LightTouch™— Non-invasive Spectroscopic Detection of Cervical Cancer with High Sensitivity and Superior Specificity

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Introduction

The present study was designed to assess the potential impact on cervical disease management that spectroscopic imaging would have if employed as a pre-colposcopy triage test. The study endpoints included both sensitivity to detect biopsy proven cervical dysplasia, especially moderate and severe forms (CIN2+), as well as specificity to rule out benign cervical conditions that also had been scheduled for colposcopy and biopsy. The study population included women with a history of abnormal cervical cytology or other risk factors, such as previously documented histopathology of cervical dysplasia in need of follow-up.

Objective

The objective of the study was to evaluate the potential safety and effectiveness of tissue spectroscopy for the diagnosis of cervical cancer in a prospective multicenter, IRB approved study of women scheduled for colposcopy on the basis of an abnormal Pap test or other risk factors.

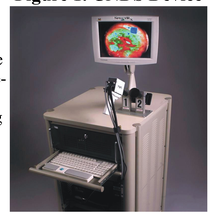
Cervical Neoplasia Detection System (CNDS)

The device system (Guided Therapeutics, Inc. Norcross, GA, USA) used in the study is a nonsignificant risk device by FDA standards that noninvasively and automatically scans the ectocervix and distal endocervix for disease related changes in fluorescence and reflectance spectra (see Figure 1).

Alterations in fluorescence spectra are indicative of metabolic changes associated with neoplasia, while alterations in reflectance and scattering are indicative of structural changes associated with neoplasia, such as epithelial thickening, nuclear size, nuclear content and angiogenesis.[1,2,3,4]

A plurality of equally spaced points over a one-inch diameter area of the cervix was automatically scanned during a four-minute period using a filtered xenon arc lamp as an illumination source.

Figure 1. CNDS Device



For cervical tissue reflectance measurements, broadband spectral output ranging from about 350 to 900nm was automatically applied under software control to the cervix using the same xenon arc lamp. The resultant reflectance spectral output from the cervical tissue was imaged onto the CCD camera and stored for processing and analysis.

For cervical tissue fluorescence measurements, light from the arc lamp was band pass filtered to limit exposure of the cervix to bands within the 300 to 500 nm range. These spectral bands are known to excite fluorophores associated with neoplastic processes as described above. Each of the fluorescence wavelengths were applied automatically under software control in a predetermined order and scan pattern. The resultant fluorescent spectral output of the cervical tissue was imaged onto a charge coupled device (CCD) camera and stored for processing and analysis.

The system consists of two main physical components, the hand-held unit and the base unit (Figure 1). The handheld unit is connected to the base unit via fiberoptic cables for transmission of light to and from the base unit, which contains the xenon arc lamp, optical processing elements (e.g., filters and lenses) and the CCD camera on a rolling cart (CNDS Device).

The other major component of the CNDS is a computer for control and data processing. This includes the capability for a diagnostic algorithm based on spectroscopic information measured from the cervix, calibration data and other patient data, such as Pap results or patient demographic data.

Figure 2. Race Distribution Study 1

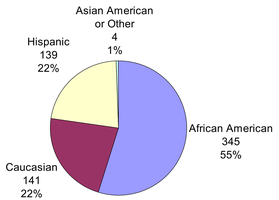


Table 1. Age Distribution Study 1

Category	Number	Percent
Median	27.7	--
Range	18-75	--
< Age 30	354	57.6
> Age 30	275	42.4

Methods

Subjects meeting the inclusion criteria underwent spectroscopy of the cervix during their colposcopy visit. Spectroscopy measurements taken over a four minute scan period were integrated by a cross-validated pattern recognition algorithm and compared with expert panel resolved histopathology to yield sensitivity and specificity of cervical spectroscopy. After the spectroscopy measurements were taken, a sample for Pap testing was taken, followed by colposcopy and biopsy, if indicated.

Histopathology Quality Control

Each clinical site fixed tissue per current clinical practice. An additional slide adjacent to the diagnostic slide was also cut and sent to a clinical pathology lab (QA 1). If QA 1 disagreed with the diagnosis of the clinical site pathologist, the slide was then sent to a third pathologist (QA 2). A case was assigned to a diagnostic category when either the site and QA 1 agreed or two out of three diagnoses agreed (i.e., either benign, CIN1, CIN2+, using the most severe disease grade for each case). A case was considered nonevaluable when all three pathologists disagreed.

Results

Two studies are presented here. The first study was conducted at four clinical sites (University of Miami, Medical College of Georgia, University of Connecticut, and Emory University). The second study was conducted at the University of Texas Southwestern Medical Center.

In the first study, 648 consecutive women from the four clinical sites met inclusion criteria, signed the consent form and were therefore eligible to participate in the study. Data could not be collected from 19 subjects because they withdrew before the study could be completed. Demographic data for these subjects are summarized in Figure 2 and Table 1. No adverse events were reported.

All women underwent colposcopic examination and all but 42 had a Pap test result and valid histopathology available. Of the 587 subjects that had valid Pap and histopathology and/or colposcopy results available, 15 cases could not be analyzed because of a device or operator error. Thus sensitivity and specificity of the test was calculated for the remaining 572 evaluable subjects (see Table 2).

Table 2. Sensitivity and specificity of spectroscopy by disease category for Study 1.

