Mobile High-Resolution Microendoscopy (mHRME) for the Detection of Cervical Dysplasia in El Salvador

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1.1 Objectives:

1. To evaluate the performance of mobile high-resolution microendoscopy (mHRME) compared with HPV DNA testing alone and HPV testing followed by VIA in San Salvador, El Salvador.
2. To evaluate the feasibility and usability of the mHRME system in a rural setting in El Salvador.
3. To assess the performance of new technologies for HPV DNA and oncoprotein and compare their performance in patients with and without cervical precancer and cancer.

Introduction:
Cervical cancer is still the most common type of cancer and the second leading cause of cancer-related deaths among women in developing countries, due to the lack of appropriate screening programs. There are economic, political, and logistic barriers for the implementation of mass vaccination programs, especially in Low-and-Middle-Income-Countries (LMIC). Furthermore, currently available vaccines do not bring protection against all types of high-risk HPV strains and are not effective in curing already existing infections. Therefore, cervical cancer screenings will continue to be necessary for years to come. One of the ways to screen for cervical cancer is through visual inspection with acetic acid (VIA). The VIA's sensitivity in the detection of cervical dysplasia and cancer is similar to that of a colposcopy, but with less specificity. The low specificity of this test can translate into false positives, leading to overtreatment of benign conditions, which not only increases the programs' cost, but causes unnecessary concern for the patients. These reasons highlight the need to have diagnostic methods with increased specificity that allow the identification of patients requiring intervention. This is particularly relevant in developing countries, where colposcopy-guided biopsies and histopathological testing are not readily available. The mHRME imaging system is a low-cost method that allows for the evaluation of epithelial cells *in situ,* thus eliminating the need for cervical biopsies. This study intends to validate the performance characteristics of HRME as a diagnostic tool.

2.0 Background and Significance

Early Detection:
For a while, cervical cancer was considered the leading cause of cancer-related deaths among women in the United States. However, its prevalence and death rate have decreased by approximately 70% over the last 40 years. This reduction is greatly linked to the implementation of the Papanicolaou test in 1941, which promoted increased efforts in the detection of precancerous lesions (1). Cervical cancer is still the most common type of cancer and the second leading cause of cancer-related deaths among women in developing countries, due to the lack of appropriate screening programs (2). Today, it is a known fact that almost all cases of cervical cancer are caused by high-risk strains of the Human Papilloma Virus (HPV) (3). Despite the development of preventive vaccines, in use since 2006 (4, 5), administration rates for said vaccine remain very low, with less than 50% of minors being vaccinated in the United States of America (6, 7). There are economic, political, and logistic barriers to the implementation of mass vaccination programs, especially in Low-and-Middle-Income-Countries (LMIC). Furthermore, currently available vaccines do not bring protection against all types of high-risk HPV strains and are not effective in curing already existing infections. Therefore, cervical cancer screenings will continue to be necessary for years to come.

Cervical Cancer Screening and Prevention in LMICs:

Current approaches when it comes to cervical cancer screening in developing countries include cervical cytology and HPV detection testing. Patients with abnormal results undergo a colposcopy during which biopsies of the areas that are considered abnormal are taken. If significant lesions with the potential of developing into cancer are found, a cryotherapy or a cone biopsy (LEEP) is performed. These methods are effective; however, they are also very costly and require appropriate infrastructure and qualified personnel. Furthermore, the patient has to be seen in three different occasions, including one visit to be informed about the test results. This is why alternative solutions are highly needed, especially in developing countries. A visual inspection with acetic acid (VIA) is an option. During this test, the acetic acid is applied to the cervical epithelium, and upon the identification of any acetowhite areas, indicative of precancerous lesions, a cryotherapy or a cone biopsy (LEEP) can be performed during the same visit ("See and Treat"). The VIA's sensitivity in the detection of cervical dysplasia and cancer is similar to that of a colposcopy but with less specificity (8-12). The low specificity of this test can translate into false positives, leading to overtreatment of benign conditions, which not only increases the programs' cost, but causes unnecessary concern for the patients. A similar "See and Treat" protocol in higher-income regions has been implemented in the Netherlands, where patients undergo a cone biopsy (LEEP) immediately after a colposcopy has identified high-risk lesions. Even in cases where a colposcopy was performed, an overtreatment rate of 18.1% was observed (13). This highlights the need to have diagnostic methods with increased specificity that allow the identification of patients requiring intervention. This is particularly relevant in developing countries, where colposcopy-guided biopsies and histopathological testing are not readily available.

