

# Performance of a Semiautomated Papanicolaou Smear Screening System

## *Results of a Population-Based Study Conducted in Guanacaste, Costa Rica*

Mark E. Sherman, M.D.<sup>1,2</sup>

Mark Schiffman, M.D., M.P.H.<sup>2</sup>

Rolando Herrero, M.D., Ph.D.<sup>3</sup>

Deidra Kelly, B.A., CT(ASCP)<sup>1</sup>

Concepcion Bratti, M.D.<sup>4</sup>

Laurie J. Mango, M.D.<sup>5</sup>

Mario Alfaro, M.D.<sup>6</sup>

Martha L. Hutchinson, M.D., Ph.D.<sup>7</sup>

Fernando Mena, M.D.<sup>6</sup>

Allan Hildesheim, Ph.D.<sup>2</sup>

Jorge Morales, M.D.<sup>6</sup>

Mitchell D. Greenberg, M.D.<sup>8</sup>

Ileana Balmaceda, M.D.<sup>6</sup>

Attila T. Lorincz, Ph.D.<sup>9</sup>

<sup>1</sup> Department of Pathology, the Johns Hopkins Medical Institutions, Baltimore, Maryland.

<sup>2</sup> Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, Maryland.

<sup>3</sup> International Agency for Research on Cancer, Lyon, France.

<sup>4</sup> Ministerio de Salud, San José, Costa Rica.

<sup>5</sup> Neuromedical Systems Inc., Suffern, New York.

<sup>6</sup> Caja Costarricense de Seguro Social, San José, Costa Rica.

<sup>7</sup> Department of Pathology, Rhode Island Infant and Women's Hospital, Providence, Rhode Island.

<sup>8</sup> Omnia Corporation, Philadelphia, Pennsylvania.

<sup>9</sup> Digene Corporation, Silver Spring, Maryland.

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**BACKGROUND.** Automated cytology devices have utility in quality assurance applications, but the effectiveness of these devices in primary screening is unknown.

**METHODS.** Enrollment smears obtained from 7323 women participating in a population-based study sponsored by the National Cancer Institute were screened manually in Costa Rica and then evaluated independently in the U.S. with the PAPNET system (Neuromedical Systems, Inc., Suffern, NY), a semiautomated, neural network-based device. Smears with abnormal PAPNET images were microscopically rescreened and then diagnosed by a U.S. cytopathologist. ThinPrep slides (Cytec Corporation, Boxborough, MA), prepared from rinses of the cytologic sampler, and cervigrams (National Testing Laboratories, Fenton, MO) were also evaluated. Women with any abnormal cytologic diagnosis or a positive cervigram were referred for colposcopy with biopsy and definitive therapy if indicated.

**RESULTS.** Based on the U.S. cytotechnologist's review of the PAPNET images, 1017 (13.9%) of 7323 smears were selected for manual screening, resulting in the selection of 492 (6.7%) possibly abnormal slides for referral to the U.S. pathologist. Ultimately, 312 smears (4.3% of the total) were diagnosed as containing squamous cells of undetermined significance or a more severe abnormality ( $\geq$ ASCUS), resulting, hypothetically, in the referral of 66.5% of women with a final diagnosis of a squamous intraepithelial lesion or a more severe abnormality ( $\geq$ SIL) and 86.0% of patients with  $\geq$ high grade SIL. Conventional microscopic screening performed in Costa Rica resulted in the hypothetical referral of 6.5% of patients with  $\geq$ ASCUS for colposcopy, including 69.5% of patients with  $\geq$ SIL and 79.8% of those with  $\geq$ high grade SIL.

**CONCLUSIONS.** In this study, PAPNET-assisted cytologic screening accurately identified smears obtained from women with high grade SIL or carcinoma. Determination of the clinical cost-effectiveness of PAPNET-assisted screening in routine practice awaits future study. [See editorial on pages 269-72, this issue.] *Cancer (Cancer Cytopathol)* 1998;84:273-80. © 1998 American Cancer Society.

