical Laboratory Improvement Amendments of 1988; FOV, field of view.

[a] Multiple responses were allowed for these questions.

imaging system microscope (3.2%; 5 of 158). Multiple responses were allowed for this question and some laboratories used more than one method.

Table 3 summarizes data about imaging systems as they pertained to quality assurance practices. The percentage of image-assisted Pap tests results requiring full manual review was reported by 129 laboratories. On average, 33.8% of slides required full manual review, with the middle 80% of respondents reporting review rates from 10% to 100%, with a median of 25%. The mean (SD) atypical squamous cells of undetermined significance/squamous intraepithelial lesion (ASC/SIL) ratio for image-assisted Pap tests, as reported by 133 laboratories, was 2.43 (2.81) (median, 1.6), as compared with a mean (SD) ASC/SIL ratio for nonimage-assisted Pap tests of 2.67 (3.74) (median, 1.7) as reported by 446 participants. Most respondents (74.1% [86 of 116] of the laboratories for image-assisted slides; 78.8% [312 of 396] of the laboratories for nonimage-assisted slides) used actual data for their answers; the remainder provided estimated values.

A series of questions on quality assurance activities for gynecologic cytology screening processes revealed that most respondents (96.5%; 573 of 594) did not perform rapid prescreening of Pap tests and did not use rapid rescreening of Pap tests (88.8%; 521 of 587) as quality measures. The remaining results are expressed in Table 3. We asked whether laboratories retrospectively reviewed slides on the imaging system, if available in the laboratory, to detect certain errors for quality assurance purposes. Multiple responses were allowed for this question. Most respondents (89.2%; 107 of 120) reported using the imaging system to retrospectively determine whether there were atypical cells present that were not displayed in the FOV review. Eighty-five percent (102 of 120) of the laboratories reported using the imaging system to determine whether a human locator error occurred, and 74.2% (89 of 120) used it to detect human interpretive error, whereby the cells were identified by the imaging system and marked by the CT but not deemed clinically significant. Most laboratories included imaged, negative Pap slides as part of their Clinical Laboratory Improvement Amendments of 1988 (CLIA)–mandated 10% negative review, both when slides did not require full manual review (93.6%; 147 of 157) and when they did (92.4%; 145 of 157). More than one-half of the respondents (58.8%; 100 of 170) used the imaging system to retrospectively review slides to determine whether there were abnormal cells in the field of view, while the rest (41.2%; 70 of 170) did not.

We asked several "yes" or "no" questions about reagents. Most laboratories (77.3%; 408 of 528) did not record the expiration dates of liquid-based cytology (LBC) Pap test vials that are sent to other clinics or facilities, but 55.4% (298 of 538) monitored the inventory of vials sent to these sites. Laboratories generally did not record the expiration dates of the LBC Pap test vials received (79.8%; 423 of 530). Once it was determined that a received LBC Pap test specimen was past the expiration date, 39.4% (184 of 467) rejected the specimen, 36.4% (170 of 467) performed a morphologic evaluation but included a disclaimer in the report, and 24.2% (113 of 467) performed a morphologic evaluation without a comment in the report.

Most laboratories (88.6%; 504 of 569) received requests for ancillary testing on LBC vials in addition to the Pap test. Although we asked a series of questions on ancillary testing for HPV, not all results are included. Table 4 shows responses to questions on ancillary testing. High-risk HPV testing was performed within the institution in 37.5% (215 of 573) of laboratories, with only 8.2% (47 of 573) of them performing the test in the cytology laboratory. Fifty-six

Table 4. Responses to Ancillary Testing	
Question	**Response, No. (%)**
Where is high-risk HPV testing performed? (n = 573)	
Sent out to a reference/referral laboratory	321 (56.0)
Within the institution, not in the cytology laboratory	168 (29.3)
In the cytology laboratory	47 (8.2)
Not performed	37 (6.5)
Other than HPV testing, what additional nonmorphologic tests does your laboratory perform from a liquid-based cytology vial?[a] (n = 511)	
No additional tests offered	295 (57.7)
Chlamydia trachomatis	215 (42.1)
Neisseria gonorrhea	202 (39.5)
Herpes simplex virus	39 (7.6)
Bacterial vaginosis	21 (4.1)
Cystic fibrosis	14 (2.7)
Hepatitis C virus	8 (1.6)
Human immunodeficiency virus	7 (1.4)
Epstein Barr virus	3 (0.6)
TERC (gain of 3q) analysis	1 (0.2)

Abbreviations: HPV, human papillomavirus; *TERC*, telomerase ribonucleic acid component.

