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Comparison of the SurePath™ Liquid-Based Papanicolaou Smear with the Conventional Papanicolaou Smear in a Multisite Direct-to-Vial Study

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BACKGROUND. Split-sample clinical trials for liquid-based Papanicolaou (Pap) smears demonstrated that the liquid-based Pap smear was a safe and effective replacement for the conventional Pap smear. However, clinical intended use of liquid-based technology employs direct-to-vial collection methods. The current study compared the cytologic detection rates of the liquid-based Pap smear with conventional Pap smears in a direct-to-vial study performed at three clinical sites.

METHODS. Data from 58,580 prospective SurePath™ slides and 58,988 historic conventional slides were collected. Results were statistically compared with regard to disease prevalence and adequacy to include biopsy follow-up data for conventional and SurePath tests.

RESULTS. The SurePath method was found to provide a statistically significantly greater detection rate for clinically important categories of high-grade squamous intraepithelial lesion (HSIL+) and low-grade squamous intraepithelial lesion (LSIL+) (64% and 107%, respectively; $P < 0.00001$ for each lesion) compared with conventional slides. The clinical significance of increased cytologic detection using SurePath was supported by biopsy data that essentially demonstrated concordance with regard to biopsy interpretation for HSIL+ ($P = 0.9105$ at Site 1; $P = 1.0000$ at Site 2; and $P = 1.0000$ at Site 3) and LSIL+ ($P = 0.6966$ at Site 1; $P = 0.8052$ at Site 2; and $P = 1.00$ at Site 3). The detection rate of atypical squamous cells of undetermined significance (ASCUS+) was found to be significantly increased (75.12%; $P < 0.00001$). A statistically significantly lower proportion of unsatisfactory slides using the SurePath test compared with conventional slides was noted (-58%; $P < 0.00001$). The ASCUS/LSIL+ ratio was found to be reduced overall when using SurePath (-28.9%), regardless of whether the study sites were combined or considered individually. The rate of false-negative results noted with SurePath (10.43%) and conventional slides (12.97%) was essentially equivalent.

CONCLUSIONS. The SurePath Pap smear was found to outperform conventional slides in the detection of HSIL+ and LSIL+ cytologic lesions of the cervix and reduced the number of unsatisfactory diagnoses. The HSIL+ advantage for SurePath is not limited to HSIL but appears to extend to carcinoma as well. ***Cancer (Cancer Cytopathol)* 2004;102:269–79.** © *2004 American Cancer Society.*

KEYWORDS: SurePath™, conventional slide, Papanicolaou (Pap) smear, cytology, liquid-based, thin-layer, cervicovaginal smear.

Liquid-based preparation methods for cervical cytology have been shown repeatedly during the last decade to be highly effective in the detection of abnormalities. Currently, two such methods have received approval by the Food and Drug Administration (FDA) for use in the clinical laboratory. The most recent approval, in May 2000, was for the PrepStain™ system (TriPath Imaging®, Inc., Burlington, NC)

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upon the successful outcome of the company's split-sample clinical trial.[1]

After approval of the PrepStain system, which produces SurePath™ liquid-based slides, TriPath Imaging (the sponsor) began a direct-to-vial study at three clinical sites to substantiate the performance of the PrepStain system. In the study, 58,580 prospective SurePath slides prepared by the PrepStain slide processor were compared with 58,988 historic conventional slides. The SurePath slides were prepared according to the intended-use labeling for the PrepStain slide processor. Both SurePath and conventional slides were screened using routine laboratory practices.

The study analysis compared each slide type with regard to the detection of the abnormal categories of high-grade squamous intraepithelial lesion-positive (HSIL+), low-grade squamous intraepithelial lesion-positive (LSIL+), and atypical squamous cells of undetermined significance-positive (ASCUS+), and in the detection of the adequacy category of "unsatisfactory for evaluation. . .". The analyses also compared the false-negative fractions and computed the ASCUS-to-LSIL+ ratio for each slide type. Papanicolaou (Pap) smear and biopsy correlation data also were obtained from each site. The current study reports the performance of the PrepStain system in the direct-to-vial study compared with historic conventional Pap smears on large, unmatched populations at three clinical sites.

MATERIALS AND METHODS

The direct-to-vial study was conducted at three clinical laboratory sites that met the following criteria: 1) the clinical laboratory site collected specimens from clinics that had converted $>$ 98% to the SurePath collection method by the beginning of the clinical study; 2) the clinical laboratory site collected specimens from clinics that used primarily conventional collection methods prior to the conversion to SurePath; and 3) the clinical laboratory site electronically stored slide results in a database, and allowed retrieval by the sponsor, TriPath Imaging.

The Western Investigational Review Board approved the current study. The participating sites were: Site 1: Seacoast Pathology, P.A. in Exeter, New Hampshire; Site 2: AmeriPath Reference Pathology Services in Sandy, Utah; and Site 3: Chappell-Joyce Pathology Association in Texarkana, Texas. The personnel at these sites had previously received the SurePath morphology training offered by TriPath Imaging at time of initial installation and implementation of the PrepStain system and had experience with regard to cytologic review and evaluation of SurePath slides prior to initiation of the current study.

SurePath specimens were collected at each of the 3 sites, beginning on June 4, 2001, from a total of 57 healthcare providers. Qualified medical personnel used a broom-type sampling device (Rover's CervexBrush®; Andwin Scientific, Warner Center, CA) to collect the gynecologic specimen. The head of the sampling device was placed into the SurePath collection vial, which was capped, labeled, and sent with appropriate paperwork to the laboratory for processing. The head of the sampling device was never removed from the SurePath preservative vial during the entire PrepStain preparation process. In the laboratory, the preserved cellular sample was mixed by vortexing to homogenize the sample. The cell solution then was transferred automatically onto a PrepStain density reagent using the PrepMate® automated accessory (TriPath Imaging, Inc.). An enrichment step, comprised of centrifugal sedimentation through density reagent, partially removes nondiagnostic debris and excess inflammatory cells from the sample. After centrifugation, the pelleted cells were resuspended, mixed, and transferred to a PrepStain settling chamber mounted on a microscope slide. The slides were coated with a PrepStain slide coat to enhance cell adhesion. The cells were sedimented by gravity, then stained on the PrepStain slide processor using a modified Pap-staining procedure. The slide was cleared with xylene or a xylene substitute and coverslipped.