HRME Imaging:

The mHRME imaging system is a low-cost method that allows for the evaluation of epithelial cells in situ, thus eliminating the need for cervical biopsies. A topical contrast marker called Proflavine is applied to the cervix in the same way that the acetic acid is applied during VIA and colposcopy procedures. The tip of a small optic fiber probe is placed on the cervix and the presence of proflavine-stained fluorescent cells is transmitted to the HRME system and to a computer monitor or tablet (Fig. 1). Some of the morphological features usually assessed by pathologists, such as nuclear clustering, pleomorphism, and the nuclear-cytoplasmic (N:C) ratio can be evaluated live, in real time. An imaging analysis software is then used to quantify the nuclear morphology parameters and calculate the N:C ratio. This approach allows for real-time detection of high-grade precancerous lesions without the need for a biopsy. Moreover, the lesions can be treated during that same visit either with cryotherapy or with a cone biopsy (LEEP), if necessary. The HRME system is portable, resistant, and battery-operated. Its cost is around $2,500 if the device is used with a computer.

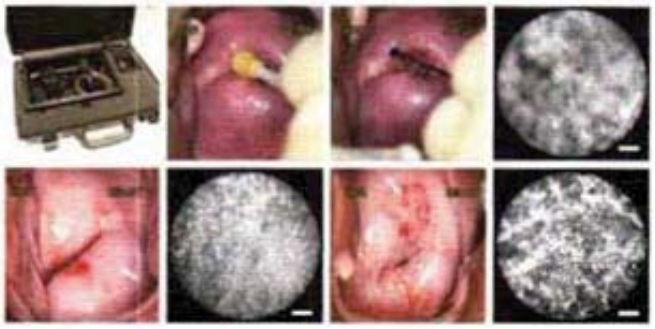

Fig. 1 (a) HRME system; (b) Proflavine application; (c) Fiber optic probe, (d) Real-time, high-resolution image on a computer; (c) Colposcopic image of a lesion located at 5:00 o'clock; (d) Resulting HRME-created image. Histopathological diagnosis was normal, consistent with the HRME image, showing small homogeneously-distributed nuclei; (g) Colposcopic image of a lesion located at 1:00 o'clock; (f) Resulting HRME-created image. The diagnosis for this site was CIN 3, consistent with the HRME image, showing

A more inexpensive system that uses a smartphone (mHRME) has been developed recently (Fig. 2). The phone model used was a Samsung Galaxy Note 3, with a 2.3 GHz processor, 3 GB RAM, and a 12-MP camera. An optic adaptor is placed on the phone. The system measures 150mm x 60mm x 80mm. The 4.9-micron resolution is appropriate for the maximum resolution allowed by the fiber optic used. The optic performance of the mHRME is equivalent to that of the HRME system. Production costs of mHRME systems are estimated to be under $500 if around 10,000 units are manufactured.

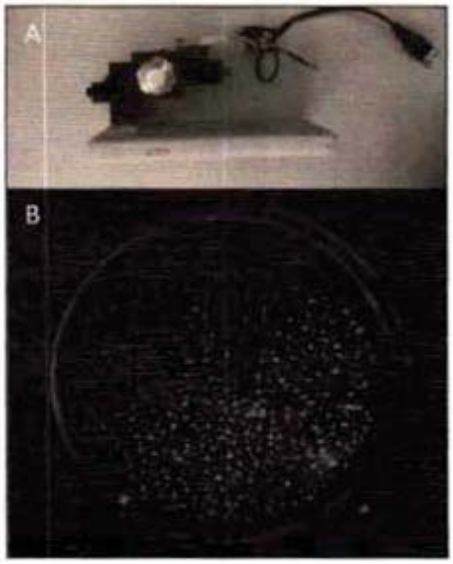

Fig. 2 (a) mHRME system; (b) mHRME-created cervical tissue image.

