Evaluation of Transvaginal Colposcopy as a Screening Device for Cervical Cancer among International Physicians

by

Elizabeth Asma

Duke Global Health Institute
Duke University

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Approved:

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Nimmi Ramanujam, Supervisor

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J.S. Malliga

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Lawrence P. Park

_____
John Schmitt

Thesis submitted in partial fulfillment of
the requirements for the degree of
Master of Science in the Duke Global Health Institute
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ABSTRACT

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Abstract

Cervical cancer disproportionately burdens women in low-resource settings, with over 85% of cervical cancer deaths occurring in developing countries due to lack of access to effective, high-quality screening programs that facilitate early detection and treatment. The aim of this study is to evaluate whether the performance of a transvaginal digital colposcope (TVDC) developed at Duke University is equivalent to the more expensive standard-of-care colposcope at identifying precancerous lesions of the cervix. Thirty-five paired cervix images, with confirmed pathologies and blinded by device, were sent electronically to six physicians, at four separate institutions, Duke University Medical Center (Durham, North Carolina, USA), La Liga Peruana de Lucha Contra el Cancer (Lima, Peru), Cancer Institute WIA (Chennai, India), and Kenyatta University (Nairobi, Kenya). Physicians completed a 1-page survey assessing cervix characteristics and overall severity of precancerous lesions for each image. Analysis included percent agreement between devices as well as identifying patterns across misdiagnosed images. The agreement between physicians using each device is 80.1% with kappa of 0.6049. The TVDC performed equivalent to standard-of-care colposcopy at identifying precancerous lesions of the cervix. Implications of these findings have the potential to create increased access to a culturally appropriate screening technology, thus reducing the burden of cervical cancer throughout the developing world.

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List of Abbreviations

ACCP	Alliance for Cervical Cancer Prevention
CIN	Cervical intraepithelial neoplasia
DUMC	Duke University Medical Center
ECC	Endocervical curettage (endocervical scraping)
FDA	Food & Drug Administration
HPV	Human Papilloma Virus
HSIL	High-grade squamous intraepithelial
IARC	International Agency for Research on Cancer
IRB	Institutional Review Board
LED	Light Emitting Diode
LEEP	Loop Electrosurgical Excision Procedure
LSIL	Low-grade squamous intraepithelial
LO2	Leisegang Optik 2 (Colposcope at DUMC)
Pap	Papanicolaou test (Pap smear)
PDF	Portable Document Format
REDCap	Research Electronic Data Capture
TVDC	Transvaginal digital colposcope
USD	United States Dollar
VIA	Visual Inspection with Acetic Acid
VIAM	VIA Inspection with Acetic Acid using Magnification
WHO	World Health Organization

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1. Introduction

Cervical cancer kills 270,000 women each year and more than 85% of these deaths occur in developing countries [2]. In sub-Saharan Africa, 34.8 new cases of cervical cancer are diagnosed per 100,000 women annually, and 22.5 per 100,000 women die from the disease. These figures compare with 6.6 and 2.5 per 100,000 women, respectively, in North America [3]. These drastic differences can be explained by lack of access to effective, high-quality screening programs that facilitate early detection and treatment [4]. The heaviest burden of cervical cancer is disproportionately seen throughout Africa, Central and South America and parts of Asia, as demonstrated by Figure 1, where red indicates countries experiencing high cervical cancer incidence [1].

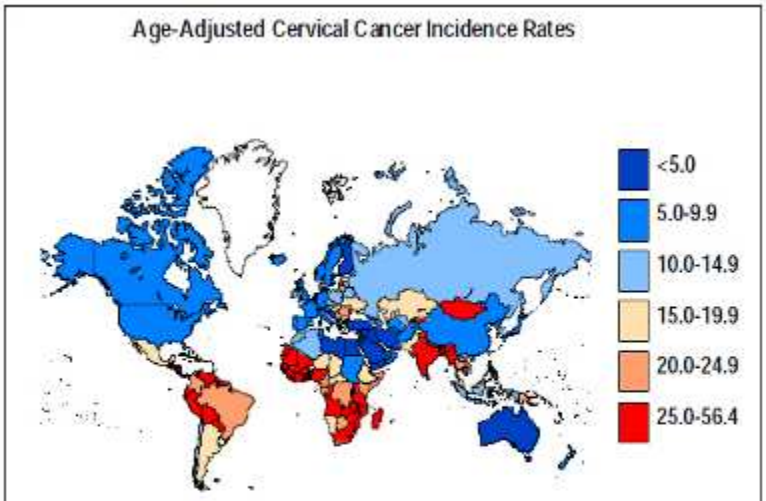


Figure 1: Age-Adjusted Cervical Cancer Incidence Rates per 100,000 (GLOBOCAN 2008)

Precancerous lesions of the cervix are typically asymptomatic, and therefore when preventative screening is not common, cervical cancer is usually only detected at an advanced incurable stage [5]. However, unlike most other types of cancer, cervical cancer is preventable when precancerous lesions are detected and treated; meaning early detection of precancerous lesions is the best strategy to reduce the high incidence and mortality of cervical cancer throughout the developing world.

1.1 Cervical Cancer Screening & Treatment

All current methods to screen for cervical intraepithelial neoplasia (CIN), a precancerous cell growth on the surface of the cervix which can develop into cancer, require a pelvic exam (speculum-based). Cytology (Papanicolaou smear) swabs a sample from the surface of the cervix to test for abnormal cell changes. Pap smears are the standard-of-care for cervical cancer screening in the United States and require trained physicians, laboratory technicians, appropriate infrastructure to support sample processing and a return visit by patients to receive test results. Loss to follow up and lack of laboratory capacity to process samples are primary reasons Pap smears are not a successful screening method for cervical cancer in low-resource settings [6].

The HPV DNA test has become increasing popular to screen for the presence of human papillomavirus (HPV). The current HPV vaccine protects against strains of HPV (Types 6, 11, 16 & 18) which are known to cause over 70% of cervical cancers [7]. A new HPV vaccine just approved by the FDA protects against 9 types of HPV, increasing the

potential of the vaccine to prevent approximately 90% of cervical cancer [8]. However, the vaccine cannot protect against all strains of HPV, so even those who were vaccinated will still need to be screened for cervical cancer as adults [9]. Also vaccination does not protect women who have already been exposed to the virus [6]. HPV DNA testing, similar to Pap smear, cannot succeed in low-resource settings due to lack of laboratory capacity to process samples.

The World Health Organization (WHO) recommends visual inspection with acetic acid (VIA) as the most efficient and effective strategy for detecting cervical cancer precursors in low-resource settings [10] [4]. VIA applies acetic acid to the cervix during a speculum-based pelvic exam, which the clinician then evaluates for presence of any aceto-whitening on the cervix with the naked eye. Studies have demonstrated screening by VIA is a simple, affordable, and sensitive test that can identify precancerous changes of the cervix [6]. During VIA, the cervix is viewed with no magnification, only the naked eye, no laboratory processing is required, the results are immediate and if necessary, treatment can be provided in the same visit [11]. VIA has a high sensitivity but low specificity, leading to overtreatment and since interpretation is subjective, the success of VIA is highly dependent on health worker training.

Cervicography is a screening method in which a non-physician captures an image of the cervix during a VIA exam and then submits the image to a physician for interpretation. This method is considered visual inspection with acetic acid using

magnification (VIAM) because magnification of the cervix image can occur depending on the features of the camera.

Colposcopy is the primary diagnostic method used to visualize CIN following an abnormal Pap smear and also a type of VIAM. Colposcopy involves illuminated magnification of the cervix, which enables providers to identify and locate lesions and if necessary, biopsy abnormal cervical cell growth [12]. During a colposcopy exam, a 3-5% solution of acetic acid (vinegar) is applied to the cervix, causing abnormal cells to turn white (aceto-whiten). Lesion characteristics such as color, opacity, margin shape, blood vessel caliber, and inter-capillary spacing are considered by physicians to derive a clinical diagnosis, however the color and appearance of images vary with colposcope instruments, camera setting and light source [12].

A colposcope is a microscope-like instrument with a powerful light source used to magnify and illuminate cervix epithelium cells and vascular patterns, which enable clinicians to recognize and predict pathology. Colposcopes range in price from $5,000-$20,000 (USD) depending on the following options: range of magnification, type of light source, auto/manual focus, green filter, solid support base and accessory camera or video capture capabilities [13]. Traditional colposcopes are expensive, require specialized training for proficient use and may not be practical in many low-resource settings due to the cost of equipment and training [11].