[a] Multiple responses were allowed for this question.

percent (321 of 573) of laboratories sent out their high-risk HPV tests to a reference laboratory. Although most laboratories (57.7%; 295 of 511) did not offer additional tests other than HPV "off the vial," others reported offering the following nonmorphologic tests from residual specimen in the LBC vial (multiple responses were allowed): *Chlamydia trachomatis* (42.1%; 215 of 511), *Neisseria gonorrhoeae* (39.5%; 202 of 511), herpes simplex virus (7.6%; 39 of 511); bacterial vaginosis (4.1%; 21 of 511), and cystic fibrosis (2.7%; 14 of 511), among others.

COMMENT

In February 2011, the 1604 laboratories enrolled in the Pap Interlaboratory Comparison Program in Gynecologic Cytology program received the demographics and supplemental questionnaires. Six hundred and eight laboratories (38%) responded to most of the supplemental questions that were included in their first mailing of the Pap Interlaboratory Comparison Program in Gynecologic Cytology.

Terminology and Reporting

After the basic demographic questions, the first query on the survey asked laboratories how they reported Pap tests that showed obvious LSIL cells but also contained a few cells that might represent HSIL. Most respondents (80.9%; 466 of 576) used a term that was not part of the 2001 Bethesda System (TBS 2001) for reporting cervical cytology: *low-grade squamous intraepithelial lesion, cannot exclude high-grade squamous intraepithelial lesion (LSIL-H)*. The TBS 2001 divides SILs into *LSIL*, *HSIL*, and *SIL of indeterminate grade*, but does not have a specific category for Pap tests with LSILs that show a few cells suspicious for a higher-grade lesion (LSIL$^+$). Clinical management decisions are based on the risk of finding high-grade disease on follow-up biopsies; for LSILs on cytology, the cumulative risk of an underlying cervical intraepithelial neoplasm grade 2 or 3 is 11% to 16%,[1–2] whereas, for HSIL on Pap tests, the risk is greater than 50%.[3–6]

Although equivocal cytologic diagnoses should be minimized, the cytologic finding of a SIL of indeterminate grade does occur and needs to be addressed by laboratories to ensure proper clinical management. Most laboratories queried used an indeterminate interpretation category rather than choosing between a LSIL or HSIL interpretation for reporting rare, possible HSIL cells in a predominately LSIL Pap test. This finding supports the need for terminology that identifies this situation, so that women with LSIL who may have a higher-grade lesion receive colposcopies and possibly biopsies to exclude HSIL. Several studies[7–11] indicate that an LSIL-H interpretation has a higher likelihood of a cervical intraepithelial neoplasm 2 or 3 biopsy on follow-up than does LSIL alone.

A recent, large study by Zhou et al[12] concluded that LSIL-H had a distinct HPV genotype distribution that more closely mirrored the HPV genotype distribution of HSIL than it did LSIL. They found that the proportion of HPV-16 infections in women with LSIL-H (36%; 9 of 25) was comparable to their findings in HSIL (45%; 25 of 56) and was significantly different ($P = .007$) from LSIL (14%; 23 of 167) or ASC-H (0%; 0 of 9). Reporting the possible presence of HSIL in patients with LSIL may become increasingly important as more women with LSIL cytology are followed by colposcopic examination and repeat Pap testing without biopsies. As screening intervals increase, it is important to alert clinicians to the possibility of a high-grade lesion to ensure adequate surveillance. Although our survey showed that many laboratories use the terminology of LSIL-H in practice, there are strong arguments for maintaining a 2-tier reporting terminology that correlates with the biology of HPV infection and cervical carcinogenesis. Adding terminology, such as LSIL-H, might create a 3-tiered system that could negate the beneficial aspects of the 2-tiered TBS nomenclature. Furthermore, the 2012 American Society for Colposcopy and Cervical Pathology management guidelines[13] use *LSIL* and *HSIL* nomenclature without an intermediate category. Recent World Health Organization[14] terminology for reporting cervical lesions also adopts a 2-tier system that mirrors TBS-LSIL or HSIL.