HPV E7 Assay:

The presence of HPV DNA is associated with an increased risk of cervical cancer. Nevertheless, said presence alone does not imply the existence of a persistent or precancerous lesion. In fact, most HPV infections are cleared by the immune system. In low-grade lesions, the HPV DNA exists as an extrachromosomal episome. However, in high-grade and cancer lesions, the viral genomes are often found in the chromosome of the host cell (14), resulting in a non-regulated expression of the E6 and E7 viral genes, prompting the lesion to progress to a cancer. The HPV DNA test cannot discriminate between episomal and integrated DNA. Therefore, this method has high sensitivity and negative predictive value, but low specificity and positive predictive value for cervical cancer. However, the overexpression of viral oncoproteins (ex. E6 and E7) is often associated with genetic integration and represents a high risk of disease progression (15). HPV DNA testing renders positive results for many patients who do not require treatment. Screening for other HPV biomarkers that have been linked to neoplastic progression could improve care management for infected women, as it would allow the identification of those patients with a higher risk of having a precancerous or cancerous lesion.

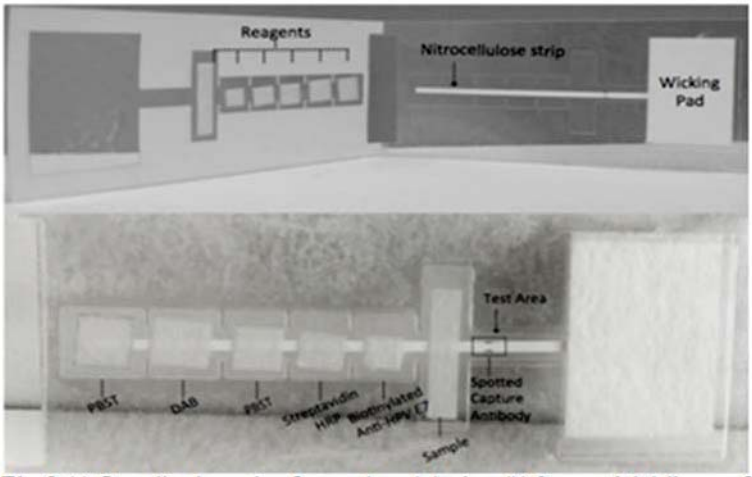

Fig. 3 (a) Overall schematic of paper-based device; (b) Sequential delivery of E7 sandwich ELISA reagents to a test area. For the HPV DNA test, the overall schematic is designed to deliver HPV RNA:DNA hybrid sandwich ELISA reagents to the test area.

Recently, two ELISA tests reported the presence of E6 proteins in cervical cytology (16, 17). The presence of this protein has been more closely correlated to the presence of CIN II than the results obtained with the HPV DNA test. An epidemiologic study to identify the presence of HPV E6 oncoprotein, types 16, 18, and 45 (OncoE6™, Arbor Vita Corporation, Fremont, CA, USA), showed that the detection of E6 has a high

specificity (<2% positive), resulting in a higher positive predictive value (30-35%) of precancerous and cancerous lesions, in comparison to a positive predictive value of <10% using VIA, HC2, and careHPV (18). This method's sensitivity was about 50%, partly because it only screens for three out of all the virus strains responsible for 75% of all cervical cancer cases. Despite these results proving that these tests are efficient at detecting the presence of HPV oncoproteins and that these proteins are specific to cancerous and precancerous lesions, its use at health care centers is still not ideal, since they require specific infrastructure (ex. microtiter plates) (16) and several properly-timed wash and incubation periods (17).

HPV DNA assay

The HPV DNA test has been broadly used in cervical screening protocols due to its high sensitivity and negative predictive value. Recent studies show that HPV DNA screening is more effective than the use of a cervical cytology test or VIA tests (R Sankaranarayanan et al, 2009), with the number of advance-stage cervical cancer cases and cervical cancer deaths dropping down to 50%. Unfortunately, the HPV DNA tests that are commercially available are expensive and require a large infrastructure. The *Digene* HC2 HPV DNA test has a cost of $40-$60 per test, takes over 4.5 hours to perform, and must be done at a laboratory equipped with an agitator and a plate reader (which cost around $500 and $20,000, respectively) (PE Gravitt et al, 2011). While it is not commercially available yet, the HPV DNA test, known as careHPV, has a projected cost of $5 per test. Despite the great improvement cost-wise when compared to the *Digene* HC2 test, the price of each test remains high for many low-income locations. Moreover, it takes about 2.5 hours to perform the careHPV test, in which 90 samples are processed simultaneously; the test also requires an agitator and a plate reader.