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carcinoma screening, but their work on this project was rigorously masked. Drs. Sherman and Schiffman, who conducted the analysis and wrote the article, have no current or planned financial interests in Neuromedical Systems, Inc., or its competitors. Dr. Sherman has previously received research contract support from Neuromedical Systems, Inc., and Cytec Corporation, currently receives contract support from Upjohn, and has purchased human papillomavirus tests from and collaborated with Digene Corporation. Drs. Sherman and Schiffman are overseen by the Ethics

Office of the National Cancer Institute. Deidra Kelly worked on the PAPNET Food and Drug Administration Trial under a contractual agreement between the Johns Hopkins Medical Institutions and Neuromedical Systems, Inc.

Address for reprints: Mark E. Sherman, M.D., NIH, NCI/EEB, Executive Plaza North, Room 443, 6130 Executive Blvd., Rockville, MD 20892-7374.

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The success of Papanicolaou smear screening programs is highly dependent on the ability of cytotechnologists to identify abnormal smears using conventional light microscopy. Because the vast majority of smears are normal, screening is a monotonous task that is prone to human error. In particular, data indicate that smears obscured by inflammation, blood, or air-drying artifact may be readily misinterpreted as normal on routine screening, especially if abnormal cells are rare.<sup>1</sup> Consequently, the development of automated screening methods capable of specifically identifying the relatively small percentage of abnormal smears that require intensive manual microscopic screening would be desirable.

The PAPNET system (Neuromedical Systems, Inc., Suffern, NY) is a semiautomated, neural network-based device that is capable of facilitating the identification of false-negative smears, including those that may precede or are associated with the diagnosis of a high grade squamous intraepithelial lesion or carcinoma.<sup>2-10</sup> In the PAPNET system, conventionally prepared and stained smears are optically scanned and then evaluated with morphometric and neural network-based computer software, which results in the selection of 128 images most likely to represent possibly abnormal cells.<sup>2-13</sup> The images are recorded on a digital tape for later review by trained cytotechnologists. In the United States, the PAPNET system has received approval from the Food and Drug Administration for use as an adjunct test in the quality assurance review of normal smears. However, the reported experience with the PAPNET system in primary screening is limited.<sup>11-13</sup>

In this study, PAPNET testing results obtained for 7323 smears collected in a population-based, National Cancer Institute-sponsored screening study conducted in Guanacaste, Costa Rica, are presented. Guanacaste is a Pacific coastal province with a high incidence of cervical carcinoma, which perennially exceeds 30 per 100,000 women per year despite an existing screening program.<sup>14</sup> The smears included in the current analysis were examined twice. Initially, the slides were screened using conventional microscopy in Costa Rica and diagnosed locally for clinical purposes. Then the slides were subjected to PAPNET testing in the U.S. Based on a study cytotechnologist's interpretation of the images, slides were selected for manual rescreening and referral to a pathologist if indicated. Because smears were selected for microscopic screening in the U.S. based on the review of PAPNET images, the design of this study simulates the use of the instrument in primary screening.

## **MATERIALS AND METHODS**

### **Case Selection**

The subjects included in this study were selected from a cohort of randomly selected women participating in a National Cancer Institute-sponsored cervical carcinoma screening study conducted in Guanacaste, Costa Rica. The design of this multiyear, population-based, prospective study is presented in detail elsewhere.<sup>14</sup>

In brief, the cohort was assembled in 1993-1994 by conducting a comprehensive door-to-door survey of all adult women residing in randomly chosen census segments of Guanacaste. In total, 11,742 women were identified, of whom 10,738 were eligible to participate in the study (e.g., living, age 18 years or older, full-time residents, and mentally competent) and 10,049 (93.6%) were actually interviewed. Enrollment pelvic examinations were not performed on 583 virgins, and an additional 291 women refused or were physically unable to undergo examination. Thus, a pelvic examination was completed for 9175 participants, representing over 90% of the eligible, nonvirgin population.