Reporting the presence or absence of a TZ component (endocervical or metaplastic cells) is recommended in TBS[15] and may help clinicians determine whether the intended target was adequately sampled, especially in cases where few HSIL cells are identified. The clinical significance of the absence of the EC/TZ component in a cervical cytology specimen has been controversial, and management decisions for follow-up vary. In our study, most laboratories (75.3%; 445 of 591) reported on the presence *and* the absence of the EC/TZ component, but 18.8% (111 of 591) laboratories reported only on its absence.

CT Workload

Because of national interest in the influence of factors, such as fatigue, on the ability of CTs to perform accurate Pap test screening, we asked laboratories if they limited screening to certain times of the day. Responses included 94.9% (544 of 573) of the laboratories that did not limit screening during a work shift, but 23 respondents (4%; 23 of 573) reported limiting screening to the first part of the work shift, and 6 participants (1%; 6 of 573) reported limiting screening to the second part of the work shift. The impact of

CT workload productivity on the accuracy of screening is well recognized.[16–19]

In their study, Elsheikh et al[20] found significant differences in the detection rates of abnormal cases by CTs. Detection rates differed according to the time of the day and the day of the week for some CTs. Even though their study demonstrated that CT screening performance generally deteriorated during the second half of the day, laboratories may be unaware of the study or unable to change current shifts to provide CTs time away from screening. Those laboratories that do limit screening to the first or second part of the work shift may do so because of other variables. For instance, CTs may primarily assist with fine-needle aspiration procurement in the afternoons, leaving only the morning hours for Pap test screening. In our survey, most laboratories did not limit screening to a particular time during the work shift, but those that did limited screening to the first portion of the work shift. Further study is required to determine what processes laboratories use to divide workload or to restrict screening time.

Only one-third of the laboratories (32.3%, 188 of 582) surveyed used an image-assisted screening instrument, so these devices are not widely used. These instruments may be used primarily by high-volume laboratories to increase efficiency. On July 27, 2010, the FDA released an alert clarifying the workload recording for semiautomated gynecologic-cytology screening devices[21] and describing how laboratories can safely calculate workload for FDA-approved, semiautomated gynecologic-cytology screening devices. According to that alert, any slide that was reviewed on an image-assisted screening device using FOVs only was assigned a workload unit of 0.5. However, when a manual review was necessary, the slide was assigned an additional full unit of 1.0, thereby raising the workload unit of that slide to 1.5. Most laboratories (89%; 108 of 121) responding to our survey appeared to be aware of this alert and were counting the slides that required full manual review differently than those that did not. However, although three-fourths (74.3%; 104 of 140) counted image-assisted slide reviews that did *not* require full manual review as 0.5 unit, most of the other respondents (21.4%; 30 of 140) counted those image-assisted slides as 1.0 unit. Some laboratories may find it easier to count slides as one unit, regardless of the work required, when calculating CT workload. Respondents may also have misread the survey question because we did not ask a similar question about image-assisted slides that *do* require full manual review. The FDA approval for the ThinPrep Imaging System (Hologic, Inc, Marlborough, Massachusetts) allows for a maximum screening volume of 200 slides in 24 hours that are not to be screened in less than an 8-hour work day.[22]

The BD FocalPoint GS Imaging System (Becton, Dickinson and Company, Franklin Lakes, New Jersey), an automated, guided imaging system that screens conventional Pap smears and BD SurePath Pap tests, received FDA premarket approval for an individual CT to use the device to screen no more than 170 slides in 24 hours.[23] Only the FOV review can be examined for those slides to count as 0.5 units. Cytotechnologists are still restricted to 200 SurePath slides in 24 hours, screened in no less than an 8-hour work day, as required by CLIA for slides that have material covering less than one-half of the slide surface. Individual state regulations supersede CLIA regulations when they impose more-stringent guidelines limiting CT workload.