The PrepStain slide processor (as described earlier) converts the liquid suspension of a cervical cell sample into a discretely stained, homogeneous thin layer of cells while maintaining diagnostic cell clusters. The process includes cell preservation, randomization, enrichment of diagnostic material, pipetting, sedimentation, staining, and coverslipping to create a SurePath slide for use in routine cytology screening and categorization as defined by the Bethesda system. The SurePath slide presents a well preserved population of stained cells present within a circle measuring 13 mm in greatest dimension. Air-drying artifact and obscuring, overlapping cellular material and debris are largely eliminated. The numbers of leukocytes are significantly reduced, allowing for the easier visualization of epithelial cells, diagnostically relevant cells, and infectious organisms.

Conventional Pap smears were collected by qualified medical personnel primarily using the combination spatula/endocervical brush. Harvested cells were smeared onto the glass slide, fixed according to the protocol of each individual site, and sent to the laboratory for processing.

The control population of historic, conventional

slides was collected from the same clinics that provided the SurePath slides. The conventional slides were collected beginning with the most recent slides before the clinics converted to SurePath, and then going back in time until the conventional and SurePath slide populations were approximately equal in number. Data collection proceeded until enough SurePath and conventional HSIL+ slides were collected to support a valid statistical comparison of HSIL+ (HSIL, adenocarcinoma in situ, and carcinoma) between the 2 methods, namely until at least 245 conventional HSIL+ and at least 245 SurePath HSIL+ cases were diagnosed and counted. The sample size for the current study was derived to provide 80% power with which to test the primary hypothesis that SurePath slides were at least equivalent to conventional slides in the detection of HSIL+ within a region of indifference of 2% with a P value of 0.05. Although not used in the determination of sample size, a second hypothesis of superiority also was evaluated. With regard to the biopsy data, only the results of the Fisher exact test of superiority are provided because there was no region of indifference declared at the time of initiation of the study.

All diagnostic data were collected electronically from the databases of the three clinical laboratory sites according to routine site procedures. The final diagnoses and other required information for both slide populations were extracted from the databases of the respective sites and stored in the TriPath Imaging database for analysis. Neither TriPath Imaging nor the participating sites reprocessed the slides in any way for the current study.

This study was designed to evaluate the performance of the PrepStain system using direct-to-vial methods rather than split-sample methods, which may be less favorable to the second (liquid-based) sample. The primary objectives of the study were to demonstrate 1) that the detection rate of HSIL+ disease by trained cytology professionals when screening SurePath slides is equivalent to or better than that when conventional slides are screened and 2) that the false-negative fraction (FNF) of SurePath and conventional slide practices are equivalent.

The FNF is the estimation of the screening proficiency of a site, and is a measurement used to assess the overall quality or accuracy of a site's screening practice. The calculation of the FNF is based on a seminal industry article,[2] which states that a false-negative slide should be defined as a slide previously diagnosed as negative, but later found to be LSIL or higher. As shown in the article, the FNF is computed as:

TABLE 1
Age of the Subjects

Age (yrs)	CN No. (%)	SurePath No. (%)
≤19	2997 (5.08)	2875 (4.91)
20–29	10,934 (18.54)	11,688 (19.95)
30–39	15,499 (26.27)	14,719 (25.13)
40–49	14,015 (23.76)	13,799 (23.56)
≥50	15,498 (26.27)	15,492 (26.45)
Unknown	45 (0.08)	7 (0.01)
Total	58,988	58,580

CN: conventional.

$$\text{FNF}(\%) = \frac{\text{Estimated False-Negatives} \times 100}{\text{Initial Positives} + \text{Estimated False-Negatives}}$$ where:

$$\text{Estimated False-Negatives} = \left(\frac{\text{No. of LSIL + slides found during QC review}}{\text{Total No. of WNL slides QC reviewed}}\right) \times \text{Total No. of WNL slides}$$

$$\text{Initial Positives} = \text{No. of LSIL + slides found during intial screening}$$

with the "Initial Positives" indicating the number of LSIL+ slides detected during the initial screening, "WNL" indicating "within normal limits," and "QC" indicating "quality control."

Additional evaluation of the data included 1) a comparison of the demographic variables for each slide type population; 2) a comparison of SurePath and conventional slides in the detection of the Bethesda system categories of ASCUS+, LSIL+, and HSIL+; 3) a comparison of detection rates by clinical site; 4) a comparison of SurePath and conventional slides in the detection of the Bethesda system adequacy category of "Unsatisfactory for evaluation…"; 5) a comparison of the ASCUS to-LSIL+ ratio; and 6) a comparison of the correlation data between Pap smear and biopsy using SurePath and conventional slides.

RESULTS

Demographics

The demographics of conventional slide and SurePath populations were evaluated and compared with regard to subject age, clinical history (data not shown), and high-risk status. Table 1 shows that the subject age demographics of both slide preparation types were similar. The number and percentage of high-risk slides for each slide type were found to be similar

TABLE 2
High-Risk Slide Population

Total high-risk slides—all sites	
CN (n = 58,988 slides) No. (%)	**SurePath** (n = 58,580 slides) No. (%)
4208 (7.13)	4832 (8.25)

Percentage of high-risk slides	No. of clinics	
	CN	SurePath
0–5%	35	33
> 5–10%	15	15
> 10–15%	5	3
> 15–20%	0	2
> 20–25%	0	1
> 25%	2	3
Total	57	57

CN: Conventional.