PAPNET testing results were analyzed for only 7323 of the subjects who had a pelvic examination. Smears obtained from 1720 sequential unselected women examined in the middle of enrollment were not tested with PAPNET due to a backlog in the evaluation process. The continued delay in evaluations rendered the missing diagnoses less useful to the field staff; thus, we chose to skip the long-delayed PAPNET evaluations and move ahead to the timely evaluation of recently enrolled subjects. During the hiatus in PAPNET testing, a site visit to the cytopathology laboratory in Costa Rica was conducted to improve the technical quality of the Papanicolaou stain, which was suboptimal. Because the sensitivity of PAPNET testing (defined as the percentage of women with a final case diagnosis of SIL or carcinoma whose disease was detected by the test) was not significantly different before and after the hiatus (chi-square with Yates correction,  $P = 0.55$ ), all available PAPNET testing results were combined for this presentation.

There were two smaller sets of exclusions. PAPNET testing was not performed on 61 smears that were unsatisfactory for scanning. In addition, 71 PAPNET-tested cases were deleted from the statistical analysis because either the conventional or the PAPNET-assisted cytologic readings were unavailable due to mistakes in shipping or data recording.

### **Clinical Specimens**

Exfoliated cervical cells collected with the Cervex brush (Unimar, CT) were prepared as conventional

smears that were fixed with Pap Perfect (Medscand, FL) and stained by the Papanicolaou method in Costa Rica. Residual cells that remained on the brush were rinsed in vials containing 20 mL of PreservCyt (Cytyc Corporation, Boxborough, MA) and prepared as ThinPrep cytologic specimens (Cytyc Corporation) in the U.S.<sup>15</sup> A second cell sample obtained with a Dacron swab was placed in Specimen Transport Medium (Digene Corporation, Silver Spring, MD) and then shipped frozen to the U.S. for human papillomavirus (HPV) DNA testing using the Hybrid Capture tube test (Digene Corporation).<sup>16</sup> Because data indicate that only cancer-associated HPV types correlate closely with disease status, virologic data for these types alone (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, and 58) are presented.<sup>17</sup> Finally, the cervix was rinsed with 5% acetic acid, and two cervigrams (photographs of the cervix) were obtained (National Testing Laboratories, Fenton, MO) for visual screening.<sup>18</sup>

### Clinical Evaluation

Conventional smears were screened in Costa Rica and diagnosed by an expert Costa Rican cytopathologist (M.A.). ThinPreps were prepared in the U.S., stained using a modified Papanicolaou method, screened, and diagnosed by another expert cytopathologist (M.L.H.). The two cytologic specimens were diagnosed according to the Bethesda System (TBS) as within normal limits or as reactive cellular changes (negative), atypical squamous cell of undetermined significance (ASCUS), low grade squamous intraepithelial lesion (LSIL), high grade squamous intraepithelial lesion (HSIL), or carcinoma.<sup>19</sup> Cervigrams were prepared at National Testing Laboratories and classified by an expert evaluator (M.G.) as "normal" (including atypical) or "positive" with graded severity.

### PAPNET Review

After diagnosis in Costa Rica, the conventional Pap smear slides were forwarded to the PAPNET central facility in Suffern, NY, where dots placed by the cytotechnologists in Costa Rica indicating the location of abnormal cells were removed. The slides were bar coded and screened with the PAPNET system.

Briefly, the PAPNET system is a computer-assisted device that uses a combination of morphometric and neural network-based software.<sup>2-13</sup> For each smear, PAPNET review selects 128 images of possibly abnormal cells that are recorded on a digital tape as two screens. The first screen consists mainly of single cells, whereas the second primarily contains cell fragments and aggregates. The tapes are reviewed at a designated work station by a trained cytotechnologist. The two screens are initially reviewed as quadrants of 16 im-

ages each at a magnification of approximately  $\times 200$ . At the reviewer's discretion, individual images may be examined at  $\times 400$ .