Several types of biopsies can be used to diagnose cervical precancerous lesions. If the biopsy can remove all the abnormal tissue, it might be the only treatment needed [14]. A standard biopsy removes a small section of the abnormal area from the surface of the cervix. Endocervical curettage (ECC) scrapes into the endocervical canal to remove a tissue sample to test for abnormalities. Loop electrosurgical excision procedure (LEEP) uses a thin, low-voltage electrified wire loop to cut out larger sections of abnormal cells and cold knife cone biopsy uses a surgical scalpel instead of a heated wire to remove tissue.

Once cervical cancer has been diagnosed, there are several treatment options for removal of the cancerous tissue; LEEP, cryotherapy or laser ablation. Cryotherapy, which destroys abnormal tissue by freezing, is the most efficient and effective strategy for treatment of cervical cancer precursors in low-resource settings [4]. Liquid nitrogen is circulated through a probe, which is applied to cancerous tissue and the freezing temperatures destroy the cancer cells. Laser surgery involves a laser which emits intense heat and light at close range to vaporize and burn off abnormal cancerous tissue [14].

1.2 Development of the Transvaginal Digital Colposcope

The Tissue Optical Spectroscopy Lab within the department of Biomedical Engineering at Pratt School of Engineering at Duke University has developed a low cost transvaginal digital colposcope for cervical cancer screening in low resource settings.

The transvaginal digital colposcope (TVDC) combines the benefits of colposcopy and cervicography for use in settings where only VIA was previously possible. The TVDC has a comparable field of view, resolution and contrast compared to a standard digital colposcope. The transvaginal design was inspired by the tampon, putting the camera at a much closer distance to the cervix (30 mm) than a traditional colposcope (300 mm), which provides comparable image quality with decreased cost due to decreased camera megapixel requirements. The TVDC is significantly less expensive (several hundred USD) than the traditional standard-of-care colposcope (several thousand USD). The device is portable, runs on battery power, and digitally captures images. These features enable clinicians in remote environments to send images to physicians for diagnosis.

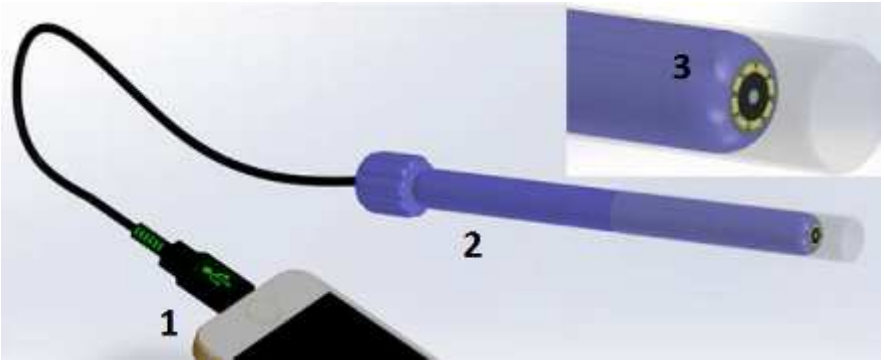


Figure 2: Picture of Transvaginal Digital Colposcope; (1) Digital image capture capabilities, (2) tampon-like introducer, (3) LEDs for illumination

The TVDC is currently undergoing a clinical trial collecting paired cervix images at DUMC under Duke University Medical IRB approved protocol (Durham, North Carolina, USA). This protocol completes standard-of-care colposcopy by capturing cervical images using the Leisegang Optik 2 (LO2) colposcope at DUMC, followed by

image capture using the TVDC of the same patient's cervix. The procedure is then completed with appropriate standard of care biopsy with pathology results interpreted at DUMC. Participants include adult females undergoing routine colposcopy and/or LEEP with paired cervical images ranging from normal cervices to those with low or high-grade precancerous lesions. The purpose of this study is to show functionality of the TVDC as well as analyze the concordance between image pairs, ultimately demonstrating the equivalence of TVDC to standard-of-care digital colposcopy.

1.3 Research Question

The main barriers to cervical cancer screening include lack of accessible and available high quality services, a lack of comfort and privacy in health centers, high cost of services, and anxiety related to waiting for test results [15]. These factors combined are major impediments to creating widespread cervical cancer prevention throughout developing countries, where the incidence and mortality of cervical cancer is highest. Significant concerns about embarrassment and pain due to screening from the speculum, gender of the health care provider, privacy and access to screening centers are some of the main reasons why women do not receive screening for cervical cancer [16]. The TVDC strives to address these issues by delivering a cost-effective cervical cancer screening solution for low resource environments.

This primary research question for this study: **Is transvaginal digital colposcopy equivalent to current standard-of-care colposcopy at identifying precancerous cervical**

abnormalities? A blinded and randomized evaluation of thirty-five (35) image pairs, each taken of the same cervix with a standard-of-care digital colposcope and then the TVDC, were completed by six (6) highly-trained physicians from both developed and developing countries. Technical and clinical qualities of each image were quantitatively assessed to explore whether transvaginal digital colposcopy is equivalent to digital colposcopy at identifying precancerous cervical abnormalities.

2. Methods

Surveys and fully de-identified image panels were sent electronically to six (6) physicians, at four (4) separate institutions. All participating institutions signed a Data Transfer Agreement, approved by data security at Duke University, for the sharing of cervix images collected under Duke University Medical IRB approved protocol Pro00008173. Survey responses were returned via email or through a secure survey link, depending on physician preference. Completed responses were entered twice (for verification purposes) into the data management tool REDCap electronic data capture hosted at Duke University. REDCap (Research Electronic Data Capture) is a secure, web-based application designed to support data capture for research studies by providing an intuitive interface for validated data entry and automated export procedures for data downloads to common statistical packages [17]. The dataset was then exported for statistical analysis using Stata SE 13.0 [18].

2.1 Setting

Paired cervix image collection occurred at DUMC (Durham, North Carolina, USA). These cervix images were then blinded by device and randomized for the purposes of our study. A total of six (6) physicians were surveyed: three (3) from DUMC (Durham, North Carolina, USA), one (1) from La Liga Peruana de Lucha Contra el Cancer (Lima, Peru), one (1) from Cancer Institute (WIA) Chennai (Chennai, India) and one (1) from Kenyatta University (Nairobi, Kenya).

2.2 Participants

All six (6) physician participants were trained in obstetrics and gynecology and are currently practicing obstetrics and gynecology, benign gynecology or gynecologic oncology at their respective hospitals. All six (6) physicians are highly-trained and familiar with identifying cervix abnormalities through visual inspection and/or colposcopy.

2.3 Procedures

Paired cervix images were collected under Duke University Medical IRB approved protocol Pro0000817 at DUMC. This protocol completed standard of care colposcopy by capturing cervical images using the Leisegang Optik 2 (LO2) colposcope at DUMC. This was followed by image capture using the TVDC of the same patient's cervix. The procedure was then completed with appropriate standard-of-care biopsy with pathology results interpreted at DUMC. Participants included adult females undergoing routine colposcopy and/or LEEP with paired cervical images ranging from normal cervices to those with low or high-grade precancerous cervical lesions. All potential participants were introduced to the study by a healthcare professional familiar with them and their medical history and agreed to participation with signed informed consent.

This study focused on the interpretation and evaluation of the collected cervix image pairs. Image pairs were randomized and placed into four (4) PDF documents for

physicians to review one image per page. A total of seventy (70) images were evaluated by each physician (35 randomized image pairs). Four separate PDF documents were created due to email attachment size restrictions. Panel A contained twenty-one (21) images, Panel B contained twenty-one (21) images, Panel C contained fourteen (14) images and Panel D contained fourteen (14) images.

Cervix images were blinded by labeling the upper left hand corner with an image identifier. No other identifying marks were listed on the image except an orientation label locating 12 o'clock. Panels A & B were created from the first batch of images collected during the DUMC protocol whereas panels C & D contained image pairs collected later on during the study. The panel letter did not indicate which device captured the image; for example Panel A contained images captured by both the LO2 and TVDC. The master key was only available to researchers and contained the randomized image identifiers, indicating which images were paired (of the same cervix), the colposcope which captured the image (LO2 or TVDC) and the resulting pathology of any biopsy.

All four image panels (A, B C & D) were shared electronically with physicians. All participating institutions signed a Data Transfer Agreement for the sharing of cervix images collected under the Duke University Medical IRB approved protocol.

One (1) physician participant from DUMC reviewed two (2) side-by-side cervix image panels instead of the blinded four (4) image panels. This physician reviewed the

exact same seventy (70) cervix images; however image pairs were viewed on the same page, with the LO2 image always on the left and the TVDC image always on the right. An individual survey response was completed for each image, however since the evaluations of these images were not blinded, it will be appropriately labeled when this physician's interpretations are included in any sections of analysis. Evaluating images in pairs and un-blinded by device may confound the dataset because it is known image pairs are of the same cervix, and therefore have the same precancerous lesion diagnosis.