To address these limitations, we have developed an HPV DNA test with a lateral flow method that can be produced for less than $2 per test and does not require expensive equipment. Our test uses an RNA hybridization probe, complementary to an HPV DNA sample, and will allow the screening of RNA hybrids: DNA through a sandwich-ELISA assay using the same two-dimension paper technology described above. The test does not require electricity; it relies on capillary reactions and it is disposable. Additionally, it can be performed on a one by one basis, without the need to gather different samples. Our test uses specimens collected from previously attained Papanicolaou tests using a swab, which have been preserved in a bottle. To perform the test, a drop of a denaturizing agent will be added to the bottle to lyse the cells and for DNA denaturation purposes. Then, the RNA hybridization probes will hybridize the denatured DNA and the bottle contents will be transferred to the sample board. An operator will simply rehydrate the dried-up reactors with water and fold the device in half to start the flow. The sample will flow through the testing area, followed by the necessary colorimetric reactors. If HPV DNA is present in the patient's sample, there will be a colorimetric change on the test site itself, which is visible without the need of any additional equipment. Our lateral flow DNA test will allow for the fast and accessible detection of HPV DNA at the health care center. In the following pages, we will assess the performance of this test and if said performance will improve when combined with the HRME.

To address these limitations, we have developed a low-cost ELISA test to screen for the presence of the E7 protein, which can be implemented with a simple, one-step procedure. A new generation, bi-dimensional ELISA method with a lateral flow network lies the foundations for multi-step testing with minimal operator intervention. With our technology, we have developed a nitrocellulose-based platform which uses capillarity to sequentially bring the ELISA reactor to an area where there is no need for the operator's interaction once the test has started. (Fig.3) Due to the absorption properties of nitrocellulose, the test does not require electricity. This disposable test is produced using dried reactors in disposable fiberglass patches. To perform the test, the operator simply adds the patient's cervical swab and water to the dry patches, and then folds the device in half to start the test. Both the sample and the colorimetric reactors flow to the reaction area at the same time. If HPV E7 is present in the patient's sample, there will be a color change that is easily visible

without the need for additional equipment. This testing device will allow for the detection of HPV E7 at health care centers, thus reducing the number of patients that fail to continue their follow-up visits. The performance of this test will be evaluated observing if its performance in combination with HRME imaging testing renders better results.

3.0 Preliminary Data

A pilot study that evaluated HRME testing was recently carried out with 174 female participants in a rural area in China (19). All patients received HPV testing, VIA, colposcopy and HRME imaging. Out of the 69 women who showed abnormalities in the colposcopy, only 12 (17%) were diagnosed with a high-grade lesion after the biopsy. The HRME imaging system correctly classified as "abnormal" all 12 areas (100%) having a high-grade lesion, and it correctly classified 38 out of the 57 remaining samples (67%) as "normal". Moreover, when the patients were stratified according to a positive high-risk HPV DNA detection test, the HRME system correctly identified 100% of patients with CIN 2 or higher. Out of the 30 patients with a positive high-risk HPV DNA detection test with no histological evidence of the disease, only 6 patients (20%) were misidentified as "abnormal" using the HRME imaging system. These preliminary findings suggest that the HRME has an improved specificity when compared to VIA and colposcopy testing, potentially leading to a more accurate screening of patients needing treatment, as part of a "See and Treat" protocol. The study results also suggest that the HRME system can be an effective initial diagnostic tool after a positive cervical cytology or an HPV test. Recently, our collaborators completed a pilot study at the Barretos Cancer Hospital (BCH) to assess the feasibility of using HRME in Brazil (MDACC Protocol 2011-0396). All patients underwent a colposcopy as the standard of care and all acetowhite lesions were recorded. HRME images were obtained for each lesion observed by colposcopy. Abnormal areas were biopsied and assessed by two independent pathologists, and the results were compared to the HRME findings. A total of 79 acetowhite lesions were identified among 46 patients, all of which were biopsied. Adequate images were obtained using the HRME for 59 of these lesions (75%). Biopsy results showed normal tissue (n=9, 15%), CIN 1 (n=16, 27%), CIN 2 (n=12, 20%), CIN 3 (n=19, 32%) and invasive tumors (n=3, 5%). The HRME's positivity percentages were 22% for normal, 31% for CIN 1, 83% for CIN 2, 95% for CIN 3, and 100% for cancer. As for the HRME's sensitivity and specificity rates for CIN 2+ verified by biopsy (n=34), the former was 89% and the latter 77%. These results suggest that the HRME system can be a precise, low-cost alternative to colposcopies and guided cervical biopsies for the diagnosis of cervical dysplasia in low income locations, where the availability of colposcopy and pathology services is low.