In this study, the review of the PAPNET images (and the microscopic screening, if indicated) were performed by a single senior cytotechnologist at Johns Hopkins (D.K.). Cases in which the PAPNET images appeared normal were designated as "Normal 1" and then archived without further study. Cases in which the PAPNET images were considered abnormal were screened microscopically. Before the entire slide was screened, the coordinates of the suspicious PAPNET tiles were identified on the slide, and the cells in question were examined microscopically. Smears classified as normal following microscopic screening were designated as "Normal 2" and then filed. Reactive and possibly abnormal smears were forwarded along with the cytotechnologist's impression and the study data collection form to a pathologist (M.E.S.) for final diagnosis. Smears classified as negative by the pathologist were designated as "Normal 3" or "reactive," and the remaining, abnormal cases were classified according to TBS.

### Referral for Colposcopy

As individual screening results became known, patients with any of the following conditions were referred for colposcopy: 1) a physical examination suspicious for cancer; 2) a cytologic diagnosis of ASCUS or a more severe abnormality rendered on the conventional smear in Costa Rica, the ThinPrep cytologic slide, or the PAPNET-assisted review of the smear in the U.S.; or 3) a positive cervigram. Colposcopically directed biopsies of visible lesions were all performed by one experienced gynecologist (J.M.). In some cases, the placement of the biopsy was guided by suggestions from the cervigram evaluator. The biopsies were prepared as hematoxylin and eosin-stained sections in Costa Rica and diagnosed locally for clinical purposes (F.M.).

Patients with a histologic diagnosis of HSIL or carcinoma and women with a cytologic diagnosis of HSIL rendered by two observers were referred for large loop excision of the transformation zone (LEEP), cold knife cone, or hysterectomy. Some women with clinically evident cancer were treated with radiation therapy following biopsy confirmation. Patients with a single initial cytologic diagnosis of HSIL that was confirmed on review but not associated with HSIL on biopsy were also referred for LEEP if a lesion was identified colposcopically, there was no contraindication to treatment, and the patient consented. The remainder of these patients were referred for careful follow-up through the Costa Rican Social Security Sys-

tem. Final treatment decisions were made by the responsible physicians in Costa Rica.

As a quality control measure, a sample of 5% of enrolled women with normal screening results were examined colposcopically to test the sensitivity of the screening protocol relative to colposcopy. The absence of pathology in this group suggests that the combined screening protocol was sensitive in identifying women with colposcopic abnormalities.

### Final Diagnoses

Final case diagnoses were based on a review of all of the pathologic material (by M.E.S.) without knowledge of HPV DNA testing results. Final "negative" case diagnoses included patients with negative screening tests, as well as women referred for colposcopy based on ASCUS cytology who were subsequently judged to be normal on colposcopy (including the biopsy, if taken). A final diagnosis of "equivocal" was assigned to cases with various combinations of inconclusive test results, including the following: a cytologic diagnosis of LSIL using one cytologic technique that was not corroborated by the other techniques nor histologically confirmed, a positive cervigram with normal cytology and histopathology, or equivocal results following review of all available tests. Final case diagnoses of LSIL or HSIL were confirmed by biopsy and sometimes also by agreement of at least two of the three cytologic diagnoses. Histologic confirmation was obtained for 39% of LSIL cases, 93% of HSIL cases, and 100% of carcinoma cases.

### Data Analysis

The data analysis was limited to the 7323 women who received both a conventional smear diagnosis and a PAPNET-assisted diagnosis on a satisfactory specimen. The performance of the PAPNET system was assessed primarily by comparing the diagnoses of smears based on PAPNET screening with the final case diagnoses. For reference, the original diagnoses rendered in Costa Rica using manual screening were compared with final diagnoses in an analogous manner. In addition, the frequencies with which cancer-associated types of HPV DNA were detected in women using the two screening approaches were compared. For women with abnormal cytologic diagnoses rendered by only one of the two cytologic methods, final case diagnoses were compared and HPV prevalences were computed as independent adjudications of the discrepancies.