Physicians were emailed a one-page fillable PDF survey to complete for each cervix image. The PDF survey had a fixed-layout with drop down menu options, radio button selections or free text response to answer questions (Appendix A). A web-based version of the survey was also available and depending on physician preference, could be completed through a secure online survey link created using REDCap (Appendix B). Both the PDF and web-based version of the survey contained identical questions and formatting however; the method of survey completion was by physician preference. All surveys were returned to researchers via email or the survey link, totaling seventy (70) surveys per physician. The development and contents of this survey are described below in Measures.

Original survey responses returned electronically are stored in a password protected Dropbox folder only available to the research team. The survey responses

submitted through the secure online link are automatically housed in the password protected REDCap database hosted at Duke University.

2.4 Measures

The survey was piloted, reviewed and approved by an experienced physician at DUMC prior to distribution. The survey includes the image identifier code, basic demographic information, technical questions regarding image quality and clinical questions evaluating properties of the cervix. Demographic questions include the physician name, site affiliation and standard-of-care for cervical imaging at their practice. Technical questions include field of view, magnification and focus.

Clinical questions include whether the os and transformation zone were visible as well as a summary of aceto-whitening, lesion margin and vascularization. The os (ostium of uterus), the external orifice of the uterus, when viewed through the vaginal canal, is a small rounded opening near the center of the cervix. The transformation zone is an area of changing cells and the most common place on the cervix for abnormal cells to develop. Aceto-whitening refers to the color change when 3-5% solution of acetic acid (vinegar) is applied to the cervix, highlighting areas of increased cellular protein and nuclear density. Lesion margin refers to shape definition of any abnormal areas highlighted by aceto-whitening. Vascularization refers to the characteristic and size of any visible vessels. Answers to these clinical questions were developed using the modified Reid colposcopic index [19]. The clinical evaluation questions also included the

number and location of any potential lesions as well as the ultimate grading of the cervix, ability to confidently grade, and any overall comments.

The modified Reid colposcopic index (available as Appendix C) is a systematic method for grading the severity of precancerous cervical lesions. The index calculates a score based on the severity of properties observed in the following four categories: lesion margin, color of aceto-whitening, blood vessels and iodine staining [19]. This objective score helps clinicians accurately predict the histology of any visible cervical lesions. The Reid colposcopic index is widely used in colposcopy training by the International Agency for Research on Cancer (IARC) and Alliance for Cervical Cancer Prevention (ACCP).

2.5 Analysis

An electronic database was created using REDCap to manage all survey responses. When surveys were returned via email, they were entered twice for data verification purposes into the REDCap database. Survey responses submitted via the online link were automatically entered into the database upon submission.

The original dataset exported from REDCap contained individual records per randomized image; therefore Stata was used to create a new database by matching records by image identifiers to the master key. This new database contained survey responses per image pair, aiding in easier analysis by colposcope device and pathology.

It is important to note that among all sections of analysis, the survey responses from the single physician who viewed side-by-side image panels (instead of the blinded image panels) will be appropriately labeled when included in results.

Physicians were de-identified by name; however a participant demographic summary table was created to identify physicians by site affiliation, location and current standard of care for cervical imaging at their practice.

Image pairs were assessed for accuracy based on the response to "What is your grading of this cervix?" between corresponding TVDC and LO2 images. Responses were classified as either clinically normal or abnormal (Table 1).

Table 1: Classification of image results into Normal & Abnormal

	Normal	Abnormal
What is your grading of this cervix? Evaluated by each physician for every image [Normal, CIN 0, CIN 1, CIN 2, CIN 3, Cancer]	Normal CIN 0	CIN 1 CIN 2 CIN 3 Cancer
Pathology Results interpreted at DUMC [Normal, LSIL\CIN1, HSIL\CIN2, HSIL\CIN3]	Normal	LSIL\CIN 1 HSIL\CIN 2 HSIL\CIN 3

Pathology results were also compared separately to each image by each colposcope device and for that comparison; cervix grades and pathologies were displayed in 3 classifications (Table 2).

Table 2: Classification of pathology, image results into Normal, LSIL & HSIL

	Normal	LSIL	HSIL
What is your grading of this cervix? Evaluated by each physician for each image [Normal, CIN 0, CIN 1, CIN 2, CIN 3, Cancer]	Normal CIN 0	CIN 1	CIN 2 CIN 3 Cancer
Pathology Results interpreted at DUMC [Normal, LSIL\CIN1, HSIL\CIN2, HSIL\CIN3]	Normal	LSIL\CIN 1	HSIL\CIN 2 HSIL\CIN 3

Contingency tables and kappa statistics were used to assess agreement between colposcopy and pathology results. Percent (%) agreement values were calculated by dividing the number of correctly classified image pairs by the total number of evaluated image pairs. Agreement between paired images was analyzed across devices (TVDC & LO2) and then each colposcope device separately compared to pathology results.

The kappa statistic (K) accounts for the agreement, which may occur by chance if images were randomly assigned grades. The kappa statistic ranges from -1 to 1, and values approaching 1 indicate stronger agreement not caused by random effects. Appendix D includes a chart to help interpret the kappa statistic [20]. The kappa statistics is calculated using the equation: K = (observed proportion of agreement – proportion of agreement expected simply by chance)/ (1- proportion of agreement expected simply by chance).

False positive and false negative rates were also calculated to quantify misdiagnosis rates. False positives were defined as the images which were identified as abnormal, when the standard of care classified the image or biopsy as normal. False

negatives were defined as the images which were identified as normal, when the standard of care classified the image or biopsy as abnormal. For 3 x 3 contingency tables, LSIL and HSIL categories were combined into "abnormal" for the calculation of these rates. The false negative rate (FNR) was calculated as the number of false negatives (FN) divided by the total false negatives and true positives (TP), FNR = FN/ (FN+TP). The false positive rate (FPR) was calculated as the number of false positives (FP) divided by the total of false positives and true negatives (TN), FPR = FP/ (FP+TN). For 3 x 3 contingency tables, LSIL and HSIL categories were combined into "abnormal" for the calculation of false negative and false positive rates.

Statistical comparisons were performed for three scenarios: 1) each TVDC image was matched with its corresponding LO2 image, 2) each TVDC image was matched with its corresponding pathology and 3) each LO2 image was matched with its corresponding pathology. An overall summary table was compiled from the six (6) physician responses. Breakdown of agreement levels and kappa statistics was also summarized by each individual physician for each of these three scenarios.

De-identified cervix image pairs were evaluated for differences between matched TVDC and LO2 images for the variables; field of view, magnification, focus, os, transformation zone, aceto-whitening, opacity, color, shiny, lesion margin, vascularization and confidence in ability to diagnose precancerous cervical lesions. These differences were summarized along with representative image pairs for the cases

where pathology, TVDC and LO2 all reached same conclusion, for when pathology and LO2 reached same conclusion but TVDC was misdiagnosed, and for when pathology and TVDC reached same conclusion but LO2 was misdiagnosed. Normal, LSIL and HSIL pathologies were included for each scenario.

A univariate logistic regression model was fit with the response variable concordant image diagnosis between LO2 and TVDC with the predictor variable aceto-whitening.

3. Results

3.1 Participant Demographics

Table 3: Demographics of Physician Participants

Demographics of Physician Participants			
Physician	**Site Affiliation**	**Location**	**Standard of Care**
1	Duke University Medical Center (DUMC)	North Carolina, USA	VIAM with Colposcopy
2*	Duke University Medical Center (DUMC)	North Carolina, USA	VIAM with Colposcopy
3	Duke University Medical Center (DUMC)	North Carolina, USA	VIAM with Colposcopy
4	La Liga Peruana de Lucha Contra el Cáncer	Lima, Peru	VIAM with Colposcopy
5	Cancer Institute (WIA) Chennai	Chennai, India	VIAM with Colposcopy
6	Kenyatta University	Nairobi, Kenya	VIA (naked eye)

*Images viewed by physician were not blinded by colposcope device

All six (6) physicians were trained in obstetrics and gynecology and are currently practicing obstetrics and gynecology, benign gynecology or gynecologic oncology at their respective hospitals. All six (6) physicians are highly trained and familiar with identifying cervix abnormalities through visual inspection and/or colposcopy. Five (5) physicians are most familiar with visual inspection with acetic acid using magnification (VIAM) through colposcopy. One (1) physician is most familiar with visual inspection with acetic acid (VIA) through naked eye evaluation. By nature of study design, it is important to note viewing image captures from colposcopy could also be defined as cervicography, because a record of the cervix image exists for later interpretation. Type and brand of colposcopes vary across institutions but the standard of care colposcope device currently used at DUMC is the Leisegang Optik 2.