This research proposal will be developed based on the findings of the two aforementioned studies, in an attempt to validate the performance characteristics of HRME testing as a diagnostic tool, following an abnormal screening, in a prospective study in El Salvador. The study will be carried out at two different sites: Basic Health International and the ICES.

Duration of the Study:
This study will last 5 years, starting in September 2015.

Projected Number of Research Subjects:
A total of 3,000 women will take part in this study.
The sample size is based on the following premise: our goal is to determine whether the addition of the mHRME to VIA improves the specificity without significantly reducing the sensitivity (non-inferiority trial) in this screening population. During our preliminary work (19), we found that the use of HRME did not reduce the VIA's sensitivity; however, we saw a considerable improvement in specificity (VIA + HRME = 82% vs. VIA alone = 64%) when compared to the histopathology gold standard. The following sample size was estimated using the PASS software (24): a sample size of 45 CIN 2+ cases diagnosed by VIA reaches at least 80% power (84%) to ensure a non-inferior proportion (mHRME sensitivity for CIN 2+ diagnosed by VIA) (P0) of 0.7500, using a one-sided binomial test for non-inferiority. The target significance level is 0.0500. The actual significance level attained by this test is 0.0446. These results assume that the actual proportion (sensitivity) (PI) is 0.9000. If the mHRME sensitivity for CIN 2+ diagnosed by VIA is over 90%, as observed in the preliminary HRME study, the power will be greater. Assuming that 1) VIA's actual sensitivity for all CIN 2+ cases is at least 75% (25); and 2) that the total prevalence of CIN 2+ in El Salvador is 2%, a total sample of 3,000 women will need to be screened using VIA. Finally, assuming that 10% of women have a positive VIA (n = 300, 255 VIA+ without CIN 2+), a 2% reduction among women with VIA+ (2% improvement in specificity) would be statistically significant (ex., 98% positive, 95% confidence interval = 95%-99%).

4.0 Eligibility Criteria

Inclusion Criteria**:**
1. Women who are between the ages of 30 and 49 years of age
2. All women must have a negative urine or serum pregnancy test prior to any study procedure (within 7 days)
3. Intact cervix (patients who have undergone previous LEEP, cone and/or cryotherapy are not eligible)
4. No history of invasive cervical cancer
5. Able and willing to provide informed consent

Exclusion Criteria:
1. Women < 30 years of age or over 49 years of age
2. Women who have undergone a hysterectomy with removal of the cervix
3. Women who have had a previous LEEP, Cold knife cone and/or cryotherapy
4. Women who are pregnant or breastfeeding
5. Women with a history of invasive cervical cancer
6. Unable or unwilling to provide informed consent

5.0 Research Plan and Methods

This is a prospective cohort study.
Eligibility is open to women 30-49 years old, who are not pregnant, have an intact cervix and no history of cervical cancer.

Visit 1:
Patients will enroll at the screening clinic of the *Instituto del Cáncer de El Salvador* (El Salvador Cancer Institute, ICES). An informed consent will be obtained. They will be given a urine pregnancy test; if

negative, two cervical samples will be collected. The first sample will be used to screen for HPV DNA. New technologies to screen for oncoproteins and HPV will be applied to the second sample. This sample will be stored at a temperature of at least -20°C until it is used. 3-5% acetic acid will also be applied to the cervix to perform the VIA. The health care provider will record their impression: positive VIA, (in which case they will describe the lesion area), or negative. Images of the cervix will be taken and any personally identifiable information removed; these will also be stored in the REDCap database.