The sensitivity and specificity of PAPNET review and conventional cytology were calculated by defining final case diagnoses of LSIL, HSIL, or carcinoma as cases of "disease" and defining final diagnoses of

"negative" (normal or reactive) and "equivocal" as "no disease." Sensitivity was recalculated for HSIL and carcinoma, excluding LSIL, because of the extreme importance of these high grade lesions. Although final case diagnoses of "equivocal" represented a heterogeneous group of conditions, undoubtedly including some unrecognized true lesions, these cases were best categorized as "no disease" in aggregate based on an assessment of all available data.

Before the study, a cytologic diagnosis of ASCUS was set as the threshold for colposcopy referral for all of the three cytologic screening techniques. Therefore, diagnoses of ASCUS, SIL, and carcinoma were considered to represent positive cytologic test results.

For statistical testing, standard contingency table methods were used.

Based on agreements made between collaborators at the start of the study, this analysis will be limited to the performance of PAPNET testing only. The performance of other screening methods will be examined in other publications.

## RESULTS

### PAPNET-Directed Screening

The results of PAPNET review are compared with the final case diagnoses in Table 1. Of the 7323 PAPNET-tested smears analyzed, 1017 smears (13.9%) were selected for manual rescreening (Normal 2 or greater), and the remainder were classified as normal based on the images alone (Normal 1). The ability of the PAPNET system to detect and display abnormal cells is shown by the cross-tabulation in Table 1. Compared with the final diagnoses, 99 (63.9%) of 155 cases with a final diagnosis of LSIL, 91 (88.4%) of 103 with HSIL, and 11 (100.0%) of 11 with carcinoma were selected for manual rescreening based on the U.S. cytopathologist's interpretation of the PAPNET video images.

In 525 cases (7.2%), a smear that had been classified as possibly abnormal based on the review of the PAPNET images was considered to be negative by the cytotechnologist following manual screening (Normal 2). These smears were not forwarded to the pathologist. Two cases in this group had final diagnoses of HSIL. Of the 492 smears reviewed by the U.S. pathologist, 180 (36.6%) were classified as normal (Normal 3) or reactive (including one final diagnosis of carcinoma and another of HSIL).

With a cytologic diagnosis of ASCUS considered a positive screening result, PAPNET testing yielded a sensitivity of 66.5% for lesions of LSIL or worse, and 86.0% restricted to the detection of HSIL or carcinoma. To obtain this sensitivity, 4.3% of this random, community-based population would have been referred for colposcopy. The specificity of the approach

**TABLE 1**  
Automated Screening Compared with Final Case Diagnoses<sup>a</sup>

Automated screening	Final case diagnosis					Total
	Negative	Equivocal	LSIL	HSIL	CA	
Normal 1	5892 (89.8)	346 (70.5)	56 (36.1)	12 (11.6)	0 (0.0)	6306 (86.1)
Normal 2	471 (7.2)	41 (8.4)	11 (7.1)	2 (1.9)	0 (0.0)	525 (7.2)
Normal 3	39 (0.6)	4 (0.8)	0 (0.0)	0 (0.0)	0 (0.0)	43 (0.6)
Reactive	99 (1.5)	29 (5.9)	7 (4.5)	1 (1.0)	1 (9.1)	137 (1.9)
ASCUS	60 (0.9)	55 (11.2)	23 (14.8)	25 (24.3)	4 (36.4)	167 (2.3)
LSIL	1 (<0.1)	12 (2.4)	53 (34.2)	13 (12.6)	0 (0.0)	79 (1.1)
HSIL	1 (<0.1)	4 (0.8)	5 (3.2)	46 (44.7)	5 (45.4)	61 (0.8)
CA	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.9)	1 (9.1)	5 (0.1)
Total	6563	491	155	103	11	7323

Normal 1: negative based on review of PAPNET images alone; Normal 2: negative based on cytotechnologist's screening of smear following classification of PAPNET images as "review"; Normal 3: negative based on pathologist's interpretation of smear following PAPNET review and manual screening of smear; ASCUS: atypical squamous cells of undetermined significance; LSIL: low grade squamous intraepithelial lesion; HSIL: high grade squamous intraepithelial lesion; CA: carcinoma.