3.2 Physician Accuracy Statistics

The following tables [4 & 5] summarize physician accuracy statistics when comparing cervical lesion diagnosis among paired images. Table 5 provides a summary excluding the physician who viewed images side-by-side. An image pair was deemed accurate if the blinded image evaluation resulted in a matched response to the question "What is your grading of this cervix?" for both the TVDC and LO2 paired images. Possible lesion grades were classified as normal or abnormal based on the classification divisions (Table 1). Each colposcope image was also compared to the reported pathology for that image pair and pathologies were classified as Normal, LSIL or HSIL (Table 2). Agreement levels and kappa statistics across the six (6) physicians are summarized by bar graphs as Figures 3, 4 & 5.

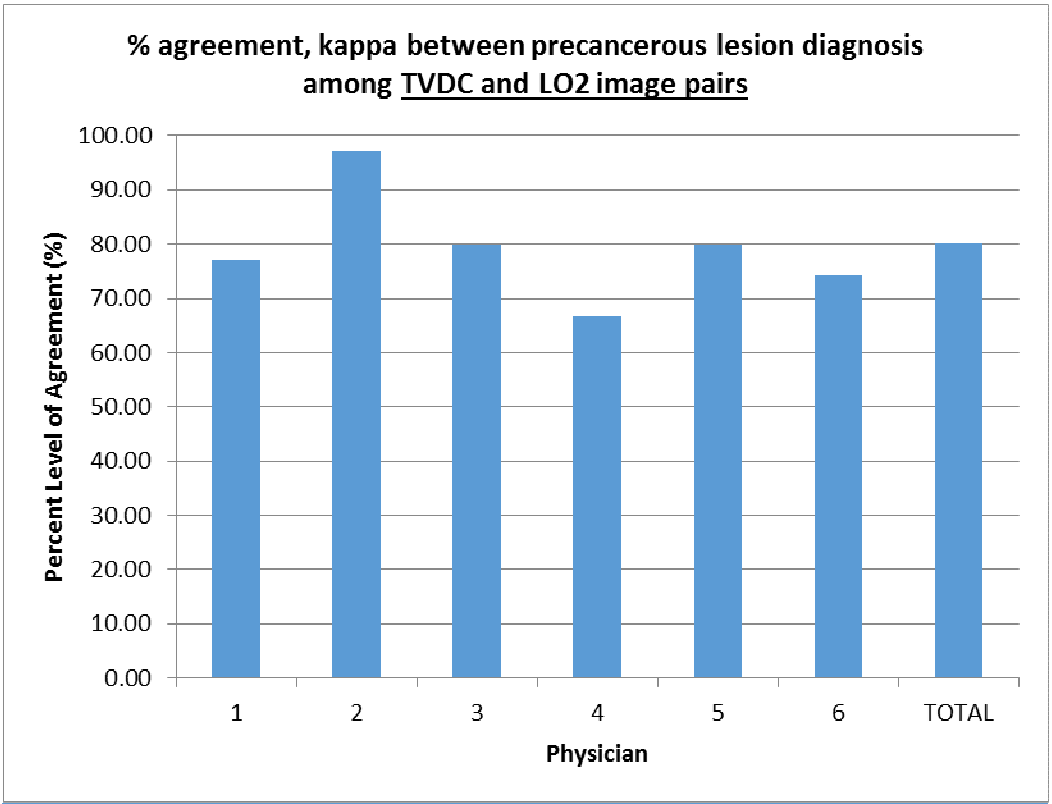
Table 4: All physician survey responses classifying severity of precancerous lesions

All physician survey responses classifying severity of precancerous lesions									
		Leisegang Optik 2*				**False Negative Rate**	**False Positive Rate**	**%Agreement**	**Kappa**
TVDC	Classification	Normal		Abnormal	**Total**				
	Normal	**74**		29	103	**25.9%**	**11.9%**	**80.10%**	**0.6049**
	Abnormal	10		**83**	93				
	Total	84		112	196				
		Pathology*				**False Negative Rate**	**False Positive Rate**	**% Agreement**	**Kappa**
TVDC	Classification	Normal	LSIL	HSIL	**Total**				
	Normal	**60**	23	20	103	**43.9%**	**38.8%**	**45.41%**	**0.1175**
	LSIL	22	**10**	10	42				
	HSIL	16	16	**19**	51				
	Total	98	49	49	196				
		Pathology*				**False Negative Rate**	**False Positive Rate**	**% Agreement**	**Kappa**
LO2	Classification	Normal	LSIL	HSIL	**Total**				
	Normal	**50**	20	14	84	**34.7%**	**49.0%**	**45.92%**	**0.1587**
	LSIL	30	**11**	6	47				
	HSIL	18	18	**29**	65				
	Total	98	49	49	196				
*Considered gold standard									

Table 5: Physicians survey responses classifying severity of precancerous lesions (excluding side-by-side analysis)

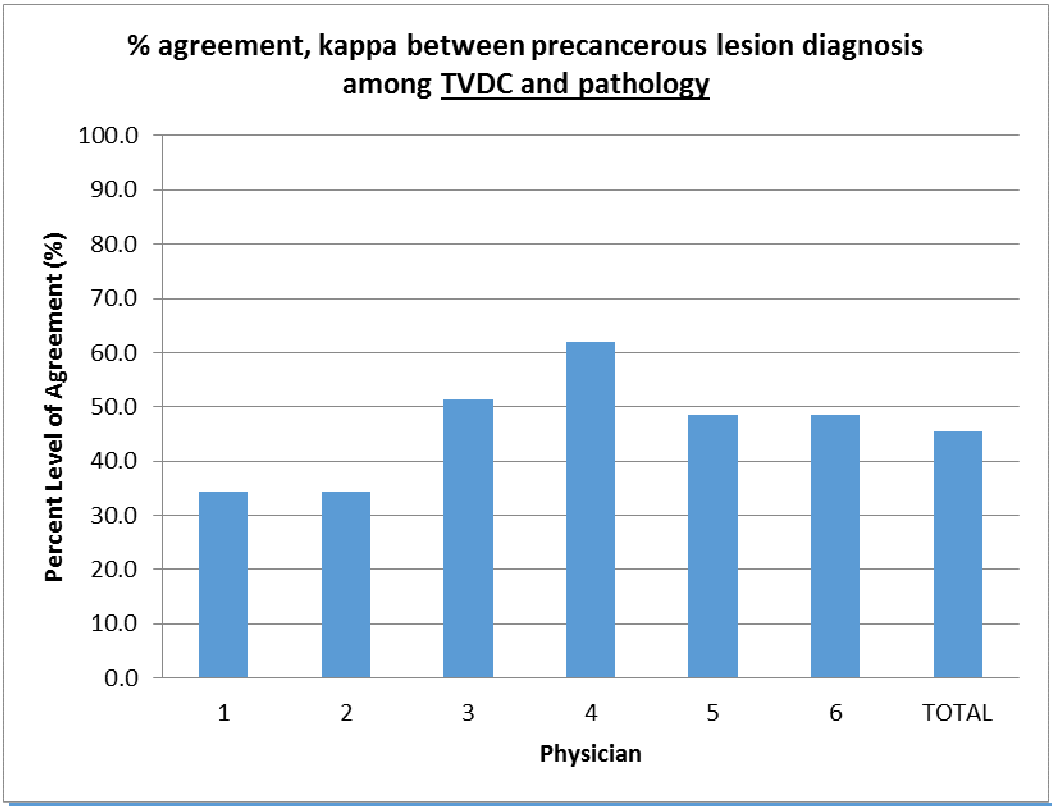
Physician survey responses classifying severity of precancerous lesions (excluding side-by-side analysis)									
		Leisegang Optik 2*				**False Negative Rate**	**False Positive Rate**	**%Agreement**	**Kappa**
TVDC	Classification	Normal		Abnormal	**Total**				
	Normal	**68**		28	96	**33.7%**	**12.8%**	**76.40%**	**0.5308**
	Abnormal	10		**55**	65				
	Total	78		83	161				
		Pathology*				**False Negative Rate**	**False Positive Rate**	**% Agreement**	**Kappa**
TVDC	Classification	Normal	LSIL	HSIL	**Total**				
	Normal	**56**	22	18	96	**50.0%**	**30.9%**	**47.83%**	**0.1300**
	LSIL	12	**7**	8	27				
	HSIL	13	11	**14**	38				
	Total	81	40	40	161				
		Pathology*				**False Negative Rate**	**False Positive Rate**	**% Agreement**	**Kappa**
LO2	Classification	Normal	LSIL	HSIL	**Total**				
	Normal	**46**	19	13	78	**40.0%**	**43.2%**	**47.83%**	**0.1694**
	LSIL	19	**8**	4	31				
	HSIL	16	13	**23**	52				
	Total	81	40	40	161				

