Evaluate the Agreement of High-risk Human Papillomavirus Type Between Self-collected Vaginal Discharge Sample Using "HygeiaTouch Self Sampling Kit for Woman" and Physician Collected Sample From the Cervix

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Study Protocol

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Experimental theme

Comparing the consistency of self-collected vaginal specimens versus physician-collected cervical specimens for high-risk human papillomavirus testing using the "Hygeia Touch Self Sampling Kit for Women"

Study purposes

"Hygeia Touch Self Sampling Kit for Women" was created and commissioned by Hygeia Touch Inc. This kit includes a self-sampling swab developed for Oriental women. It is constructed of non-toxic and non-allergenic materials and can be used by you. Collect enough exfoliated vaginal cells to test for human papillomavirus. This is a single-use sterilizing product. It is simple to use and allows women to self-collection freely and comfortably. The collected specimens should be forwarded to a reputable laboratory at room temperature for high-risk HPV type testing. We intend to give an alternative for women who do not obtain regular cervical screening, raise the national cervical screening rate, and further lower the incidence of cervical cancer.

The trial will involve 1,200 women who will self-sample using a kit for high-risk HPV testing, and their results will be compared to cervical samples collected by physicians. The physician-collected samples will serve as the standard for evaluating the consistency of HPV typing results between the two methods. This kit is classified as a Class II medical device by the TFDA of the Ministry of Health and Welfare. If the results demonstrate strong consistency between self-sampling and physician testing, an application will be made to classify the kit as a Class 2 medical device for women's self-sampling of high-risk HPV types.

A. Primary objective:

For the test group, the consistency (agreement) of whether there is high-risk HPV detected by using the samples obtained by the self-collection kit and the samples collected by the doctor.

B. Secondary objectives:

1. Determine if women can effectively perform self-sampling by following provided instructions or videos.
2. Assess the safety of testing, including evaluations by physicians and self-sampling.
3. Evaluate the success rate of self-sampling, consistency, and compare results among different groups.
4. Examine the correlation between HPV test results selected by physicians or self-sampling, and compare with smear and histopathological diagnosis results.
5. Evaluate satisfaction with self-sampling.

Duration

2019/12/1-2021/12/31

Study design and methods

A. The primary aim of this study is to compare the reliability of self-collected specimens versus physician-collected specimens in detecting high-risk HPV. This clinical trial is being carried out at multiple centers in Taiwan with laboratory technicians being blind of the sample sources.

B. The trial aims to assess how well regular women can collect specimens using self-collection kits without professional assistance

C. To ensure effective specimen collection by ordinary women, visual aids and pre-recorded audio and video materials will be provided for self-learning. Participants will then answer a questionnaire to verify the clarity of the instructional materials. Before HPV testing, the laboratory will verify the specimen's integrity by testing for beta-globin DNA.

D. Subject inclusion procedures:

1. Eligible women who agree to participate will sign a consent form after receiving an explanation from the doctor.
2. Once the predetermined number of participants is reached, no further subjects will be enrolled, and the test results will be revealed and analyzed statistically.

E. Sample collection procedure:

1. After obtaining consent, the study nurse will provide a coded self-sampling swab to each subject for self-collection. Cervical cell collection by the physician will follow routine procedures in the outpatient clinic. The self-sampling swabs are randomly coded to ensure blinding for laboratory test operators.
2. Physician examination:

(1) Doctors use a cytological brush to collect cervical cells in accordance with medical routine during the outpatient clinic.

(2) The specimens collected for examination will be kept by nurses and sent to the National Tsing Hua University laboratory.

(3) Vaginal lavage or intravaginal medication should be avoided before specimen collection. Additional tests like smear examination or colposcopy may be conducted if necessary.

3. Self-sampling:

(1) Subjects will receive a "HygeiaTouch Self-sampling Kit for Women" containing a sterile self-sampling swab and sample collection tube. They must read the instruction manual, watch an instructional video, and confirm their understanding by answering questions.

(2) After self-sampling, subjects will place the swab in the collection tube and hand it to the study nurse.

(3) When collecting self-sampled specimens from subjects, the study nurse asked the subjects to complete the self-sampling questionnaire in the case report form and provided assistance. After the subjects fill out the questionnaire, the research nurse will take back the case report form.

(4) Subjects can self-sampling on the day of the outpatient clinic or thereafter, but no biopsy, medicine, surgery or any form of treatment is allowed between the subject's self-sampling and the doctor's sampling. The self-collected specimens are placed into the collection tube of the set and stored at room temperature. Specimens must be sent to the study nurse within one week of collection.

F. Specimen number and test results:

1. Test work bag:

(1) In order to facilitate the conduct of the trial and avoid confusion and errors, each subject will be assigned a trial work bag with a subject number on it according to the order of inclusion in the trial after signing the subject consent form.