<sup>a</sup> Data are expressed as no. (%) column total.

**TABLE 2**  
Manual Screening Compared with Final Case Diagnosis<sup>a</sup>

Manual screening	Final case diagnosis					Total
	Negative	Equivocal	LSIL	HSIL	CA	
Normal	4283 (65.3)	200 (40.7)	35 (22.6)	9 (8.7)	1 (9.1)	4528 (61.8)
Reactive	2171 (33.1)	108 (22.0)	24 (15.5)	13 (12.6)	0 (0.0)	2316 (31.6)
ASCUS	94 (1.4)	23 (4.7)	4 (2.6)	3 (2.9)	1 (9.1)	125 (1.7)
LSIL	0 (0.0)	142 (28.9)	65 (41.9)	10 (9.7)	0 (0.0)	217 (3.0)
HSIL	15 (0.2)	16 (3.3)	27 (17.4)	56 (54.4)	3 (27.3)	117 (1.6)
CA	0 (0.0)	2 (0.4)	0 (0.0)	12 (11.7)	6 (54.6)	20 (0.3)
Total	6563	491	155	103	11	7323

ASCUS: atypical squamous cells of undetermined significance; LSIL: low grade squamous intraepithelial lesion; HSIL: high grade squamous intraepithelial lesion; CA: carcinoma.

<sup>a</sup> Data are expressed as no. (%) column total.

(i.e., the percentage of normal or reactive PAPNET tests among women found not to have LSIL, HSIL, or carcinoma) was 98.1%.

Of note, 23 LSILs (14.8%), 25 HSILs (24.3%), and 4 carcinomas (36.4%) were diagnosed as ASCUS by the U.S. pathologist. Many of these smears were considered suspicious, but an unequivocal diagnosis was precluded by air-drying artifact, atrophy-related changes, obscuring blood and inflammation, or sub-optimal staining.

### Conventional Microscopic Screening

As a point of reference, the results of conventional microscopic screening performed in Costa Rica are compared with final case diagnoses in Table 2. Manual screening, with a threshold of ASCUS, resulted in the colposcopy referral of 6.5% of the subjects, including 96 (61.9%) of 155 with LSIL, 81 (78.6%) of 103 with

HSIL, and 10 (90.9%) of 11 with carcinoma. Thus, the sensitivity of conventional screening was 69.5% for the detection of LSIL or a more severe lesion and 79.8% restricted to HSIL and carcinoma. The specificity was 95.9%. In the conventional review (performed in Costa Rica), therefore, a higher percentage of women received a cytologic diagnosis of SIL or carcinoma (4.8%, compared with 2.0% for PAPNET), but the specificity of these diagnoses based on the final case diagnoses was lower than in the PAPNET review (conducted in the U.S.).

### Detection of HPV DNA versus Cytologic Diagnosis

Cancer-associated HPV types were detected in a similar percentage of women with normal cytologic diagnoses based on PAPNET review alone (Normal 1) and in those who were classified as negative following manual screening prompted by PAPNET review (Nor-

**TABLE 3**  
**Detection of Cancer-Associated Types of HPV DNA vs. PAPNET-Assisted Cytologic Diagnosis<sup>a</sup>**

Automated screening diagnosis	No. (%) with HPV DNA detected
Normal 1 (n = 6301)	311 (4.9)
Normal 2 (n = 525)	29 (5.5)
Normal 3 (n = 43)	3 (7.0)
Reactive (n = 137)	22 (16.1)
ASCUS (n = 167)	71 (42.5)
LSIL (n = 78)	53 (68.0)
HSIL (n = 61)	52 (85.2)
Carcinoma (n = 5)	4 (80.0)

Normal 1: negative based on review of PAPNET images alone; Normal 2: negative based on cytotechnologist's screening of smear following classification of PAPNET images as "review"; Normal 3: negative based on pathologist's interpretation of smear following PAPNET review and manual screening of smear; ASCUS: atypical squamous cells of undetermined significance; LSIL: low grade squamous intraepithelial lesion; HSIL: high grade squamous intraepithelial lesion; CA: carcinoma.