According to our data, CTs screened a mean (SD) of 8.9 (3.5) slides per hour, well within the CLIA requirements. It is not clear how laboratories determined that number, and our results were compiled from laboratories that count workloads in different ways. Of interest, the maximum number of gynecologic-cytology slides reportedly screened in 1 hour by one CT was 68! One can only hope that answer was an erroneous entry. Because of established guidelines and regulations, CT Pap test workload records should be kept to distinguish between image-assisted slides and nonimage-assisted slides. Approximately two-thirds of laboratories (62.2%; 117 of 188) that did use image-assisted screening instruments kept workload records distinguishing between image-assisted and nonimage-assisted slides. Most often, a full manual review was performed by the same CT who performed the FOV review (93.6%; 132 of 141), immediately following the initial review and using the same imaging system microscope (77.2%; 121 of 197). This suggests that CTs were assigned responsibility for particular slides and were expected to see the case to its completion. Another model would allow laboratories to have CTs performing only the FOV review on an imaging system and then passing the potentially abnormal slides to another CT for full manual review, but based on our data, this approach was not popular. Some CTs appeared to prefer to use a nonimaging microscopic for full manual review because 44.3% (70 of 158) responded that they reviewed those slides immediately or at a later time using a nonimaging system microscope. Multiple responses were permitted to this question, indicating that some CTs used different methods in the same laboratory. Laboratories that allow CTs to perform a full manual review on a separate, non-ThinPrep Imaging System microscope would not be in compliance with the FDA-approved method of review and would have to separately validate that process. In some laboratories, the imaging microscopes are separate from the CT's work space, and those CTs may prefer to use the microscope in their own work space for manual review. For the laboratories surveyed, a mean (SD) of 33.8% (28.4%) of the image-assisted Pap tests required a full manual review. That number included both abnormal Pap tests and mandatory 10% quality assurance review slides. The FocalPoint GS Imaging System received FDA premarket approval to allow 25% of imaged conventional or SurePath slide results to be released as normal without further human review, and of the remaining 75%, at least 15% must receive full manual review for quality control purposes.[21] There are no national or federal guidelines established for the number of ThinPrep Imaging System slides that must receive full manual review if they are otherwise interpreted as negative on initial FOV review. However, negative imaged slides would still be subject to the mandatory CLIA requirement for 10% prospective rescreening of negative slides.[24] Most laboratories included imaged slides that required full manual review (92.4%; 145 of 157) and that did not require full manual review (93.6%; 147 of 157) in their CLIA-mandated 10% negative review (Table 3).

Quality Assurance

Rapid prescreening has not taken hold in gynecologic cytology in the United States. Only 3.5% (21 of 594) of those surveyed used rapid prescreening, and slightly more respondents (11.2%; 66 of 587) rapidly rescreened Pap tests, but most laboratories did neither. Rapid prescreening[25] and rapid rescreening[26] have been proposed as cost-effective

processes that could replace the 10% random review of negative for intraepithelial lesion or malignancy cases and prove more effective at detecting missed HSIL. The 120 laboratories that used imaging systems for quality assurance purposes were inclined to use them for retrospective review of gynecologic cytology slides to detect imaging (89.2%; 107 of 120), human locator (85%; 102 of 120), and human interpretive (74.2%; 89 of 120) error. Imaging errors are the presence of atypical cells that are not presented in the FOVs. Human locator errors are expressed as cells that are presented in the FOV but are not recognized and marked as significant by the CT. Human interpretive error is when the atypical cells are present in the FOV, recognized, and marked but interpreted as not significant. Fifty-nine percent (58.8%; 100 of 170) of laboratories used the imaging system, when applicable, on cases selected for retrospective review to determine if there were abnormal cells in the FOV. This is a powerful and effective method of screening for instrument error and can help cytology professionals determine interfaces between humans and instruments that are most prone to error. For instance, there may be abnormal cells, such as koilocytes, that fall between the FOVs. Reviewing negative Pap tests in cases with a cervical intraepithelial neoplasm 1 biopsy result, using the imaging instrument to demonstrate selected FOVs, may reveal that the most diagnostically significant cells were not included in the FOVs. When professionals become aware of specific instrument limitations, it alerts them to potential errors and encourages additional diligence or processes to prevent those errors. Of 157 laboratories, most included negative imaged Pap tests in the CLIA-mandated 10% negative review both when they did not require full manual review (93.6%; 147 of 157) and when they did (92.4%; 145 of 157). This probably occurs because the selection of cases for 10% prospective review must include slides selected randomly from negative cases.[24]