TABLE 3
Numbers of SurePath and CN Slides by Diagnostic Category–All Sites Combined

Diagnosis	CN No. (%)	SurePath No. (%)	Total No. (%)
Unsat	315 (0.53)	130 (0.22)	445 (0.38)
WNL	56,611 (95.97)	54,864 (93.66)	111,475 (94.82)
ASCUS	414 (0.70)	480 (0.82)	894 (0.76)
AGUS	14 (0.02)	20 (0.03)	34 (0.03)
Atypia[a]	674 (1.14)	1111 (1.90)	1785 (1.52)
LSIL	712 (1.21)	1570 (2.68)	2282 (1.94)
HSIL	244 (0.41)	390 (0.67)	634 (0.54)
Carcinoma[b]	4 (0.01)	15 (0.03)	19 (0.02)
Total	58,988	58,580	117,568

CN: conventional; Unsat: unsatisfactory; WNL: within normal limits; ASCUS: atypical squamous cells of undetermined significance; AGUS: atypical glandular cells of undetermined significance; LSIL: low-grade squamous intraepithelial lesion; HSIL: high-grade squamous intraepithelial lesion.

[a] The site(s) did not specify the slide as "ASCUS" or "AGUS", but reported it as "atypia".

[b] Included two SurePath cases of adenocarcinoma not otherwise specified (NOS), two SurePath cases of carcinoma NOS, one SurePath case of endocervical adenocarcinoma, five SurePath cases of endometrial adenocarcinoma, one conventional case of endometrial adenocarcinoma, five SurePath cases of squamous cell carcinomas, and three conventional cases of squamous cell carcinoma.

among the 57 clinics in the study (7.13% for conventional slides and 8.25% for SurePath) (Table 2).

Performance

Table 3 provides the counts and relative distribution by diagnostic category of SurePath and conventional slides for all sites combined. Table 4 shows the same data presented according to site. The comparison data are given in Table 5. As can be noted, the SurePath direct-to-vial study results indicated that the SurePath method provides a statistically significantly greater detection rate for the clinically important categories of HSIL+ (64.4%) and LSIL+ (107.16%) and a statistically significantly lower proportion of unsatisfactory slides (–58.44%) regardless of whether the study sites were combined or examined individually. The HSIL+ advantage for SurePath is not limited to HSIL but extended to carcinoma as well ($P < 0.0092$ by the 1-sided Fisher exact test). For the ASCUS+ category, only SurePath results at Site 3 failed to demonstrate a statistically significant superiority.

The ASCUS-to-LSIL+ ratio, a measure of the specificity of cervical cytology screening, is shown in Table 6. The SurePath ratio demonstrated an average reduction across 3 sites of 28.9% from the conventional slide ratio.

The data used to calculate the overall FNF for each slide type (SurePath or conventional) are shown in Table 7. The following example illustrates the calculation of the overall SurePath FNF of 10.43% as shown in Table 8.

Estimated False-Negatives for SurePath

$$= \left(\frac{34}{7829}\right) \times 51{,}113 = 221.975$$

$$\text{FNF SurePath} = \frac{221.975 \times 100}{1906 + 221.975} = 10.43\%$$

All the FNFs for each slide type with associated 95% confidence intervals (95% CIs) are shown in Table 8. The FNF for the SurePath slides was nearly equal to that of conventional slides at Site 1, and the approximate 95% CI for the conventional slides completely overlapped the SurePath interval. At the other two sites and overall, the estimate of the FNF for SurePath slides was found to be lower than that for conventional slides, but the 95% CIs for conventional slides was reported to nearly completely overlap the 95% CIs for SurePath slides. This indicates that the FNF for the SurePath slides is no worse than that for conventional slides. The QC selection and screening processes were similar for the SurePath and conventional slides in each laboratory.

The rate of available biopsy follow-up for all diagnostic categories (ASCUS+, LSIL+, HSIL+, and carcinoma) was determined at each site for conventional slides and SurePath during the study period (see Tables 9–12). Pooled data from all 3 sites demonstrate the following biopsy follow-up rates for each category: carcinoma: 100% for conventional slides (4 of 4 slides) versus 67% for SurePath (10 of 15 slides); HSIL+: 82% for conventional slides (204 of 248 slides) versus 75% for SurePath (305 of 405 slides); LSIL+: 70% for conventional slides (675 of 960 slides) versus 62% for

TABLE 4
Numbers of SurePath and CN Slides by Diagnostic Category and Site

	Site 1[a]			Site 2[b]			Site 3[c]		
Diagnosis	CN No. (%)	SurePath No. (%)	Total	CN No. (%)	SurePath No. (%)	Total	CN No. (%)	SurePath No. (%)	Total
Unsat	132 (0.32)	37 (0.09)	169 (0.21)	163 (1.56)	89 (0.83)	252 (1.19)	20 (0.27)	4 (0.06)	24 (0.17)
WNL	39,703 (96.19)	38,086 (93.50)	77,789 (94.85)	9911 (95.11)	9898 (92.71)	19,809 (93.89)	6997 (95.94)	6880 (95.97)	13,877 (95.95)
ASCUS	0 (0.00)	0 (0.00)	0 (0.00)	246 (2.36)	325 (3.04)	571 (2.71)	168 (2.30)	155 (2.16)	323 (2.23)
AGUS	0 (0.00)	0 (0.00)	0 (0.00)	5 (0.05)	17 (0.16)	22 (0.10)	9 (0.12)	3 (0.04)	12 (0.08)
Atypia[d]	674 (1.63)	1111 (2.73)	1785 (2.18)	0 (0.0)	0 (0.00)	0 (0.0)	0 (0.00)	0 (0.00)	0 (0.00)
LSIL	549 (1.33)	1201 (2.95)	1750 (2.13)	77 (0.74)	269 (2.52)	346 (1.64)	86 (1.18)	100 (1.39)	186 (1.29)
HSIL	215 (0.52)	293 (0.72)	508 (0.62)	19 (0.18)	72 (0.67)	91 (0.43)	10 (0.14)	25 (0.35)	35 (0.24)
Cancer	1 (0.00)	7 (0.02)	8 (0.01)	0 (0.00)	6 (0.06)	6 (0.03)	3 (0.04)	2 (0.03)	5 (0.03)
Total	41,274	40,735	82,009	10,421	10,676	21,097	7293	7169	14,462

CN: conventional; Unsat: unsatisfactory; WNL: within normal limits; ASCUS: atypical squamous cells of undetermined significance; AGUS: atypical glandular cells of undetermined significance; LSIL: low-grade squamous intraepithelial lesion; HSIL: high-grade squamous intraepithelial lesion.