<sup>a</sup> Six women with missing HPV test results were excluded from the table.

**TABLE 4**  
**Detection of Cancer-Associated Types of HPV DNA vs. Cytologic Diagnosis Using Conventional Screening<sup>a</sup>**

Conventional screening diagnosis	No. (%) with HPV DNA detected
Normal (n = 4525)	208 (4.6)
Reactive (n = 2314)	157 (6.8)
ASCUS (n = 125)	15 (12.0)
LSIL (n = 216)	71 (32.9)
HSIL (n = 117)	81 (69.2)
Carcinoma (n = 20)	13 (65.0)

HPV: human papillomavirus; ASCUS: atypical squamous cells of undetermined significance; LSIL: low grade squamous intraepithelial lesion; HSIL: high grade squamous intraepithelial lesion.

<sup>a</sup> Six women with missing HPV test results were excluded from this table.

mal 2, Table 3). HPV DNA detection was more frequent among women diagnosed with reactive changes than among women in the normal categories. Women diagnosed with ASCUS using PAPNET-assisted screening more frequently had a positive HPV test than women with negative cytology, but were less likely to be positive than women with SIL or carcinoma.

The frequency with which cancer-associated HPV types were detected in women with normal smear results based on manual screening and diagnosis in Costa Rica was comparable to that obtained with the PAPNET-assisted review (Table 4). However, HPV detection was appreciably lower in cases of SIL diagnosed in Costa Rica as compared with those diagnosed in the U.S.

### Colposcopy Referral Based on a Positive Result with a Single Cytologic Technique

There were 189 women referred for colposcopy who had both an abnormal conventional reading and an

**TABLE 5**  
**Final Case Diagnoses for Women Referred for Colposcopy after Either an Abnormal Conventional or PAPNET-Assisted Cytologic Diagnosis ( $\geq$ ASCUS)**

Final case diagnosis	Referred after PAPNET testing only	Referred after conventional cytology only
Negative	53 (43.1)	100 (34.5)
Equivocal	39 (31.7)	151 (52.1)
LSIL	19 (15.4)	34 (11.7)
HSIL	11 (8.9)	4 (1.4)
Carcinoma	1 (0.8)	1 (0.3)
Total	123 (100.0)	290 (100.0)

LSIL: low grade squamous intraepithelial lesion; HSIL: high grade squamous intraepithelial lesion.

abnormal PAPNET-assisted diagnosis ( $\geq$ ASCUS), 123 referred with abnormal PAPNET testing results alone, and 290 referred for abnormal conventional cytology only. The final case diagnoses for women who were referred for colposcopy with an abnormal result from only one of the two techniques are shown in Table 5. PAPNET testing alone identified 19 LSILs, 11 HSILs, and 1 carcinoma, whereas conventional cytology identified 34 LSILs, 4 HSILs, and 1 carcinoma. Cancer-associated HPV types were detected in 46 (37.4%) of 123 women with abnormal PAPNET testing results only, as compared with 46 (15.9%) of 290 women who had only abnormal conventional cytology ( $P < 0.001$ ).

### DISCUSSION

We assessed the performance of a semiautomated, neural network-based cervical carcinoma screening device in a population-based study conducted in Guanacaste, Costa Rica. The design of this study simulated the use of the device in primary cancer screening.