The mean (SD) percentage of image-assisted gynecologic slides that had full manual review was 33.8% (28.4%) for 129 responding laboratories with imaging systems. The mean ASC/SIL ratio for image-assisted cases was 2.43, with 75% (100 of 133) of laboratories reporting ASC/SIL ratios under 2, whereas the mean (SD) for nonimage-assisted Pap test slides from 446 laboratory respondents was slightly higher (2.67 [3.74]), with 90% (401 of 446) of laboratories reporting ASC/SIL ratios below 4.1. The difference between these two means was not statistically significant ($P = .60$; t test). Whether slides were imaged or nonimaged did not have an effect on the mean ASC/SIL ratio for laboratories, but these results may be flawed because of the inclusion of both actual and estimated data and inclusion of data from different imaging systems that select abnormal cells through different algorithms. Renshaw et al[27] investigated the ASC/SIL ratio as a monitor of a CT's screening sensitivity when correlated with other statistics and reported that laboratories using location-guided screening were less likely to have CTs who have ASC/SIL ratios less than 1.5 (1 of 20; 5%) than those without imaging systems. The CT ASC/SIL ratios did not correlate with volume of slides, workload, or preparation type (conventional versus LBC). Our data suggest that the ASC/SIL ratio is affected by the use of imaging systems, but more-robust studies in this area would be necessary to confirm this hypothesis because of the variables discussed above.

Reagents

Four survey questions dealt with LBC reagent-collection vials for cervical cytology relating to inventory control and to the fixative expiration date printed on each vial as provided by the company. These questions were posed because items relating to reagent storage, labeling, expiration, and lot verification have recently been added to the CAP Accreditation Program checklists.[28]

We explored whether laboratories monitored the inventory of the LBC vials sent to clinics and other facilities sending Pap test samples to the laboratory. Inventory control, as it relates to the stability of reagents, is mandated by CLIA[29] and is addressed by multiple declarative statements (COM.30300, COM.30350, COM.30400, and COM.30450) in the CAP Accreditation Program All Common Checklist.[28] These statements address reagent labeling, storage, expiration date, and new reagent-lot verification, respectively. The intent of these statements is to ensure that laboratories have established procedures to ensure specimen integrity from the clinical services submitting specimens and to ensure the stability of reagents necessary to perform the tests within the laboratory. Most laboratories (55.4%; 298 of 538) responded that they did monitor the inventory of vials that they sent to clinics and facilities. This may be a service provided by the laboratory to ensure that clinics are appropriately stocked with necessary submission receptacles, and if laboratories included the cost of the vials in their test price, this may also be a prudent mechanism to prevent waste of resources. It would not be cost effective to allow clinics to stock dozens of unused vials that expire and are never submitted with a sample. Laboratories that did not provide LBC vials, but that required clinics to purchase their own vials, would have had no incentive for monitoring clinical inventory.

Three questions addressed the expiration date on the collection vials. ThinPrep vials have a shelf life of 2 years,[30] and the shelf life of BD SurePath vials is 3 years.[31] The CAP All Common Checklist,[28] statement COM.30400, requires confirmation that all reagents are used within their indicated expiration date. Most laboratories (77.3%; 408 of 528) did not record expiration dates of LBC Pap test vials that were *sent* to clinics, even though that would be a reasonable action to ensure that clinics did not receive expired reagent. It is not clear whether the remaining laboratories did not monitor expiration dates at all, did not send expired vials to clinics, did not *record* the dates, or simply did not send vials to clinics and, therefore, did not need to monitor reagent expiration dates on the vials. We did not provide a "not applicable" response for these questions. Remarkably, most laboratories (79.8%; 423 of 530) did not record the expiration dates of LBC vials that they *received* from clinical practices. Some of them did reject the specimen outright (39.4%; 184 of 467) if the vial had expired or provided a disclaimer about expiration in the final report (36.4%; 170 of 467), but one-quarter of laboratories (24.2%; 113 of 467) simply reported the results. This set of findings implies that laboratories were aware that a specimen was submitted in a vial that had expired but did not record the number of those vials or where they were received from, even though they may have rejected the specimen or commented on the specimen status in the report. It may be that CTs and pathologists noticed that the morphology of cells was not impaired in expired vials and, therefore, believed that it was a safe practice to report morphologic findings. Only 22.7%

