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Original Research

Performance of multimodal hyperspectroscopy for scanning of cervix in pregnant women: a preliminary study

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Summary

Purpose: This study aimed to measure the performance of cervical multimodal hyperspectroscopy (MHS) when compared to Pap smear, and HPV testing in low-risk pregnant women followed up in a university clinical setting. *Materials and Methods:* Each of 91 pregnant women underwent Pap smear, HPV testing, and during the first 20 weeks of pregnancy. In 84 subjects evaluable data were collected and analyzed. *Results:* Average age of women was 30 ± 5.2 years. The specificity of MHS performed in pregnant women seems to be significantly better ($p < 0.01$) than the data of non-pregnant women published earlier. The specificity of MHS in pregnant women with normal Pap smear result is estimated to be 92.9%. The procedure was also well-tolerated with a patient satisfaction rate of 92%. *Conclusions:* Scanning of the cervix by MHS may perform similar to Pap smear + HPV testing and may have a higher specificity in low-risk pregnant women.

Key words: Cervical multimodal hyperspectroscopy; Pap smear; HPV testing; Cervical scanning.

Introduction

Although there have been recent developments in preventing and the introduction of new management guidelines for cervical disease, it continues to be a significant health problem in developing countries. Introduction of HPV vaccines and Pap smear plus HPV testing are chief among these, but both are too costly to be repeatedly covered by national insurance system for preventive or screening purposes. Also, despite tremendous efforts, some women are not screened because they do not show up to their routine examinations.

In developing countries, pregnancy is a particular period in the sense that even women who do not attend their gynecologic exams, do so when they become pregnant. It represents an opportunity for the clinician to screen for cervical pre-invasive disease.

The authors report the results of a preliminary clinical study to evaluate the potential of multimodal hyperspectroscopy (MHS) for screening purposes in the first half of the pregnancy. MHS is the concurrent use of multiple types of tissue spectroscopy, whereby specific wavelengths of light are focused on the cervix and the response of cells and cellular structures, as manifested in the reflected light, is resolved spectrally and imaged onto a high-resolution sensor [1]. The primary goal of this study was to compare the results of MHS to Pap smear plus HPV test screening in low-risk pregnant women with no known history of HPV or cervical pre-invasive disease. As far as the present authors are aware, this is the first study of MHS performed on pregnant women population.

Materials and Methods

There has been increasing use of MHS to detect and evaluate the pre-invasive changes occurring in the cervix. The present study used an easily operated and cost-effective device that combines fluorescence and reflectance spectroscopy. The advantage of combining spectroscopic modes is that fluorescence spectroscopy identifies metabolic changes associated with neoplasia, while reflectance spectroscopy indicates the presence of structural changes within a tissue indicative of neoplasia [2-8]. The device is used only by medically trained personnel, whose time to learn the procedure is complete after two to three cases.

The study was performed in a university clinic outpatient setting after obtaining a university's ethical committee approval. The study population consisted of pregnant women who were at low risk for cervical preinvasive disease between 5-20 weeks of gestation. Women who have a history of HPV infection or abnormal smears and a history of any prior procedure on the cervix were left out of the study. All enrolled pregnant women had low-risk pregnancies. Pregnant women having any chronic or immunosuppressive disease or spotting/bleeding of any sort were also left out of the study. After thoroughly explaining the details of the procedure and obtaining informed consent, all patients were prepared for standard pelvic examination. First, excessive mucus or discharge was removed using saline, and then sight tube of calibrated MHS device was inserted through the vaginal speculum. Cervical os was visualized adequately, focused and then spectrophotometric measurements were made automatically under software control [1]. The examination took around one minute, and after completion, the sight tube was withdrawn. A swab sample of the cervix, both for PAP smear exam and HPV genotyping was taken. Smears were sent to two blinded experienced gyno-pathologists in

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Table 1. — *Patients' age and gestational age.*

		Number	Percent (%)
Age (years)	Median	31 years	-
	Range	(19-45 years)	-
	<30 years	41	48.8%
	31 years and older	43	51.2%
Gestational age	<10 weeks	12	14.3%
	>10 weeks	72	85.7%

Table 2. — *Comparison of specificity in pregnant and published non-pregnant women data.*

	DeSantis *et al.* [9] specificity	Current study specificity	*p*
Number tested	226	84	
Smear compatible	128	78	
Percent consistent	56.6%	92.9%	<0.001