[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[c] Site 3: Chappell-Joyce Pathology Association, Texarkana, Texas.

[d] The site(s) did not specify the slide as "ASCUS" or "AGUS," but reported it as "atypia."

SurePath (1227 of 1975 slides); and ASCUS+: 49% for conventional slides (835 of 1715 slides) versus 48% for SurePath (1391 of 2897 slides).

The rate of available biopsy follow-up for HSIL+ between the conventional and SurePath arms did not appear to differ statistically (using the Fisher exact test of superiority) at Site 1 ($P = 0.0840$) or Site 3 ($P = 1.0000$). The available biopsy follow-up rate for HSIL+ at Site 2 was reported to be higher for SurePath compared with conventional slides ($P = 0.0376$).

The rate of available biopsies for LSIL+ slides between the conventional and SurePath arms did not appear to differ statistically at Site 1 ($P = 0.0631$) or Site 2 ($P = 0.8052$). The LSIL+ biopsy rate at Site 3 was found to be higher for conventional slides compared with SurePath ($P < 0.0001$).

The rate of biopsy for ASCUS+ slides between the conventional and SurePath arms did not appear to differ statistically at Site 1 ($P = 0.9198$). The ASCUS+ biopsy rate at Site 3 was found to be higher for conventional slides compared with SurePath ($P = 0.0018$). The ASCUS+ biopsy rate for Site 2 was not provided.

The correlation data between the Pap smear and biopsy results for both conventional and SurePath slides for each site for all diagnostic categories (ASCUS+, LSIL+, HSIL+, and carcinoma) are presented in Tables 12–16. The number of biopsies undertaken for carcinoma was too small to provide meaningful statistical comparisons. Exact correlation (biopsy and Pap smear, agreed on as defined by the laboratory) for HSIL+ at Site 1 was 76% for conventional slides (145 of 190 slides) versus 76% for SurePath (187 of 247 slides), whereas the correlation at Site 2 was 100% for conventional slides (4 of 4 slides) versus 85% for SurePath (33 of 39 slides), and the correlation at Site 3 was 90% for conventional slides (9 of 10 slides) versus 89% for SurePath (17 of 19 slides).

The percentage of slides for which the Pap smear and the biopsy results for HSIL+ were found to correlate among biopsy specimens was not found to be statistically significantly different at Site 1 ($P = 0.9105$), Site 2 ($P = 1.0000$), or Site 3 ($P = 1.0000$). The correlation between the Pap smear and the biopsy for LSIL+ demonstrated no statistically significant difference at Site 1 ($P = 0.6966$), Site 2 ($P = 0.8052$), or Site 3 ($P = 1.0000$). Similar correlation data for the ASCUS+ category demonstrated that the Pap smear result and the biopsy result were not statistically significantly different at Site 1 ($P = 0.9198$) or at Site 3 ($P = 1.0000$).

DISCUSSION

The current study was conducted under an Institutional Review Board-approved protocol and evaluated a total of 117,568 SurePath and conventional slides. The SurePath slides were screened according to routine laboratory practices and the intended-use labeling for the PrepStain system (PrepStain slide processor and SurePath liquid-based Pap smear). The SurePath population was comprised of 58,580 slides from 57 clinics that had converted nearly 100% from conventional Pap smear collection to the SurePath collection method.

The conventional Pap smear population was comprised of 58,988 slides taken from the same clinics as the SurePath slides. This historic population was col-

TABLE 5
Comparison of Detection Rates by Site

HSIL+

	CN			SurePath				
Site	Total	HSIL+	(%)	Total	HSIL+	(%)	% change	*P* value
1[a]	41,274	216	0.523	40,735	300	0.736	40.73	0.0001
2[b]	10,421	19	0.182	10,676	78	0.731	300.72	< 0.00001
3[c]	7293	13	0.178	7169	27	0.377	111.28	0.0167
Total	58988	248	0.420	58,580	405	0.691	64.44	< 0.00001

LSIL+

	CN			SurePath				
Site	Total	LSIL+	(%)	Total	LSIL+	(%)	% Change	*P* value
1	41,274	765	1.853	40,735	1501	3.685	98.81	< 0.00001
2	10,421	96	0.921	10,676	347	3.250	252.82	< 0.00001
3	7293	99	1.357	7169	127	1.772	30.50	0.0261
Total	58,988	960	1.627	58,580	1975	3.371	107.16	< 0.00001

ASCUS+

	CN			SurePath				
Site	Total	ASCUS+	(%)	Total	ASCUS+	(%)	% Change	*P* value
1	41,274	1,439	3.486	40,735	2,612	6.412	83.92	< 0.00001
2	10,421	347	3.330	10,676	689	6.454	93.82	< 0.00001
3	7,293	276	3.784	7,169	285	3.975	5.05	0.2906
Total	58,988	2,062	3.496	58,580	3,586	6.122	75.12	< 0.00001

Unsatisfactory

	CN			SurePath				
Site	Total	Unsat	(%)	Total	Unsat	(%)	% Change	*P* value
1	41,274	132	0.320	40,735	37	0.091	−71.60	< 0.00001
2	10,421	163	1.564	10,676	89	0.834	−46.70	< 0.00001
3	7293	20	0.274	7169	4	0.056	−79.65	0.0009
Total	58,988	315	0.534	58,580	130	0.222	−58.44	< 0.00001

HSIL: high-grade squamous intraepithelial lesion; +: positive; CN: conventional; LSIL: low-grade squamous intraepithelial lesion; ASCUS: atypical squamous cells of undetermined significance.