Visit 2:
All participants will return for their results within three to four months. All women with a positive VIA or HPV test will undergo additional evaluation. In addition, 10% of women in the double negative group (VIA-/HPV-) will be randomly selected for evaluation. This evaluation includes: Urine pregnancy test, VIA, colposcopy, Lugol's solution application, and a HRME. 3-5% acetic acid will also be applied to the cervix to perform the VIA. The health care provider will record their impression for each of the abnormal lesions (positive or negative). Then, the physician will perform the colposcopy and record their impression for each of the abnormal lesions (positive or negative). Proflavine and Lugol's solution (2-5%) will be applied to these lesions, and once again the health care provider will record their impression for each of the abnormal lesions. Cervical images will be taken. Then, 0.01% proflavine will be topically applied to the cervix for 1 minute. Images will be obtained using the mHRME system of a visually normal site and then of all lesions identified by VIA, colposcopy, and/or Lugol's solution. If no lesions are identified by VIA, colposcopy, and/or Lugol's solution, mHRME images will be taken of each quadrant. Samples of abnormal areas identified by VIA, colposcopy, Lugol's solution, and/or mHRME will be collected for biopsy and ECC purposes. If the evaluation shows no abnormalities, a random sample will be collected from the squamocolumnar junction and an ECC will be performed (only if the patient is HPV positive). This diagnostic protocol will be used to prevent the introduction of bias from knowing the screening results. It will also avoid potential bias from auto-correlating the visual methods (colposcopy, VIA, Lugol's solution), which could falsely increase the clinical performance (22, 23). Two expert pathologists will review and classify the samples. All research results will be unbeknownst to them. They will use the following classification system: normal, CIN 1, CIN 2, CIN 3, or cancer, per standard criteria. Any discrepancies will be resolved through a new review, until consensus is reached. Women with CIN 2+ will undergo an excision or cryotherapy, according to the treatment standard. Patients diagnosed with invasive cancer will be referred to gynecology/oncology. The sensitivity and positive predictive value will be calculated for each patient with CIN 2 or a more severe diagnosis (CIN 2+), both for VIA alone, and for VIA followed by the mHRME. The results will be compared with those of the biopsy, which is the histopathology gold standard. The sensitivity and positive predictive value for the HPV DNA alone will also be calculated and compared to an HPV test with VIA/mHRME triage, as well as to the histopathological analysis. These results will enable us to compare the total number of women that could have received adequate and inadequate treatment based on four of the most predominant clinical scenarios: VIA alone, VIA/mHRME triage, HPV test alone, and HPV test integrated with VIA/mHRME triage.

7.0 Potential Side Effects

There are no known risks associated with the mHRME system used in this study. However, there is a remote potential for a severe allergic reaction to proflavine, the contrast dye used to obtain the images. Proflavine can be flammable at high temperatures. Proflavine is the active ingredient in acriflavine and has been used without reported side effects in fluorescent gastrointestinal imaging techniques, according to European, Asian, and Australian literature. It is also used as an antibacterial agent. In neonatal care, a triple dye consisting of a combination of proflavine hemisulfate and crystal violet is routinely used as a topical antibacterial agent on the newborn's umbilical cord (28), with a recent review of this practice which

classified the agent's toxicity as rare (29).
The proflavine concentration proposed for our study is lower than that of commercial products, 0.11% (Kerr Triple Dye, Vista Pharm). The solution amount required for diagnostic imaging is usually not greater than the amount used in neonatal care (0.65 ml per swab). Some research studies in humans using confocal laser scanning microscopy for gastrointestinal cancer patients currently use topical acriflavine with a concentration of 0.05% (30). We have conducted pilot studies using proflavine and the HRME system in 174 women in China (19) and 59 patients in Brazil (MDACC Protocol 2011-0396). No allergic reactions or side effects have been reported by any of the participants.