*Considered gold standard



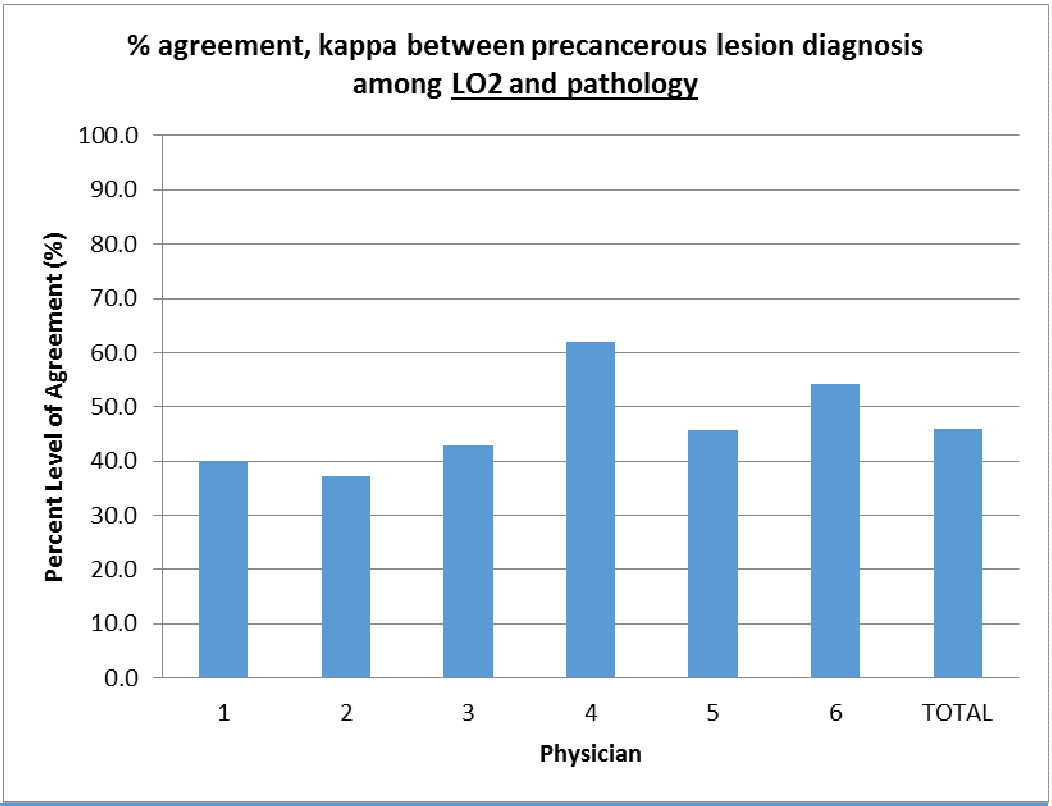
	1	2	3	4	5	6	Total
Kappa (K)	0.0728	0.9057	0.4948	0.3467	0.5826	0.4615	0.6049

Figure 3: Inter-observer variability represented by percent agreement level and kappa statistic by individual physician when comparing TVDC image diagnosis with matched LO2 image diagnosis



	1	2	3	4	5	6	Total
Kappa (K)	0.0661	0.0574	0.1524	0.2432	0.1026	0.1322	0.1175

Figure 4: Inter-observer variability represented by percent agreement level and kappa statistic by individual physician when comparing TVDC image diagnosis with pathology interpretation



	1	2	3	4	5	6	Total
Kappa (K)	0.1629	0.1067	0.0141	0.3708	0.1227	0.2533	0.1587

Figure 5: Inter-observer variability represented by percent agreement level and kappa statistic by individual physician when comparing LO2 image diagnosis with pathology interpretation

3.3 Technical and Clinical Survey Responses

Tables 7 through 9 summarize technical and clinical responses and their differences across colposcope devices. Table 7 displays de-identified representative images for Normal, LSIL and HSIL pathologies where both the TVDC and LO2 image evaluation matched pathology. Table 8 displays representative images among the three possible pathologies for cases where pathology and LO2 image were consistent but TVDC images were not concordant. Table 9 similarly displays example cervix images where pathology and TVDC diagnosis were the same but LO2 images were not concordant. Table 10 contains a univariable logistic regression model which assesses presence of aceto-whitening as a predictor variable for whether the TVDC image will match the corresponding LO2 image diagnosis.

Table 6: Technical and clinical differences for paired images (pathology, LO2 and TVDC same diagnosis)

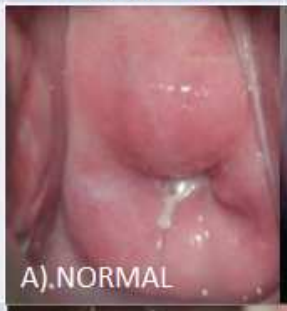
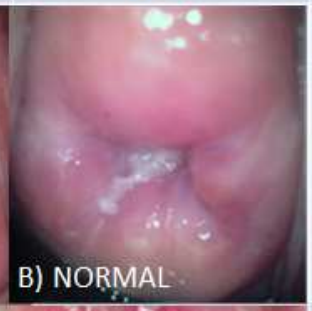
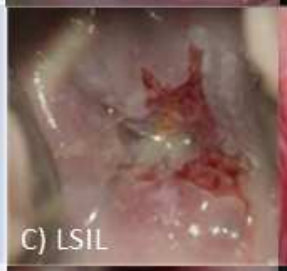
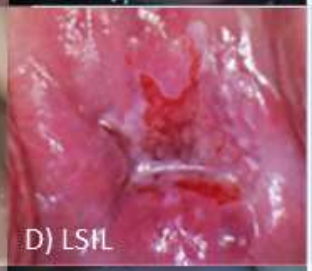
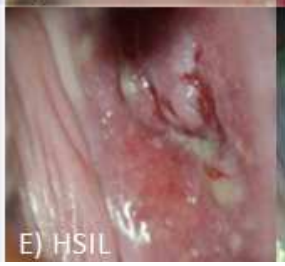
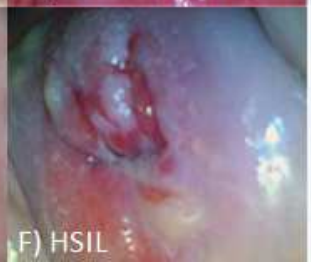
Example images where pathology, LO2 & TVDC all reached same diagnosis			
	Leisegang Optik 2	**TVDC**	**Technical/Clinical Differences**
NORMAL	 A) NORMAL	 B) NORMAL	**Surface shine:** TVDC shiny, LO2 dull **Vascularization:** TVDC saw vessels, LO2 did NOT see vessels
LSIL	 C) LSIL	 D) LSIL	**Opacity:** TVDC were semi-opaque, LO2 were transparent
HSIL	 E) HSIL	 F) HSIL	**Focus:** TVDC were NOT in focus **Aceto-whitening**: TVDC did NOT see aceto-whitening **Opacity:** TVDC were semi-opaque, LO2 were fully-opaque **Surface shine:** TVDC shiny, LO2 dull **Lesion Margin:** TVDC flat/indistinct, LO2 smooth **Confidence**: TVDC NOT confident in diagnosis

Table 7: Technical and clinical differences for paired images (pathology and LO2 same diagnosis)