[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[c] Site 3: Chappell-Joyce Pathology Association, Texarkana, Texas.

Statistical tests of noninferiority using the method of Farrington and Manning[3] and superiority using the Fisher exact test were computed for the detection of slides containing carcinoma, those that were positive for low-grade squamous intraepithelial lesions, and those that were positive for atypical squamous cells of undertimed significance, as well as unsatisfactory slides for each slide type. These results were computed for all sites and for each slide individually. Although the study was not powered to detect a noninferiority test or a superiority test for carcinoma alone, the data in Table 5 were provided as supporting evidence that the detection capabilities of the SurePath system extend beyond HSIL. The results of the noninferiority test (data not shown) were $P < 0000.1$ for all diagnostic classes and for the combined data and individual sites alone. The *P* values of the superiority test and "% change," calculated as follows, are given in Table 5:

$$\text{Percent Change}(\%) = \left(\frac{\text{SP HSIL}^+/\text{SP Total} - \text{CN HSIL}^+/\text{CN Total}}{\text{CN HSIL}^+/\text{CN Total}} \right) \times 100$$

in which SP=SurePath and CN=Conventional.

TABLE 6
ASCUS: LSIL+ Ratios

	Site	CN ratio	SurePath ratio	Percent change
ASCUS: LSIL+	1[a]	0.881	0.742	−15.8
	2[b]	2.620	0.985	−62.4
	3[c]	1.787	1.243	−30.4
	All	1.147	0.816	−28.9

ASCUS: atypical squamous cells of undetermined significance; LSIL: low-grade squamous intraepithelial lesion; +: positive; CN: conventional.

[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[c] Site 3: Chappell-Joyce Pathology Association, Texarkana, Texas.

TABLE 7
Data Used to Calculate the FNFs of CN and SurePath

No. of LSIL+ slides detected at initial or QC review		
	CN	SurePath
Initial screen	927	1906
QC screen	20	34
No. of slides QC reviewed		
	CN	SurePath
WNL total	51,696	51,113
WNLs QC reviewed	7485	7829

FNF: false-negative fraction; CN: conventional; LSIL: low-grade squamous intraepithelial lesion; +: positive; QC: quality control; WNL: within normal limits.

lected beginning with the most recent slides before the clinics converted to SurePath, and then going back in time until the laboratories' conventional and SurePath slide populations were approximately equal in number.

Because the two-slide type populations were collected during different time periods, demographic data were collected to assess their similarities or differences. A comparison of the patient populations with regard to age, high-risk status, and prior history demonstrated that the two populations were reasonably alike to allow comparison of other population parameters. The study design did not control for repeat patients or repeat positive or negative slides. However, this issue applies to both patient groups (conventional and SurePath) and should not affect the results of the current study.

The overall detection rate for HSIL+, LSIL+, and ASCUS+ increased by 64%, 107%, and 75%, respectively. The individual site results demonstrated a statistically significantly increased rate of detection of

TABLE 8
Summary of FNF and Associated 95% Approximate Confidence Intervals[a]

Site	Detection method	FNF (%)	Lower 95% CI	Upper 95% CI
1[b]	SurePath	14.38	10.58	18.17
	CN	14.29	8.93	19.64
2[c]	SurePath	4.86	0.00	14.31
	CN	8.52	0.00	25.57
3[d]	SurePath	2.68	0.00	19.07
	CN	16.78	2.39	31.18
Overall	SurePath	10.43	6.87	13.99
	CN	12.97	8.08	17.86

FNF: false-negative fraction; 95% CI: 95% confidence interval; CN: conventional.

[a] The false-negative fraction (FNF) of each slide type was compared using 95% confidence intervals. An estimate of the variance of the FNF was required to estimate the 95% confidence intervals. The variance can be estimated using the approximate formula below[4]: $\mathrm{Var}[\,f(x,y)] \approx \left(\frac{\partial f}{\partial x}\right)^2 \mathrm{Var}(y) + \left(\frac{\partial f}{\partial y}\right)^2 \mathrm{Var}(x) - 2\left(\frac{\partial f}{\partial x}\right)\left(\frac{\partial f}{\partial y}\right)\mathrm{Cov}(x,y)$,

[b] Site 1: SeaCoast Pathology, Exeter, New Hampshire. [c] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[d] Chappell-Joyce Pathology Association, Texarkana, Texas.

HSIL+ (40.7%, 300.7%, and 111.3%, respectively) and LSIL+ (98.8%, 252.8%, and 30.5%, respectively) and a statistically significant reduction in the number of unsatisfactory slides across all sites compared with conventional slides (-71.6%, -46.7%, and –79.65%, respectively). Because the number and rate of high-risk slides were nearly the same in each study arm, these results cannot be attributed to an enriched population for the SurePath arm of the trial.

Some studies indicate a rate of disease progression to carcinoma from HSIL (carcinoma in situ) that ranges from 50–90%.[5] The ASCUS–LSIL Triage Study (ALTS) study indicated a possible progression rate from high-grade lesions to carcinoma of as high as 60%.[6] Using this figure, the finding of an additional 146 HSIL slides (390–244; from Table 3) implies that up to 87 women in this population of 58,000 might have had their disease progress to carcinoma.

The number of additional LSIL slides found in the SurePath arm was 858 (Table 3). The literature indicates that LSIL will harbor HSIL up to 28% of the time,[7] which translates into 240 women in the SurePath population of the current study.