(2) The test work bag contains:

- ♦ 1 copy of "Libo cytological Brush (set)" for medical examination
- ♦ Self-sampling: 1 copy of "HygeiaTouch Self-sampling Kit for Women"
- ♦ 1 copy of subject consent form
- ♦ 1 case report form

2. Physician collects specimens:

(1) When conducting outpatient examinations, doctors will use test work bags marked with subject numbers according to the order in which subjects are included.

(2) The subject number has been marked with a sticker on the collection tube of each "Libo Libo cytological Brush (Set)".

(3) This subject number is the specimen number collected by the doctor.

(4) In the experimental laboratory, samples are examined for HPV typing, and the results will be shared with doctors for clinical purposes. These results will be given to patients and used as a benchmark for consistency comparison in the study.

3. Self-collected samples from subjects:

(1) The serial number of the self-collected specimen has been marked with a sticker on the specimen collection tube of each "HygeiaTouch Self-sampling Kit for Women".

(2) The self-collected sample number is a number obtained by random garbled calculation based on the sample number collected by the physician. This number cannot directly determine the correlation between the specimen collected by the physician and the specimen collected by the subject.

(3) When the specimens are extracted and tested in the test laboratory, the test operators do not know the correlation between the specimens.

(4) After the trial, the self-collected specimens will be unblinded and compared with the test

results of the specimens collected by the physician for consistency.

G. Transportation and inspection of specimens:

1. The research nurse will deliver the collected specimens collected by doctors and self-collected specimens to the laboratory in National Tsing Hua University by express delivery at normal temperature for storage at -20°C and DNA extraction of the specimens .

2. The extracted DNA of the specimen will be sent by frozen express to Dr. Chip Biotechnology Industrial Co., Ltd. for HPV typing testing.

3. The laboratory's specimen operations will continue with the coding on the collection tubes, and no additional coding will be performed.

Number of subjects

This trial will include 1,200 subjects .

Subject inclusion conditions

Subjects must meet the following three conditions:

A. aged 21 years and above (inclusive) and below 65 years old (inclusive) who have not undergone total hysterectomy and uterine / cervical radiation therapy

B. Understand the trial content and sign the subject consent form

C. Those who meet one of the following 5 conditions:

1. Those with no abnormal smears or history of cervical lesions

2. Those who have had mild smear or pathological tissue abnormalities within 3-12 months and need to undergo cervical smear follow-up

Mild smear abnormalities include atypical squamous cells of undetermined significance, cervical intraepithelial neoplasia grade 1, or atypical glandular cells .

3. Have had moderate to severe smear or pathological tissue abnormalities or cervical cancer, have undergone cone surgery, and are currently being followed

Including smear is atypical squamous cells favor high-grade squamous intraepithelial lesion atypical squamous cells favor high-grade squamous intraepithelial lesion, the cytopathology cannot exclude high-grade squamous intraepithelial lesion dysplasia cannot exclude high-grade squamous intraepithelial lesion, smear or histopathology is Cervical intraepithelial neoplasia grade 2, Cervical intraepithelial neoplasia grade 3, Cervical carcinoma in situ, Cervical squamous cell carcinoma, or atypical glandular cells favor neoplasm, cervical adenocarcinoma in situ, or cervical adenocarcinoma .

4. Those who have mild smear or pathological tissue abnormalities within 3 months and have not yet been treated

Mild smear abnormalities include atypical squamous cells of undetermined significance, cervical intraepithelial neoplasia grade 1, or atypical glandular cells .

5. Those who have found moderate to severe smear or pathological tissue abnormalities or cervical cancer within 3 months and have not yet received treatment

Including smear shows atypical squamous cells favor high- grade squamous intraepithelial lesion , smear test or histopathology is Cervical intraepithelial neoplasia grade 2, Cervical intraepithelial neoplasia grade 3, Cervical carcinoma in situ, Cervical squamous cell carcinoma, or atypical glandular cells favor neoplasm, cervical adenocarcinoma in situ, or cervical adenocarcinoma .

The above groupings are also the basis for stratification of the statistical analysis of this trial .

Subject exclusion conditions

Subjects with any of the following conditions will not be able to participate in this trial:

A. Those who have undergone total hysterectomy (those who have undergone subtotal hysterectomy and still have a cervix do not need to be excluded)

B. Pregnant

C. Women with cervicitis who must receive treatment

D. Those who have received treatment for cervical lesions within 90 days

E. who have received or are currently receiving uterine / cervical or vaginal radiation therapy

F. less than 21 years old or over 65 years old

G. Having sex without condom within 8 hours

H. Excessive vaginal secretions, such as excessive clear mucus during ovulation

I. Those who are receiving local treatment for vaginitis and have medicine in their vagina

J. During the menstrual period, the menstruation has not ended yet

Evaluation indicators

Main evaluation indicators:

A. HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68 are cancer-causing high-risk types. The agreement on detecting the presence of high-risk HPV between physician-sampled specimens and self-collected specimens is listed as the main purpose. Consistency is defined as follows:

B. The high-risk HPV test results are consistent (agree) . The test results indicate that either of the above-mentioned high-risk HPV types is present in both cases or that neither of them has high-risk HPV types .