(120 of 528) of the laboratories did record the expiration date of vials sent to clinical practices. Even fewer laboratories recorded the expiration date on the patient sample vials received from the clinics (20.2%; 107 of 530). This is significant because the shelf life of vials containing a patient sample is considerably shorter than the virgin vial. For example, the shelf life of a SurePath vial with a patient sample is 4 weeks at room temperature or 6 months at –17°C to –13°C (2°F to 8°F),[31] and it is 6 weeks from collection at room temperature for samples in ThinPrep medium.[30] Therefore, a specimen submitted in a vial with expired reagent would not be considered viable, and a patient sample in a medium with an expiration date after its vial expiration date would have a shorter expiration interval than expected. We did not ask about specimen expiration as it relates directly to HPV testing, but that is one area where adherence to the manufacturer's expiration dates may be critical. Some studies[32–35] show that cervical samples have viability for HPV testing beyond manufacturer's recommendations for some preservatives and HPV testing types, but testing of expired specimens under those circumstances should be approached with caution. Castle et al[34] showed that, after several years of storage in a methanol-based preservative at ambient temperature, the nuclear detail and β-globulin DNA of cervical cells deteriorated even though HPV DNA detection by Hybrid Capture 2 (Qiagen, Valencia, California) was not affected. Further studies would be helpful to provide clear guidance on specimen stability in other mediums. Laboratories seeking to extend specimens past recommended expiration dates should internally validate specimen viability before testing.

Ancillary Testing

Ancillary testing, primarily testing for high-risk HPV, has become common practice, with 88.6% (504 of 569) of laboratories receiving requests for ancillary tests on LBC Pap test vials. The most common additional out-of-the-vial tests offered were for *Chlamydia trachomatis* (42%; 215 of 511) and *Neisseria gonorrhea* (40%; 202 of 511).

Only a few laboratories in our study performed testing for herpes, bacterial vaginosis, cystic fibrosis, hepatitis C virus, human immunodeficiency virus, Epstein-Barr virus, or the evaluation of genomic amplification of the human telomerase RNA component (*TERC*) gene analysis on specimen remaining in LBC vials (Table 4). The *TERC* component (extra copies of chromosome arm 3q that cause additional telomerase genes) can be detected by fluorescence in-situ hybridization on LBC specimens and has been associated with invasive cervical carcinoma,[36] but that test was only offered by one laboratory. These findings indicate that, as a general rule, health care providers are not requesting studies other than HPV and other sexually transmitted diseases from residual Pap test specimens.

In conclusion, 2010 practice patterns in gynecologic cytology among participants of the CAP Pap Proficiency Testing Program and Pap Education Program showed that laboratories using TBS 2001 for reporting results reported the presence and absence of endocervical component on Pap tests and had adopted the term *LSIL-H*. Automated screening with imaging devices had not become common, but where they existed, most laboratories made use of the instruments for quality-improvement purposes. Rapid prescreening and rescreening had not been significantly adopted for quality assurance in cytology in the United States. Additionally, most laboratories did not limit screening to a particular time during a work shift. Usually, laboratories counted imaged Pap tests differently than manually screened Pap tests for workload capture, but confusion still existed on the proper means of reporting workload on imaged cases. In general, laboratories were not very vigilant about LBC vial expiration, for either prespecimen or postspecimen collection. In the past decade, there have been significant changes in practice in cervical cancer screening and prevention in the United States and internationally. These data will provide a useful baseline for future assessment of practice patterns of laboratory cervical-cancer screening.

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