The clinical significance of increased disease detection using SurePath was supported by biopsy data, which were collected at the request of the FDA. Biopsy data were obtained regarding 82.3% of HSIL+ cases in the SurePath population and 88.0% of HSIL+ cases in the conventional Pap smear population at Site 1, 50.0% of HSIL+ cases in the SurePath population and 21.1% of HSIL+ cases in the conventional Pap smear population at Site 2, and 70.4% of HSIL+ cases in the

TABLE 9
Biopsy Counts for Carcinoma Slides

	Site 1[a]		Site 2[b]		Site 3[c]	
Biopsy performed	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Biopsy	1 (100.00)	5 (71.43)	0 (0.00)	4 (66.67)	3 (100.00)	1 (50.00)
No biopsy	0 (0.00)	2 (28.57)	0 (0.00)	2 (33.33)	0 (0.00)	1 (50.00)
Total	1	7	0	6	3	2

CN: conventional. The numbers of biopsies for carcinoma were too small to provide meaningful statistical comparisons.
[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.
[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.
[c] Site 3: Chappell-Joyce Pathology Association, Texarkana, Texas.

TABLE 10
Biopsy Counts for HSIL+ Slides

	Site 1[a]		Site 2[b]		Site 3[c]	
Biopsy performed	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Biopsy (%)	190 (87.96)	247 (82.33)	4 (21.05)	39 (50.00)	10 (76.92)	19 (70.37)
P value	($P = 0.0840$)		($P = 0.0376$)		($P = 1.0000$)	
No biopsy (%)	26 (12.04)	53 (17.67)	15 (78.95)	39 (50.00)	3 (23.08)	8 (29.63)
Total	216	300	19	78	13	27

HSIL: high-grade squamous intraepithelial lesion; +: positive; CN: conventional.
[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.
[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.
[c] Site 3: Chappell-Joyce Pathology Association, Texarkana, Texas.

TABLE 11
Biopsy Counts for LSIL+ Slides

	Site 1[a]		Site 2[b]		Site 3[c]	
Biopsy performed	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Biopsy	562 (73.46)	1046 (69.69)	29 (30.21)	110 (31.70)	84 (84.85)	71 (55.91)
No biopsy	203 (26.54)	455 (30.31)	67 (69.79)	237 (68.30)	15 (15.15)	56 (44.09)
Total	765	1,501	96	347	99	127

LSIL: low-grade squamous intraepithelial lesion; +: positive; CN: conventional.
[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.
[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.
[c] Site 3: Chappell—Joyce Pathology Association, Texarkana, Texas.
The rate of biopsy for low-grade squamous intraepithelial lesion-positive (LSIL+) slides between the conventional and SurePath arms was not found to differ statistically at Site 1 ($P = 0.0631$) or Site 2 ($P = 0.8052$). The LSIL+ biopsy rate at Site 3 was found to be higher for the conventional compared with the SurePath arm ($P < 0.0001$).

SurePath population and 76.9% of HSIL+ cases in the conventional Pap smear population at Site 3. Because analysis of biopsy data was not required by the clinical protocol, the classification of cases was defined according to the laboratory practices of each of the clinical sites.

Although the biopsy data could not be collected with regard to all patients, the yield was sufficiently high (82.33% at Site 1, 50% at Site 2, and 70.37% at Site 3) (Table 10) to allow one to conclude that the increased detection of HSIL+ in the SurePath arm of the current study was clinically significant and did not

TABLE 12
Biopsy Counts for ASCUS+ Slides

	Site 1[a]		Site 2[b]		Site 3[c]	
Biopsy performed	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Biopsy	688 (47.81)	1277 (48.89)	NA	NA	147 (53.26)	114 (40.00)
No biopsy	751 (52.19)	1335 (51.11)	NA	NA	129 (46.74)	171 (60.00)
Total	1439	2612	NA	NA	276	285

ASCUS: atypical squamous cells of undetermined significance; +: positive; CN: conventional; NA: not applicable.

[a] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[b] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[c] Site 3: Chappell—Joyce Pathology Association, Texarkana, Texas.

The rate of biopsy for slides found to be positive for atypical squamous cells of undetermined significance (ASCUS+) slides between the conventional and SurePath arms was not found to differ statistically at Site 1 ($P = 0.9198$). The ASCUS+ biopsy rate at Site 3 was found to be higher for the conventional compared with the SurePath arm ($P < 0.0018$), and the ASCUS+ biopsy rate for Site 2 was not provided.

TABLE 13
Results for Carcinoma Biopsies

	Site 1[b]		Site 2[c]		Site 3[d]	
Biopsy result[a]	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Pap smear and biopsy correlate	0 (0.00)	4 (80.00)	0 (0.00)	4 (100.00)	2 (66.67)	1 (100.00)
Pap smear overcalled	1 (100.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Pap smear not representative	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
No correlation, Pap interpretation verified	0 (0.00)	1 (20.00)	0 (0.00)	0 (0.00)	1 (33.33)	0 (0.00)
Pap smear was undercalled	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Total	1	5	0	4	3	1

CN: conventional; Pap: Papanicolaou.

The numbers of biopsies performed for carcinoma were too small to provide meaningful statistical comparisons.

[a] The biopsy results are explained as follows: Papanicolaou (Pap) smear and biopsy correlate: the biopsy and the Pap smear were in agreement as defined by the laboratory. Pap smear overcalled: If the biopsy and the Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was overcalled. Pap smear not representative: the biopsy was diagnosed significantly higher than the Pap smear (as defined by the laboratory) and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The biopsy and the Pap smear may have been taken from different anatomic regions. No correlation, Pap interpretation verified: The Pap smear was diagnosed significantly higher than the biopsy (as defined by the laboratory), and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The Biopsy and the Pap smear may have been taken from different anatomic regions or the lesion regressed. Pap smear was undercalled: if the biopsy and Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was undercalled.