C. The high-risk HPV test results are inconsistent (disagreement). One of the samples collected by the doctor or self-sampling

D.

E. does not have any of the above-mentioned high-risk HPV types , and the other has any of the above-mentioned high-risk types.

F. The consistency between the test results of the specimens collected by the physician and those collected by the subject will be evaluated by Cohen 's kappa coefficient or compared by related tests.

Secondary evaluation indicators:

A. The success rate of HPV typing (proportion of effective specimens) for physician-collected and self-collected specimens .

B. of all detectable HPV types.

C. The incidence of adverse reactions is used as a safety evaluation index to evaluate whether adverse reactions will occur when a doctor's examination or a woman's self-sampling of vaginal secretions will occur. The test process is recorded according to the CTCAE version 5.0 grading standards.

D. Evaluate whether women can complete effective self-sampling by following the instructions or videos. The beta-globin of the test results will be used as the evaluation index of the effective specimen, and the results of the questionnaire survey will be used to evaluate the reliability of the description file.

E. The self-collection success rate (proportion of effective specimens) and consistency assessment of each group under the test group.

F. Correlation between the HPV test results collected by doctors, the HPV test results of self-collected specimens, and the cervical smear and histopathological diagnosis results at the same time. Taking the most severe pathological diagnosis as the standard, calculate separately

1. Sensitivity, specificity, positive predictive value and negative predictive value for predicting cervical lesions based on HPV test results collected by physicians

2. Sensitivity, specificity, positive predictive value and negative predictive value for predicting cervical lesions based on test results of self-collected specimens

G. Through questionnaires, subjects' self-sampling satisfaction was assessed.

Statistical methods

This study is a multi-center, operator-blinded clinical trial in Taiwan . It mainly explores the consistency of self-sampling of the product and that of doctors . This trial will use the test results of samples collected by doctors as the standard and compare them with the HPV type test results

of self-collected specimens from the same subject to verify the HPV types in the samples collected by doctors and self-sampling. consistency.

A. The trial design uses the status of cervical precancerous lesions as a stratification variable. Five groups of subjects will be recruited according to the following proportions. It is expected to recruit a total of 1,200 subjects :

The 5 groups of subjects included in the study	**Inclusion ratio**	**Estimated number of people to be included**
1. Those with no abnormal smears or history of cervical lesions	10%	120 people
2. Those who have had mild smear or pathological tissue abnormalities within 3-12 months and need to undergo cervical smear follow-up	15%	180 people
3. Have had moderate to severe smear or pathological tissue abnormalities or cervical cancer, have undergone cone surgery, and are currently being followed	20 %	240 people
4. Those who have mild smear or pathological tissue abnormalities within 3 months and have not yet been treated	20 %	240 people
5. Those who have found moderate to severe smear or pathological tissue abnormalities or cervical cancer within 3 months and have not yet received treatment	35%	420 people
The total number of subjects expected to be included		1,200 people

Based on clinical research, it is estimated that the HPV positivity rates in the above five groups are (1) 10% , (2) 70% , (3) 50%, (4) 70%, and (5) 90% respectively. Based on the experimental design, under the stratified recruitment ratio, the overall HPV positivity rate is expected to be approximately 67%.

B. According to the main evaluation indicators of the test, the consistency of this study is defined as:

1. HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68 are cancer-causing high-risk types. The agreement on detecting the presence of high-risk HPV between physician-sampled specimens and self-collected specimens is listed as the main purpose. Consistency is defined as follows :

(1) The high-risk HPV test results are consistent (agree) . The test results indicate that either of the above-mentioned high-risk HPV types is present in both cases or that neither of them has high-risk HPV types .

(2) The high-risk HPV test results are inconsistent (disagreement). One of the samples collected by the doctor or self-collected does not have any of the above-mentioned high-risk HPV types , and the other has any of the above-mentioned high-risk types .

2. The consistency between the test results of the specimens collected by the physician and the specimens collected by the subjects will be evaluated by Cohen 's kappa coefficient or related test comparisons.

C. The consistency assessment will use valid samples as the analysis group, that is, the samples collected by the verification physician and the self-collected specimens of the same subject are both valid samples. Valid specimens are defined as those who test positive for the β-globin gene in the internal control group of the Dr. Chip Human Papillomavirus Genotyping Test Kit.

D. Safety, self-sampling success rate, and satisfaction evaluation are based on the intent-to-treat population (ITT).