Table 3. — *Comparison of specificity of MHS by age group between pregnant and non-pregnant women.*

Age	Twiggs *et al.* [1] Specificity normal (%)	Current study Specificity normal (%)	*p*
21-30	33.9% (76/224)	97.4% (38/39)	<0.001
31 and older	45.8% (119/260)	88.4% (38/43)	<0.001

the university's pathology department. If both agreed, this served as the pathologic gold standard diagnosis. If there was a disagreement between the pathologists, the specimens were evaluated by an additional gyno-pathologist as a "tie-breaker." All HPV genotyping samples were sent to the same private accredited laboratory for evaluation. Although colposcopy was offered to women who were either having HR-HPV or MHS screening positive, none of them consented to this procedure. After completion of all of the procedures, all enrolling pregnant women filled out a questionnaire about their experience.

This preliminary study aimed to evaluate the feasibility and to compare the results of MHS screening for cervical disease in pregnant women and low-risk non-pregnant women. Prior published data about MHS was used for comparison. Enrollment and data collection took place from June to September 2015. McNemar and z-tests were used for comparing data, and SigmaStat 3.5 software was used for all statistical analysis.

Results

A total of 91 consecutive pregnant women fulfilling the inclusion/exclusion criteria applying at University's perinatology outpatients' clinic agreed to participate in the study. All participants were more significant than five, and less than 20 weeks of gestation. In seven patients, PAP smear test results showed a paucity of cells for evaluation; therefore these patients were dropped out. In a total of 84 subjects, evaluable data were collected and analyzed. The average age of women was 305.2 years. In Table 1, the characteristics of the patients are outlined.

In all pregnant women, PAP smear results were low-risk; in contrast, MHS screening resulted in high risk in six patients (7.1%). This MHS screening result was compared to the data published in 2006 by DeSantis *et al.* in terms of specificity in Table 2 [9]. The specificity of MHS performed in pregnant women seemed to be significantly better ($p < 0.01$) than the data of non-pregnant women published earlier. The specificity of MHS in pregnant women with normal PAP smear result is estimated to be 92.9%.

Table 4. — *Percent of hgh risk (HR) HPV positivity in MHS screened cases with normal PAP smear result.*

	Werner et al (10)	Current study	p
HR HPV (+) %	(11/33) 33.3%	(6/84) 7.1%	<0.001

Table 5. — *HR-HPV versus MHS screening results in screened pregnant women.*

	Day of study MHS result		Total
Day of study HR-HPV	Negative	Positive	
Positive	6	0	6
Negative	72	6	78
Total	78	6	84

When women in the study are divided into age groups, and the results are compared to the data of non-pregnant women published by Twiggs *et al.* in 2013, it was seen that specificity of MHS was significantly higher in the current pregnant women study in both age groups (Table3) [1]. The specificity of MHS was calculated to be 97.4% and 88.4% in age 21-30 group and age 31 and older group, respectively. Of note, in order to match and compare correctly with the published data, two patients aged 19 and 20 years were left out of the < 30-year age group, reducing the number of women to 39 in this specific group.

The proportion of high-risk HPV (HR-HPV) positivity in the present pregnant study group was also compared to available data of HR-HPV in non-pregnant women with normal PAP smear results extracted from Werner *et al.* [10]. The proportion of HR-HPV positivity was only 7.1% in the present study group, and this was significantly lower than Werner's data as outlined in Table 4.

The relationship between the results of HR-HPV and MHS screening in the present study group is also detailed in Table 5. MHS screening showed an increased risk in six cases with negative HR-HPV results, while HR-HPV test was positive in six other cases whose MHS screening were negative.

The majority, 92%, of women were quite satisfied with the procedure overall. The first reason for satisfaction was the result of the test was readily available. Of women who were dissatisfied, the reasons were the fear of bleeding or causing abortion, and relative significant dimensions of the sight tube causing discomfort. The results of the questionnaire are summarized in Table 6.