[b] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[c] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[d] Site 3: Chappell-Joyce Pathology Association, Texarkana, Texas.

result from overcalling of less significant abnormalities. The percentage of cases in which the cytologic diagnosis was found to correlate exactly with the biopsy diagnosis was nearly identical for the SurePath and conventional Pap smear arms at all the sites (75.71%, 84.6%, and 89.47%, respectively, for SurePath and 76.32%, 100% [all 4 cases], and 90%, respectively, for conventional smears). The percentage of overcalling at each of the sites was 8.50% (21 cases), 0%, and 5.26% (1 case), respectively, for SurePath slides and 11.05% (21 cases), 0%, and 0%, respectively, for conventional Pap smears. The percentage of cases designated as "no correlation, Pap diagnosis verified," cases in which the cytologic diagnosis was diagnosed at a significantly higher rate than biopsy, and rereview of the Pap smear and biopsy confirmed both the initial cytologic and biopsy diagnoses (most likely a function of sampling from different anatomic regions or disease regression), did not appear to differ significantly between the 3 sites for the SurePath and conventional Pap smears (15.38%, 15.38%, and 0%, respectively, for SurePath and 12.63%, 0%, and 10.00%, respectively, for conventional slides).

The ratio of ASCUS to LSIL+ was found to be reduced overall by 28.9% for SurePath compared with conventional slides. This reduction also was observed at each individual site (-15.8%, -62.4%, and -30.4%, respectively).

TABLE 14
Results for HSIL+ Biopsies

	Site 1[a]		Site 2[b]		Site 3[c]	
Biopsy result[a]	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Pap smear and biopsy correlate	145 (76.32)	187 (75.71)	4 (100.0)	33 (84.62)	9 (90.00)	17 (89.47)
Pap smear overcalled	21 (11.05)	21 (8.50)	0 (0.00)	0 (0.00)	0 (0.00)	1 (5.26)
Pap smear not representative	0 (0.00)	1 (0.40)	0 (0.00)	0 (0.00)	0 (0.00)	1 (5.26)
No correlation, Pap interpretation verified	24 (12.63)	38 (15.38)	0 (0.00)	6 (15.38)	1 (10.00)	0 (0.00)
Pap smear was undercalled	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Total	190	247	4	39	10	19

HSIL: high-grade squamous intraepithelial lesion; +: positive; CN: conventional; Pap: Papanicolaou.

The percentage of slides for which the Papanicolaou smear and biopsy results were correlated among the biopsied specimens was not found to be statistically significantly different at Site 1 ($P = 0.9105$), Site 2 ($P = 1.0000$), or Site 3 ($P = 1.0000$).

[a] The biopsy results are explained as follows: Papanicolaou (Pap) smear and biopsy correlate: the biopsy and the Pap smear were in agreement as defined by the laboratory. Pap smear overcalled: If the biopsy and the Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was overcalled. Pap smear not representative: the biopsy was diagnosed significantly higher than the Pap smear (as defined by the laboratory) and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The biopsy and the Pap smear may have been taken from different anatomic regions. No correlation, Pap interpretation verified: The Pap smear was diagnosed significantly higher than the biopsy (as defined by the laboratory), and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The Biopsy and the Pap smear may have been taken from different anatomic regions or the lesion regressed. Pap smear was undercalled: if the biopsy and Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was undercalled.

[b] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[c] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[d] Site 3: Chappell—Joyce Pathology Association, Texarkana, Texas.

TABLE 15
Results for LSIL+ Biopsies

	Site 1[b]		Site 2[c]		Site 3[d]	
Biopsy result[a]	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Pap smear and Biopsy correlate	375 (66.73)	708 (67.69)	17 (58.62)	75 (68.18)	74 (88.10)	63 (88.73)
Pap smear overcalled	33 (5.87)	35 (3.35)	1 (3.45)	0 (0.00)	7 (8.33)	3 (4.23)
Pap smear not representative	19 (3.38)	24 (2.29)	0 (0.00)	0 (0.00)	1 (1.19)	4 (5.63)
No correlation, Pap interpretation verified	120 (21.35)	276 (26.39)	11 (37.93)	35 (31.82)	2 (2.38)	1 (1.41)
Pap smear was undercalled	15 (2.67)	3 (0.29)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Total	562	1,046	29	110	84	71

The percentage of slides for which the Pap smear and biopsy results correlate among the biopsied specimens was not different statistically at Site 1 (p = 0.6966), Site 2 (p = 0.8052) or Site 3 (p = 1.0000).

LSIL: low-grade squamous intraepithelial lesion; +: positive; CN: conventional.

The percentage of slides for which the Papanicolaou smear and the biopsy results were correlated among the biopsied specimens was not found to be statistically significantly different at Site 1 ($P = 0.6966$), Site 2 ($P = 0.8052$), or Site 3 ($P = 1.0000$).

[a] The biopsy results are explained as follows: Papanicolaou (Pap) smear and biopsy correlate: the biopsy and the Pap smear were in agreement as defined by the laboratory. Pap smear overcalled: If the biopsy and the Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was overcalled. Pap smear not representative: the biopsy was diagnosed significantly higher than the Pap smear (as defined by the laboratory) and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The biopsy and the Pap smear may have been taken from different anatomic regions. No correlation, Pap interpretation verified: The Pap smear was diagnosed significantly higher than the biopsy (as defined by the laboratory), and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The Biopsy and the Pap smear may have been taken from different anatomic regions or the lesion regressed. Pap smear was undercalled: if the biopsy and Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was undercalled.

[b] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[c] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[d] Site 3: Chappell—Joyce Pathology Association, Texarkana, Texas.

In addition, the overall FNF was numerically reduced for SurePath (10.43%) compared with the conventional method (12.97%) The overall 95% CIs were found to overlap significantly, indicating that the two FNFs were very similar. For each site, a similar trend was observed. In all cases, the 95% CIs overlapped.