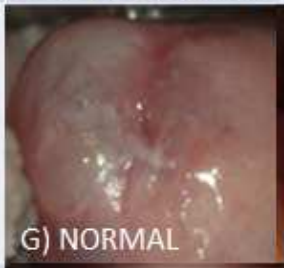
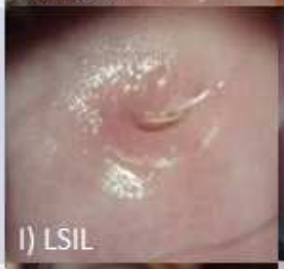
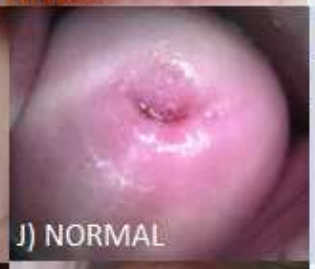
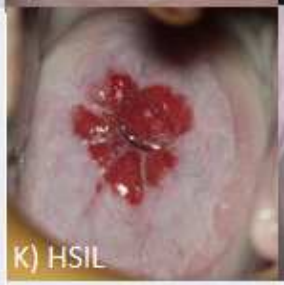
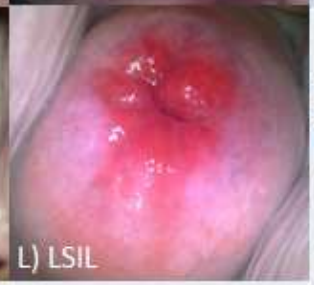
Example images where pathology, LO2 reached same diagnosis, but TVDC did not			
	Leisegang Optik 2	**TVDC**	**Technical/Clinical Differences**
NORMAL	 G) NORMAL	 H) HSIL	**Aceto-whitening:** TVDC saw aceto-whitening **Shiny:** TVDC shiny, LO2 dull **Confidence:** TVDC NOT confident in diagnosis
LSIL	 I) LSIL	 J) NORMAL	**Focus:** TVDC were NOT in focus, LO2 were in focus **Transformation Zone:** TVDC did NOT see transformation zone **Aceto-whitening:** TVDC did NOT see aceto-whitening
HSIL	 K) HSIL	 L) LSIL	**Focus:** TVDC were NOT in focus, LO2 were in focus **Aceto-whitening:** TVDC did NOT see aceto-whitening **Shiny:** TVDC shiny, LO2 dull **Transformation Zone:** TVDC did NOT see transformation zone **Opacity:** TVDC was transparent, LO2 was fully-opaque **Color:** TVDC pure white, LO2 oyster grey **Surface shine:** TVDC shiny, LO2 dull **Lesion margin:** TVDC flat/indistinct, LO2 rolled/peeled **Vascularization:** TVDC did NOT see vessels

Table 8: Technical and clinical differences for paired images (pathology and TVDC same diagnosis)

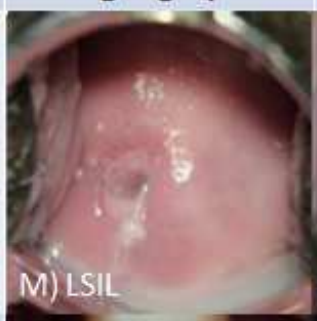
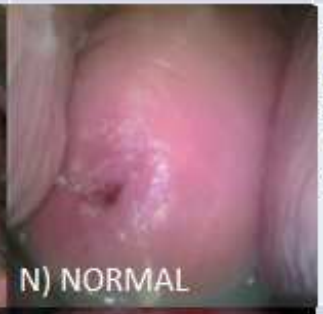
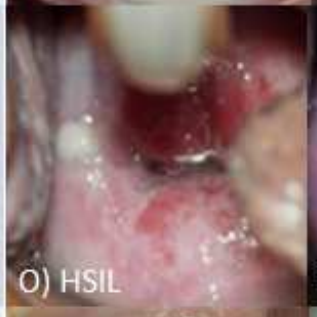
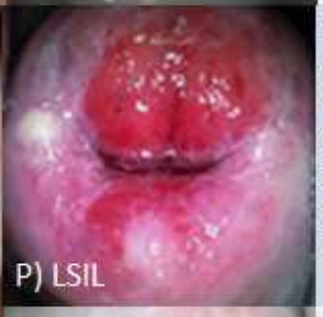
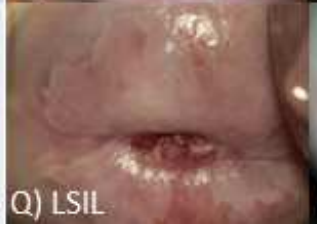
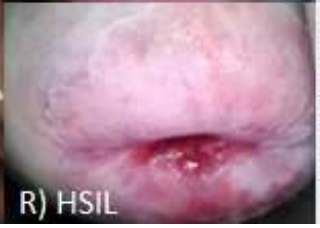
Example images where pathology, TVDC reached same diagnosis, but LO2 did not			
	Leisegang Optik 2	**TVDC**	**Technical/Clinical Differences**
NORMAL			**Focus:** TVDC NOT in focus **Aceto-whitening:** TVDC did NOT see aceto-whitening **Lesion margin:** TVDC indistinct, LO2 smooth **Vascularization:** TVDC did NOT see vessels **Confidence:** TVDC were NOT confident in diagnosis
LSIL			**Focus:** TVDC NOT in focus **Aceto-whitening:** LO2 did NOT see aceto-whitening **Opacity:** TVDC transparent, LO2 fully opaque **Color:** TVDC pure white, LO2 grey white **Surface shine:** TVDC shiny, LO2 dull **Confidence:** TVDC were NOT confident in diagnosis **Note:** LO2 image had obstruction of view
HSIL			**Focus:** LO2 NOT in focus **Os:** LO2 did NOT capture os **Transformation zone:** TVDC did NOT see transformation zone **Opacity:** TVDC semi-opaque, LO2 fully opaque **Surface shine:** TVDC were dull, LO2 were shiny

Table 9: Univariate logistic regression model, aceto-whitening as predictor variable for image concordance between TVDC and LO2

Univariate logistic regression model, aceto-whitening as predictor variable for image concordance between TVDC and LO2			
Variable	**Response**	**OR [95%CI]**	**P-value**
Aceto-whitening	No	1	---
	Yes	2.33 [1.06, 5.13]	0.035

The odds that TVDC and LO2 images will match when aceto-whitening is seen in TVDC images are 2.33 that of TVDC and LO2 images will match when aceto-whitening is not seen in TVDC images. Univariable logistic regression models were also explored with the following as predictor variables; standard of care, field of view, magnification, focus, os, transformation zone, opacity, color, surface shine, and lesion margin, however no other variable significantly ($p<0.05$) affected the outcome of image concordance between TVDC and LO2.

4. Discussion

The purpose of this study was to explore whether transvaginal digital colposcopy is equivalent to current standard-of-care colposcopy at identifying precancerous cervical abnormalities. Analysis of a blinded image evaluation among trained physicians showed 80.1% agreement level between matched TVDC images with a kappa value 0.6049. Both devices had equivalent poor agreement and kappa results when compared to pathology. Inter-observer agreement was fairly consistent across physicians, regardless of current standard-of-care or hospital location. The most important clinical characteristic to correctly detect precancerous cervical lesions in TVDC images was presence of aceto-whitening. Other characteristics influencing a correct TVDC diagnostic outcome include surface shine, image focus and confidence in diagnosis. These results summarize the performance of transvaginal digital colposcopy relative to digital colposcopy as well as identify factors influencing misdiagnosed images.

Comparing performance between the imaging methods, TVDC images identified precancerous cervical lesions just as well as LO2 standard-of-care digital colposcopy. The percent agreement among physicians using each device is 80.1% with a kappa statistic of 0.6049 (Table 4). Kappa higher than 0.6 indicates substantial agreement between colposcope devices (Appendix D). When removing the physician who viewed side-by-side images from the analysis, TVDC images reached the same pathology

diagnosis 76.40% of the time to the matched LO2 image diagnosis, with a Kappa = 0.5308 (Table 5).

Responses across physicians were fairly consistent with agreement between TVDC & LO2 image evaluations ranging between 66.7% - 97.2% (Figure 3). The accuracy of one physician was very high (97.2%) because images viewed were not blinded by device. The next highest physician accuracy among TVDC vs LO2 was 80.0%.

Colposcopy (both LO2 and TVDC) agreed considerably less frequently with pathologic assessment than with each other. When comparing TVDC to LO2, agreement was 80.1% (K = 0.6049), but TVDC images compared to pathology had only 45.41% agreement (K = 0.1175) and LO2 images compared to pathology had only 45.92% agreement (K = 0.1587) (Table 4). A kappa value less than 0.2 indicates slight agreement (Appendix D). Responses across physicians were fairly consistent, displaying similar levels of variance across both the TVDC (Figure 4) and LO2 (Figure 5) comparisons to pathology.

It is interesting to note that both devices (TVDC and LO2) appear to have similar disconcordance when predicting histology from images. This is consistent with previous studies exploring the strength of agreement between colposcopy and histology. A population-based study with colposcopy and biopsy results for 2466 women, found the overall strength of agreement between colposcopy and histology was poor (K = 0.17) [21]. Also, there is often disagreement between physicians regarding image evaluation

and diagnosis. Another study of 939 cervical images, found that pairs of colposcopists only agreed on diagnosis for 56.8% of images [22].