Table 6. — *Pregnant women's responses to satisfaction survey about MHS screening.*

	Satisfied	Dissatisfied	Not sure
Percent (%)	92	6	2
Major reasons (%)	- Results readily available (90%) - Procedure may have the advantage of showing changes that may arise later during the next two years (65%)	- The dimension of sight tube is big and causing discomfort (85%) - Fear of abortion and unforeseen bleeding (60%) - Have questions about the reliability of the procedure (2%)	- Screening for cervical pathologies not necessary during pregnancy

Discussion

This study compared the feasibility of MHS in cervical screening purposes in low-risk pregnant women less than 20 weeks of gestation. Another possible use of this technology, as using it as a triage test for detection of high-grade dysplasia was not evaluated [10]. The aim was to compare the results of MHS screening in pregnant women to published relevant non-pregnant women data. Since low-risk and normal Pap smear result groups were selected, specificities were compared. MHS performed similar or better in pregnant women than in the non-pregnant in terms of specificity. There were no adverse events reported, and the procedure was tolerated well. As far as teh present authors are aware, this is the first study evaluating the use of MHS technology in pregnant women.

Cervix in pregnant women is not the same as non-pregnant ones. Cervix grows in size, remodeling of the surface contour occurs, vascularity increases, and endocervical mucosa is everted [11]. Although immature metaplasia, basal cell hyperplasia, decidualization, and Arias –Stella reaction complicates interpretation of smears, morphologically cervical intraepithelial neoplasia (CIN) in pregnant women is the same as CIN in the non-pregnant [12]. Whether these changes complicate interpretation of MHS screening of pregnant cervix is not known. Therefore the present authors designed this first study of MHS in pregnant women who have very low risk, no history of warts, and/or CIN and normal Pap smear results. The specificity of MHS in the present pregnant population was around 92.9%, while in both DeSantis *et al.* and Twiggs *et al.* studies, it was much lower in between 34-57% [1, 9]. This difference was present even specificities were compared according to different age groups. It might be suggested that due to the better visualization of the transformation zone, MHS might perform better in detecting true negative results, increasing specificity. It might also be speculated that due to better recognition and prominent appearance of cervical dysplasia, sensitivity for detecting CIN may also be increased; but this question is to be answered in another trial. With these results, MHS in pregnancy may have the potential to protect pregnant women from unnecessary colposcopies.

The prevalence of HPV in the present study population was around 7.1%, and this was remarkably lower from the data published by Twiggs *et al.* (prevalence 40%) and by Werner (prevalence 33%) [1, 10]. This very low level of HR-HPV positivity may also be the cause for high specificity. With a better-visualized cervix, which is unaffected by HPV, it might be quite possible for MHS to report true normal as normal; therefore increasing specificity as the present study result. This difference in specificity might also be explained by the fact that HPV infection may be present in lower genital tracts of women who do not have neoplasia as stated in Werner *et al.* [10]. It is also interesting to note that the cases, which were positive in MHS screening, are not the same as cases who had HR-HPV positivity in the present pregnant study group.

The present study's acceptability and satisfaction rates for MHS during the first half of pregnancy are quite high (92%) and compatible with previously published results. In a study by Ferris *et al.*, women supported the use of spectroscopy instead of Pap smear by 81%; but only 30.9% agreed that spectroscopy was less comfortable than Pap smear [3]. The main reason for dissatisfaction in the present study was also the large dimensions of the sight tube (in 85% of the cases) and then fear of vaginal bleeding. On the other hand, women were especially satisfied to have the results readily available right after having the procedure.

The main weakness of the study is that the present authors were unable and/or failed to perform a gold standard diagnostic procedure to diagnose the cervical pathology correctly. Although colposcopy was planned for HR-HPV positive and MHS screening positive cases, none of the pregnant women gave consent for performing the procedure. Therefore the authors had to rely on Pap smear results as gold standard while calculating specificity. Although choosing a very low-risk population for CIN with a low incidence of HR-HPV might have been helpful in reducing the errors while performing the statistics, the present conclusions are also limited by small sample size and preclude us from generalizations.

To conclude, MHS is a novel, non-invasive, rapid, easy-to-perform, and an objective test with an immediate result, suitable for screening purposes in low-risk pregnant women. Exit interviews suggest that women are receptive to MHS screening of their cervix when pregnant if needed. This study extends previous reports demonstrating the feasibility of MHS in the cervical screening of non-pregnant women to pregnant women. It also proposes that MHS may have similar or superior specificity in this group of sub-

jects. The potential role of MHS in this group of women may be further investigated since it might help to scan and diagnose CIN, especially in low-income settings.

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