All Cases (N = 572)	SENSITIVITY		SPECIFICITY*
Disease	CIN2+	CIN1	No CIN
Number Tested	142	180	250
Number Correct	135	135	138
Percent Correct	95.1	75	55.2
95% Confidence Interval	(91.55, 98.65)	(68.67, 81.33)	(49.04, 61.36)

* Note: In 149 cases, biopsy was not performed because expert colposcopy indicated a normal cervix and the Pap test did not indicate dysplasia. Most of these cases were referred to colposcopy on the basis of other risk factors and are categorized as benign for the purposes of statistical analysis.

Figure 3. Race Distribution Study 2

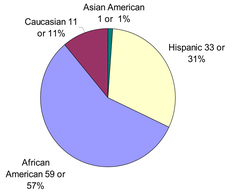


Table 3. Age Distribution Study 2

Category	Number	Percent
Median	31	--
Range	16-57	--
< Age 30	50	48

Sensitivity of cervical spectroscopy for CIN2+ lesions (n= 142) was 95.1% with a corresponding specificity for benign lesions (e.g., normal tissue, inflammation, metaplasia; n= 250) of 55.2%. The test also identified 75.0% of CIN1 lesions (n= 180) as positive. A secondary algorithm was employed to reduce the number of CIN1 lesions as positive by spectroscopy without a significant reduction in sensitivity for CIN2+ lesions.

Color enhanced maps of the cervix could be constructed from the spectroscopic data (See Figure 4).

In the second study, a comparison was done for the performance of cervical spectroscopy (CS) with HPV DNA testing (HPV) when used in conjunction with cytology in detecting CIN2+.

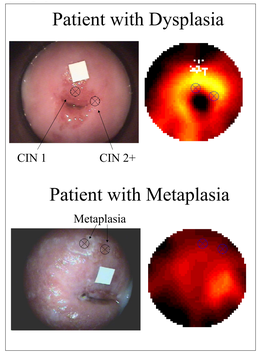
Of the 113 subjects consented to participate, 109 qualified and completed the study. The other 4 were ineligible by inclusion criteria or withdrew from the study after the consent process had been completed. Data was not collected from 5 subjects because of device malfunction (n=3) or operator error (n=2). Two study subjects' data were not included in the data analysis due to 3-way histopathology discordances pertaining to their histological diagnoses. Two cases with borderline histology (e.g., CIN1 to 2) were included in the analysis. Data from 102 study subjects were used for analysis. Demographic data are summarized in Table 3 and Figure 3.

Table 4. Sensitivity and specificity of spectroscopy by disease category for Study 2.

All Cases (102)	SENSTIVITY	SPECIFICITY	
Disease	Either Strategy (CIN2+)	Pap + HPV	Pap + CS
Number Tested	18	84	84
Number Correct	17	23	55
Percentage Correct	95	27.4	65.5

Pap+HPV and Pap+CS achieved equivalent sensitivities (95%) for high-grade lesions, with both detecting 17 of 18 histology confirmed CIN2+ lesions. Pap+HPV had a specificity of only 27.4% compared to 65.5% for Pap+CS (p<.0001) (See Table 4).[2]

Figure 4. Cervical Maps
(Biopsy sites marked with Xs)



Summary and Conclusions

Spectroscopy of the cervix has the potential to accurately detect moderate and high-grade cervical dysplasia while also reducing the false positive rate for benign cervices. The test is relatively simple to implement, and as previously reported[5], was well accepted by subjects enrolled in the study. An additional strength of the test is its safety; it is noninvasive and no adverse events were reported. If the present results are validated in pivotal clinical trials and in commercial use, a significant reduction in the number of unnecessary follow-on tests, colposcopies and biopsies could be expected. The ability to utilize spectroscopic information for mapping cervical lesions could assist the physician in making biopsy and/or treatment decisions.

Figure 5. CNDS & Next Generation Device

A next generation device (Figure 5, on right) is currently enrolling subjects in a pilot clinical study. This new device is 25% less in weight and volume, and the measurement time has been reduced to less than one minute. The cost of the new device has also been significantly reduced.

References

[1]Drezek RA, Richards-Kortum R, Brewer MA, Feld MS, Pitris C, Ferenczy A, et al. Optical Imaging of the Cervix. *Cancer (Supp.)* 2003: 98/9; 2015-2027.

[2]Werner CL, Griffith WF, Ashfaq R, Diana Gossett D, Wilkinson E, Raab S, Bambot S., Mongin D, Faupel M. Comparison of HPV Testing and Spectroscopy Combined with Cervical Cytology for the Detection of High-grade Cervical Neoplasia. . *J Lower Genital Tract Dis*. 2006. In press..

[3]Tromberg B. Detection of cervical pre-cancer using reflectance spectroscopy. Presented at the Office of Women's Health Symposium on Biomedical Optical Diagnostics and Imaging. Washington, DC, October 1997.

[4]Ferris DG, Lawhead RA, Dickman ED, Holtzapple N, Miller JA, Grogan, et al. Multimodal hyperspectral imaging for the noninvasive diagnosis of cervical neoplasia. *J Lower Genital Tract Dis*. 2001;2:

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CAUTION - Investigational device. Limited by federal law to investigational use. The availability of any product in the U.S. developed from these technologies is dependent on FDA marketing approval.

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