When looking at the performance of the LO2 images compared to pathology, the false positive rate is always higher than the false positive rate of the TVDC images compared to pathology (Tables 4 & 5). This means LO2 images are more often classified as abnormal, when pathology results indicate the suspected lesion was actually normal. This over-cautious approach could lead to overtreatment (taking biopsy when not necessary), and is of particular concern in low-resource settings where additional testing could unnecessarily burden the healthcare system. While the false positive rate among TVDC ability to match pathology (38.8%) was lower than LO2 to pathology (49.0%), this could be an artifact of TVDC images failing to see aceto-whitening when aceto-whitening was seen on their corresponding LO2 image pairs.

The largest discrepancy between image pairs that were misdiagnosed was presence of aceto-whitening (Tables 7 through 10). A major reason TVDC images did not match pathology or LO2 image diagnosis was that physicians did not see aceto-whitening on the TVDC image. Other clinical and technical characteristics would be almost identical between matched images but almost always "Can you see aceto-whitening?" was answered "No" in TVDC images which were misdiagnosed. A univariate logistic regression model between TVDC and LO2 concluded the odds that TVDC and LO2 images will match when aceto-whitening is seen in TVDC images are

2.33 that of when TVDC and LO2 images will match when aceto-whitening is not seen in TVDC images. The odds ratio (OR) is a measure of association between an exposure and an outcome. Although not the primary focus of analysis, a lack of aceto-whitening was also noticed in LO2 images when LO2 images were misdiagnosed in relation to pathology during representative image comparison.

Other differences seen from the representative image comparison included: focus, surface shine, lesion margin, confidence, and vascularization, however only aceto-whitening was statistically significant ($p< 0.05$ as seen by the logistic regression). Multiple univariable logistic regression models were run separately with standard of care, field of view, magnification, focus, os, transformation zone, opacity, color, surface shine, and lesion margin as predictor variables for response of image concordance between TVDC and LO2, however none significantly ($p< 0.05$) affected the outcome of image concordance between TVDC and LO2.

Although not statistically significant, focus, surface shine, lesion margin, confidence, and vascularization variables all saw differences across paired images. Lack of focus in TVDC images was sometimes a concern, where the corresponding LO2 image was in focus. Surface shine was another concern, often TVDC images were identified as shiny, whereas their matching LO2 images were identified as dull. In a few TVDC images, lesion margin was identified as "unknown" when the corresponding LO2 image was able to detect a smooth lesion. The physician's confidence in their ability to

diagnosis the potential lesion also saw differences across devices. When a misdiagnosis occurred, the TVDC image was often identified as not providing enough information to make a confident diagnosis. The ability to detect presence of vessels in images also affected the outcome. Misdiagnosed TVDC images did not see vascularization when their matching LO2 images did detect vascularization.

There were a few instances when TVDC images correctly identified the lesion according to pathology but the LO2 image did not. Reasons for these differences include focus or an obstructed view. Lack of image focus in LO2 images would lead to a misdiagnosis. The other reason for misdiagnosis among LO2 images was obstructed view, either by the speculum or inability to see os or transformation zone from image angle.

4.1 Recommendations

The largest difference seen among paired TVDC and LO2 images that were misdiagnosed was lack of aceto-whitening identified in TVDC images. A reason for this difference might be due to a systematic error during data collection. The protocol at DUMC applies acetic acid solution to the cervix, takes the image with the LO2 and then captures the image with the TVDC without reapplication of acetic acid. Time from application of acetic acid to image capture with the TVDC could range roughly between five to ten minutes, whereas image capture with the LO2 would occur approximately two to five minutes after application of acetic acid on the cervix. This time difference

could decrease the visual whitening of potential abnormal cervical lesions during TVDC image capture. It has been shown that acetic acid-induced whitening of high-grade squamous intraepithelial lesions maximizes around three to five minutes after application of acetic acid to the cervix, but that after five minutes the effects of whitening begin to drop off [23]. The official guide for colposcopy examination from the IARC states acetic acid should be reapplied every 2-3 minutes throughout the examination because the effects of aceto-whitening may begin to fade after one minute [19].

Recommendations to improve visualization of aceto-whitening in TVDC images include reapplication of acetic acid prior to TVDC image capture. Another improvement could be to slightly decrease the brightness of the LEDs during TVDC image capture because sometimes the shiny glare can cover up the presence of any aceto-whitening. It was noticed that misdiagnosed TVDC images were frequently labeled as shiny when corresponding LO2 images were labeled as dull. Capturing green filter TVDC images could also improve diagnostic capabilities because lack of identifying vascularization was listed as a difference among images that were misdiagnosed. Using green filtered light creates more contrast in tissue vascularization, allowing for the visualization of vessel caliber and spacing patterns.

Future avenues for research include developing a mobile health application for community health workers. The TVDC interfaces with an android or tablet, which could provide a digital resource database and virtual modules to aid in the training of

frontline health care workers performing cervical cancer screenings using the TVDC. Lack of confidence in the ability to correctly diagnosis TVDC images was much higher among misdiagnosed images than seen in the corresponding LO2 images, and a training program with the ability to provide feedback could help improve confidence and ability to correctly identify precancerous lesions on TVDC images.

4.2 Study strengths and limitations

Study strengths include that cervix image evaluations were blinded by colposcope device. Images were cropped such that no indicators (ie speculum) were visible to identify which colposcope device captured the image. Image randomization ensured individual images within image pairs were dispersed throughout the image panels.

A study design concern might include static colposcope images were evaluated by physicians when colposcopy is normally a dynamic evaluation, however past studies have explored the difference and found lack of significant difference between static and dynamic image evaluation when evaluating and interpreting colposcopy [24].

Practical limitations of this study include follow-up with physicians regarding completion of the surveys. Thirty five (35) pairs for a total of seventy (70) images was a fairly large commitment for our very busy physicians. Possible future improvements could include embedding cervix photos into a survey link, so the image and questions were all available on one screen and could also allow for saving and returning, allowing

physicians to evaluate images at their own schedule. This would ease ability to complete evaluations, and could automatically send email reminders until completion, which would decrease the amount of follow-up time required by the research team to evaluate the progress of each physician.

Other limitations of the study include physician interpretations of survey questions were fairly subjective, especially questions about opacity, color, shine, lesion margin and vascularization. Future improvements could include providing sample images for reference, although that creates bias in that physicians may rely more on provided sample image comparisons than their own experience and training. Another limitation includes non-uniformity of screens which were used to review images. The images may appear different depending on what screen displayed the image. Future studies might consider calibrating monitors and collecting image evaluations off the same device, although that provides many logistical difficulties.

4.3 Implications for policy and practice

Conclusions from this study show the performance of transvaginal digital colposcopy is equivalent to that of standard-of-care digital colposcopy in identifying precancerous lesions of the cervix. This has significant policy implications, creating the potential for widespread implementation of the TVDC. This technology could help alleviate the disproportionate burden of cervical cancer in developing countries by

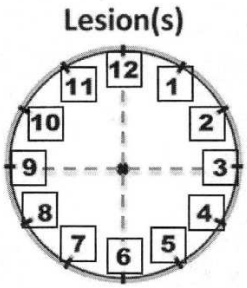
providing a low-cost, portable and culturally sensitive option to identify precancerous cervical lesions before they become fatal.

5. Conclusion

The TVDC performed equivalent to standard-of-care digital colposcopy in its ability to identify precancerous cervical lesions during a blinded image evaluation among six (6) physicians. Areas of improvement include application of acetic acid immediately preceding TVDC image capture to increase the ability to view aceto-whitening in cervix images experiencing abnormal cell growth. The unique design of the transvaginal digital colposcope, combining portability, with the low-cost ability to digitally capture images, creates an appropriate technology for countries lacking the tools and resources to effectively support cervical cancer screening programs. Wide-spread implementation of this technology has the potential to increase access to effective, high-quality screening programs and facilitate early detection of precancerous lesions in low-resource setting. Implications of these findings have the potential to create increased access to a culturally appropriate screening technology, thus reducing the burden of cervical cancer throughout the developing world.

Appendix A

Please complete the following **Clinical evaluation of Cervical Imaging** after using your standard of care and transvaginal digital colposcope (TVDC) /for VIA/VIAM for cervical cancer screening.