Based on the experienced U.S. cytotechnologist's review of computer images generated with the device, smears obtained from 13.9% of participants were selected for manual rescreening. Approximately half of these slides were referred to a pathologist for final diagnosis, whereas the remainder were considered normal microscopically by the cytotechnologist and not examined further. Using a threshold of ASCUS (or greater) for colposcopy referral, as agreed upon at the start of the study, would have resulted in colposcopy referral for 4.3% of women, with the detection of 86% of HSILs and carcinomas. Detection of all SILs and carcinomas with this approach was 66%. Though not completely comparable (because separate pathology teams were involved), manual screening of the same smears in Costa Rica would have resulted in colposcopy referral for 6.5% of the women, including 80% of

those with HSIL or carcinoma. The sensitivity for detecting all SILs and carcinomas with conventional screening was 70%. Therefore, by employing a colposcopy cutoff of ASCUS and using the device in the manner described in this study, we were able to achieve good results with automated cytology. Although experience with real-time and simulated primary screening with PAPNET is limited, previously reported results have also been favorable.<sup>11-13</sup>

The frequency with which cancer-associated types of HPV were detected in women classified as normal based solely on the interpretation of the computer-generated images was similar to that observed in women classified as normal with manual screening, supporting the conclusion that automated screening was comparably sensitive. As has been previously reported for conventional cytology, PAPNET-assisted screening resulted in diagnoses of negative, ASCUS, and SIL, which corresponded to progressively higher percentages of cancer-associated HPV detection.<sup>17</sup> Accordingly, these data suggest that computer-assisted screening permitted the appropriate stratification of patients according to cancer risk using TBS.

PAPNET-assisted screening would have resulted in the hypothetical referral of 4.3% of women for colposcopy, whereas manual screening would have resulted in the referral of 6.5% of patients. The detection of cancer-associated HPV types was considerably more frequent among women with diagnoses of ASCUS, LSIL, and HSIL based on automated screening in the U.S., as compared with the conventional cytology alone performed in Costa Rica. Because PAPNET-assisted screening was performed in the U.S. and conventional screening was conducted in Costa Rica, differences in diagnostic criteria may limit the validity of directly comparing the results achieved with the two methods. Nonetheless, automated screening achieved high sensitivity with a low percentage of colposcopy referral in this population.

In the automated review, 24% of the HSILs and 36% of the carcinomas were classified as ASCUS. Although a number of these cases were considered highly suspicious for SIL or carcinoma at the time of diagnosis, exact classification was precluded by poor staining quality, air-drying artifact, obscuring blood or inflammation, and atrophy-related changes. Because it had been decided at the outset that all women with ASCUS cytology or worse would be referred for colposcopy, the impetus to commit to a specific diagnosis for these patients was limited.

The analyses presented in this report use final case diagnoses that were based on all three cytologic techniques as well as all resulting biopsies, LEEPs, and hysterectomy specimens. This approach resulted in

7% of the subjects in this study receiving a final diagnosis of "equivocal" because women diagnosed with a diagnosis of SIL using any of the methods were so classified. Overall, the data indicate that the majority of the women in the equivocal category received a false-positive test result and were actually disease free. However, we acknowledge that the detection of cancer-associated HPV types in these women was more frequent than in normal women, and some of these patients undoubtedly had an unrecognized SIL. However, the high percentage of equivocal diagnoses could not have had an important effect on the conclusions concerning PAPNET testing results.

The interpretation of the results of this study is limited by the finding that about one-third or more of the smears examined would probably be considered limited for interpretation using TBS. This may account, at least in part, for the observation that cervical carcinoma rates in this population have remained in excess of 30 per 100,000 for decades, despite screening. Thus, the performance of various screening methods in this population might differ from that in well-screened populations with a lower prevalence of cervical carcinoma. In addition, the results obtained in clinical practice in the U.S. may differ from those obtained in a research study of this type. Finally, the performance of automated screening as performed in this study is dependent on the expertise of the cytopathology laboratory in reviewing the computer-generated images, microscopic screening, and cytologic diagnosis. The importance of experience in rendering expert evaluations of PAPNET images has been emphasized previously.<sup>5</sup>

Recently, multiple cervical screening technologies have been developed in the hope of improving cervical carcinoma screening. Whether introduction of new technical advances will improve screening in a cost-effective manner remains to be determined.

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