The results of the current direct-to-vial study corroborate the results obtained by other investiga-

TABLE 16
Results for ASCUS+ Biopsies

	Site 1[b]		Site 2[c,d]		Site 3[e]	
Biopsy result[a]	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)	CN No. (%)	SurePath No. (%)
Pap smear and biopsy correlate	464 (67.44)	857 (67.11)	NA	NA	137 (93.20)	106 (92.98)
Pap smear overcalled	40 (5.81)	47 (3.68)	NA	NA	7 (4.76)	3 (2.63)
Pap smear not representative	29 (4.22)	46 (3.60)	NA	NA	1 (0.68)	4 (3.51)
No correlation, Pap interpretation verified	133 (19.33)	320 (25.06)	NA	NA	2 (1.36)	1 (0.88)
Pap smear was undercalled	22 (3.20)	7 (0.55)	NA	NA	0 (0.00)	0 (0.00)
Total	688	1,277	NA	NA	147	114

ASCUS: atypical squamous cells of undetermined significance; +: positive; CN: conventional; NA: not applicable.

The correlation between the Papanicolaou results and the biopsy result was not found to be statistically significantly different at Site 1 ($P = 0.9198$) or Site 3 ($P = 1.0000$).

[a] The biopsy results are explained as follows: Papanicolaou (Pap) smear and biopsy correlate: the biopsy and the Pap smear were in agreement as defined by the laboratory. Pap smear overcalled: If the biopsy and the Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was overcalled. Pap smear not representative: the biopsy was diagnosed significantly higher than the Pap smear (as defined by the laboratory) and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The biopsy and the Pap smear may have been taken from different anatomic regions. No correlation, Pap interpretation verified: The Pap smear was diagnosed significantly higher than the biopsy (as defined by the laboratory), and the rereviews of the Pap smear and the biopsy showed that they were diagnosed correctly. The Biopsy and the Pap smear may have been taken from different anatomic regions or the lesion regressed. Pap smear was undercalled: if the biopsy and Pap smear did not correlate, the rereview of the Pap smear showed that the original diagnosis was undercalled.

[b] Site 1: SeaCoast Pathology, Exeter, New Hampshire.

[c] Site 2: AmeriPath-Utah Reference Pathology Services, Sandy, Utah.

[d] The correlation between the Papanicolaou (Pap) smear and the biopsy was calculated for all cases at Site 2 except those with a Pap interpretation of atypical squamous cells (ASC) (atypical squamous cells of undetermined significance [ASCUS]) and atypical glandular cells (AGC) (atypical glandular cells of undetermined significance [AGUS]). To monitor ASC/AGC, Site 2 used the frequency of disease at the time of biopsy to calculate the ASC/AGC yield, which is the total number of ASC/AGC cases with tissue biopsies showing disease (cervical intraepithelial neoplasia or worse) divided by the total number of ASC/AGC cases that have biopsies. The ASC yield was calculated at Site 2 as 36% (5 of 14 cases) and 66% (21 of 32 cases) for conventional and SurePath slides, respectively. The AGC yield was calculated as 0% (0 of 1 case) and 23% (1 of 4 cases) for conventional and SurePath slides, respectively.

[e] Site 3: Chappell—Joyce Pathology Association, Texarkana, Texas.

tors.[8–10] The current study was designed to evaluate the actual intended use of the SurePath liquid-based Pap smear and the PrepStain slide processor in a general screening population under typical clinical conditions. The prevalence of abnormalities was found to be consistent with that found in the general screening population in the U.S. As such, the results of the current study are clinically significant as they apply to the routine application of cervical carcinoma screening in the U.S.

REFERENCES

1. Bishop J, Bigner SH, Colgan TJ, et al. Multicenter masked evaluation of AutoCyte PREP thin layers with matched conventional smears. Including initial biopsy results. *Acta Cytol.* 1998;42(1):189–197.
2. Naryshkin S, Davey D. Terminology of false negative and false positive Pap smears. *Diagn Cytopathol.* 1995;14:1–2.
3. Farrington C, Manning G. Test statistics and sample size formulae for comparative binomial trials with null hypothesis of non-zero risk difference or non-unity relative risk. *Stat Med.* 1990;1:1447–1454.
4. Kotz, S, Johnson NL, Campbell B. Statistical differentials, method of. In: Kotz S, Johnson NL, editors. Encyclopedia of statistical sciences, volume 8. New York: John Wiley & Sons, Inc., 1988:646.
5. DeMay RM. The art and science of cytopathology. Chicago: ASCP Press, 1996:101
6. Syrjanen K, Kataja V, Yliskpski M, Chang F, Syrjanen S, Saarikoski S. Natural history of cervical human papillomavirus lesions does not substantiate the biologic relevance of the Bethesda System. *Obstet Gynecol.* 1992;79(5 Pt 1):675–682.
7. Lox JT, Schiffman M, Solomon D; ASCUS-LSIL Triage Study (ALTS) Group. Prospective follow-up suggests similar risk of subsequent cervical intraepithelial neoplasia grade 2 or 3 among women with cervical intraepithelial neoplasia grade 1 or negative colposcopy and directed biopsy. *Am J Obstet Gynecol.* 2003;188(6):1406–1412.
8. Vassilakos P, Saurel J, Rondez R. Direct-to-vial use of the AutoCyte PREP liquid-based preparation for cervical-vaginal specimens in three European laboratories. *Acta Cytol.* 1999;43:65–68.
9. Vassilakos P, Griffin S, Megavand E, Campana A. CytoRich liquid-based cervical cytology test: screening results in a routine cytopathology service. *Acta Cytol.* 1998;42:198–202.
10. Tench W. Preliminary assessment of the AutoCyte PREP. Direct-to-vial performance. *J Reprod Med.* 2000;45:912–915.