	Response
Reviewer ID #: (Please Select from List)	
Site Affiliation: (Please Select from List)	
Image Letter Code: (Please Select from List)	A☐ B☐ C☐ D☐ E☐ F☐
Image ID #: (Please Select from List)	1
What is the standard of care cervical imaging of your practice?	☐VIA (naked eye) ☐VIAM with Cervicography ☐VIAM with Colposcopy
Field of view - How many of the four quadrants of the cervix can you see in a single view?	☐1 ☐2 ☐3 ☐4
Magnification – Does a single image/visual provide adequate magnification?	☐yes ☐no
Is the image/visual in focus?	☐yes ☐no
Can you see the os?	☐yes ☐no
If present, can you see the transformation zone?	☐yes ☐no ☐NA
Can you see aceto-whitening?	☐yes ☐no ◯ NA
If there are aceto-whitened sites, what is the opacity?	☐transparent ☐semi-opaque ◯ NA ☐fully opaque
If there are aceto-whitened sites, what is the color?	☐pure white ☐grey white ◯ NA ☐oyster grey
Surface shine – is the cervical surface shiny?	☐intense shiny ☐shiny ◯ NA ☐dull
Lesion margin – describe the lesion margin	☐Flat/indistinct/feathered/scalloped ☐Smooth ◯ NA ☐Rolled/peeled
Vascularization – describe, if present	☐yes vessels, fine punctuation/mosaicism ☐no vessels ◯ NA ☐yes vessels, coarse punctuation/mosaicism
How many lesions are visible?	Fill in blank (___)
Please check the location of lesion(s) on the shown clock face. The cervix, viewed as a circle, is divided into 12 segments denoted by 1-12 and separated into four quadrants denoted by dashed blue line for spatial reference.	
What is your grading of this cervix?	☑Normal ☐CIN0 ☐CIN1 ☐CIN2 ☐CIN3 ◯ Can
Did the image provide enough information to make a confident diagnosis? If not, why?	☑yes ☐no Why? _____

Appendix B

This version of the survey was available online and accessed using the following link: https://redcap.dtmi.duke.edu/redcap/surveys/?s=PTkmN3ennH . The questions and format are identical to the PDF version of the survey in Appendix A.

Appendix C [19]

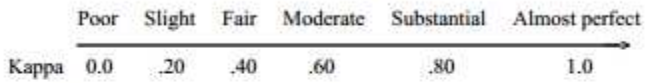
Colposcopic signs	Zero point	One point	Two points
Colour	Low-intensity acetowhitening (not completely opaque); indistinct acetowhitening; transparent or translucent acetowhitening Acetowhitening beyond the margin of the transformation zonePure snow-white colour with intense surface shine	Intermediate shade - grey/white colour and shiny surface (most lesions should be scored in this category)	Dull, opaque, oyster white; grey
Lesion margin and surface configuration	Microcondylomatous or micropapillary contour[1] Flat lesions with indistinct marginsFeathered or finely scalloped margins Angular, jagged lesions[3] Satellite lesions beyond the margin of the transformation zone	Regular-shaped, symmetrical lesions with smooth, straight outlines	Rolled, peeling edges[2] Internal demarcations between areas of differing colposcopic appearance-a central area of high-grade change and peripheral area of low-grade change
Vessels	Fine/uniform-calibre vessels[4]- closely and uniformly placed Poorly formed patterns of fine punctation and/or mosaic Vessels beyond the margin of the transformation zone Fine vessels within microcondylomatous or micropapillary lesions[6]	Absent vessels	Well defined coarse punctation or mosaic, sharply demarcated[5] - and randomly and widely placed
Iodine staining	Positive iodine uptake giving mahogany-brown color Negative uptake of insignificant lesion, i.e., yellow staining by a lesion scoring three points or less on the first three criteria Areas beyond the margin of the transformation zone, conspicuous on colposcopy, evident as iodine-negative areas (such areas are frequently due to parakeratosis)[7]	Partial iodine uptake - variegated, speckled appearance	Negative iodine uptake of significant lesion, i.e., yellow staining by a lesion already scoring four points or more on the first three criteria

Colposcopic prediction of histologic diagnosis using the Reid Colposcopic Index (RCI)

RCI (overall score)	Histology
0 - 2	Likely to be CIN 1
3 - 4	Overlapping lesion: likely to be CIN 1 or CIN 2
5 - 8	Likely to be CIN 2-3

Appendix D [20]

Interpretation of Kappa



Kappa	Agreement
< 0	Less than chance agreement
0.01–0.20	Slight agreement
0.21– 0.40	Fair agreement
0.41–0.60	Moderate agreement
0.61–0.80	Substantial agreement
0.81–0.99	Almost perfect agreement

References

1. Singh, G.K., R.E. Azuine, and M. Siahpush, *Global inequalities in cervical cancer incidence and mortality are linked to deprivation, low socioeconomic status, and human development.* Int J MCH AIDS, 2012. **1**(1): p. 17.

2. GLOBOCAN (IARC), *Cancer Fact Sheets: Cervical Cancer.* World Health Organization International Agency for Research on Cancer, 2012.

3. Cancer, I.A.f.R.o. and W.H. Organization, *Latest world cancer statistics*, in *Global cancer burden rises to 14.1 million new cases in 2012: marked increase in breast and cervix cancers must be addressed*. 2013.

4. Sherris, J., et al., *Evidence-based, alternative cervical cancer screening approaches in low-resource settings.* Int Perspect Sex Reprod Health, 2009. **35**(3): p. 147-54.

5. Almonte, M., et al., *Cervical screening by visual inspection, HPV testing, liquid-based and conventional cytology in Amazonian Peru.* Int J Cancer, 2007. **121**(4): p. 796-802.

6. Paul, P., et al., *Screen-and-treat approach to cervical cancer prevention using visual inspection with acetic acid and cryotherapy: experiences, perceptions, and beliefs from demonstration projects in Peru, Uganda, and Vietnam.* Oncologist, 2013. **18 Suppl**: p. 6-12.

7. Castellsagué, X., *Natural history and epidemiology of HPV infection and cervical cancer.* Gynecologic oncology, 2008. **110**(3): p. S4-S7.

8. Release, F.N., *FDA approves Guardasil 9 for prevention of certain cancers caused by five additional types of HPV*. 2014.

9. PATH, *Cervical cancer screening and treatment in low-resource settings: practical experience from PATH*, in *Cervical Cancer Prevention: Practical Experience Series*. 2013, PATH.

10. Organization, W.H., *Prevention of cervical cancer through screening using visual inspection with acetic acid (VIA) and treatment with cryotherapy. A demonstration project in six African countries: Malawi, Madagascar, Nigeria, Uganda, the United Republic of Tanzania, and Zambia.* 2012.

11. Prevention, A.f.C.C., *Planning and Implementing Cervical Cancer Prevention and Control Programs: A Manual for Managers.* 2004.

12. Li, W., M. Soto-Thompson, and U. Gustafsson, *A new image calibration system in digital colposcopy.* Optics Express, 2006. **14**(26): p. 12887-12901.

13. Ferris, D.G., W.A. Willner, and J.J. Ho, *Colposcopes: A Critical Review.* The Journal of Family Practice, 1991. **33**(5): p. 506-515.

14. Society, A.C., *Cervical Cancer*, A.C. Society, Editor. 2014. p. 57.

15. Agurto, I., et al., *Perceived barriers and benefits to cervical cancer screening in Latin America.* Prev Med, 2004. **39**(1): p. 91-8.

16. Paz-Soldan, V.A., et al., *Low knowledge of cervical cancer and cervical pap smears among women in Peru, and their ideas of how this could be improved.* Int Q Community Health Educ, 2010. **31**(3): p. 245-63.

17. Harris, P.A., et al., *Research electronic data capture (REDCap)—A metadata-driven methodology and workflow process for providing translational research informatics support.* Journal of Biomedical Informatics. **42**(2): p. 377-381.

18. StataCorp, *Stata Statistical Software: Release 13.* 2013. **College Station, TX: StataCorp LP**.

19. Sellors, J.W. and R. Sankaranarayanan, *Colposcopy and treatment of cervical intraepithelial neoplasia: a beginner's manual*. 2003: Diamond Pocket Books (P) Ltd.

20. Viera, A.J. and J.M. Garrett, *Understanding Interobserver Agreement: The Kappa Statistic.* Family Medicine, 2005. **37**(5): p. 360-3.

21. Ghosh, I., et al., *Study of accuracy of colposcopy in VIA and HPV detection-based cervical cancer screening program.* Australian and New Zealand Journal of Obstetrics and Gynaecology, 2014. **54**(6): p. 570-575.

22. Jeronimo, J., et al., *Interobserver Agreement in the Evaluation of Digitized Cervical Images.* Obstetrics & Gynecology, 2007. **110**(4): p. 833-840.

23. Pogue, B.W., et al., *Analysis of acetic acid-induced whitening of high-grade squamous intraepithelial lesions.* Journal of Biomedical Optics, 2001. **6**(4): p. 397-403.

24. Massad, L.S., et al., *The accuracy of colposcopic grading for detection of high grade cervical intraepithelial neoplasia.* Journal of lower genital tract disease, 2009. **13**(3): p. 137.