8.0 Data Collection:

Study data will be collected and managed with REDCap (Research Electronics Data Capture), a data management tool used by MD Anderson [13]. REDCap (www.project-redcap.org) is a secure network tool with controlled access designed for data collection in research studies. The tool provides the following features: 1) a data-input interface; 2) information auditing capabilities to identify cases of mishandling of information; 3) automatic data conversion to statistical analysis packages; 4) import capabilities from external resources. When dealing with multi-center studies, REDCap uses access groups to ensure that personnel from one institution cannot access the data from other facilities. REDCap (https://redcap.mdanderson.org) operates within a secure server at the MD Anderson Cancer Center's Department of Research Information & Technology Services. REDCap's data system has been evaluated by MD Anderson's Information Security Office to ensure that it is compliant with HIPAA, Texas 202-203 Administrative Codes, The University of Texas; Regulation 165, Federal Regulations mentioned in 21CFR, Part 11, and UTMDACC Institutional Regulation #ADM0335. The only people with access to the study's information will be the study chair and research personnel. All confidential information will be removed from the database once it is transferred from REDCap for processing purposes. All dates for each patient will be randomly replaced by a number between 0 and 364, maintaining the proper distance interval between each date.

The following is the specific data to be collected:

- Patient's age
- Ethnicity
- Parity
- History of dysplasia
- VIA and colposcopy findings
- mHRME findings
- Final histopathological findings
- Performed treatment (LEEP, cold knife cone biopsy, cryotherapy)

9.0 Patient Confidentiality

All pathology specimens, evaluations forms, reports and other patient records will be identified in a manner designed to maintain patient confidentiality. Clinical information will not be released without the written permission of the patient or the patient's guardian, except as necessary for monitoring by

regulatory authorities, or the IRB.

The investigators and all employees and co-workers involved with this study shall not disclose or use for any purpose, other than performance of the study, any data, records or other unpublished, confidential information disclosed to those individuals for the purpose of the study. No patient identifiers will be used when analyzing the data or reporting the results.

For this protocol we will only capture adverse events that are determined by the PI to be definitely related to the proflavine and/or HRME imaging. Adverse events and serious adverse events related to the standard of care colposcopy and cervical biopsies will not be captured as part of the protocol data.

Potential Benefits:
There will be no direct benefits for the participants of this study. Their entire screening, treatment, and follow-up process will be the same as if the patient did not participate in this study. The main benefit will be contributing to the research of a prevention mechanism of cervical cancer in other women.

Treatment Alternatives:
Patients will be given the option to receive appropriate treatment if their results come back positive. Women with CIN 2+ will undergo an excision or cryotherapy, according to the treatment standard. Patients diagnosed with invasive cancer will be referred to gynecology/oncology. Participation in this study will not impact the type of treatment given to a patient.

Study Payments:
Participants will receive no payment for taking part in this study.

Costs:
Participation in this study will not result in any additional costs to the participant, other than the amount that is normally billed by ICES for the medical care services rendered.

Compensation for Study-related Injuries:
Patients will not receive any compensation in case of injury and there is a minimal risk when participating in this study.

Voluntary Participation and Withdrawal:
Participation in this study is strictly voluntary. Each patient will decide whether or not to participate in this study. A decision not to enroll will not affect their medical care. Participants will be able to withdraw their consent at any time during their participation in the study, and their decision will not affect their care.

Questions/Contact:
Should there be any questions regarding this study, please contact:
Dr. Mauricio Maza, Colonia Las Mercedes Ave. Los Espliegos #5, San Salvador.
Phone: (503) 7854 8873

Conflict of Interest Disclosure:
The study's Co-chair, Dr. Miriam Cremer, is a founder of Basic Health International, INC. (BHI), a non-profit organization. BHI is a collaborator in this study. BHI and Dr. Cremer could benefit from positive advertising if this research is performed correctly. Neither Dr. Cremer nor Basic Health International have financial interests in the mobile high-resolution microendoscopy system. These financial interests are

being monitored and are within the permissible limits established by the local institution and the Conflict of Interest Policies. Should there be any questions regarding the Conflict of Interest Policy, ask your doctor or call the Cleveland Clinic at (216) 444-2924.

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