

Colposcopic Directed Biopsy in Positive Visual Inspection with Acetic Acid

Mohamed A. Mohamed, Amr A. El-Dien, Hend A. Mahgoub, Ali A. Bendary

Abstract:

Background: Cervical cancer is a leading cause of death among women, with about 528,000 new cases each year. It is the fourth most common cancer in women globally, with nearly 70% of cases occurring in less developed regions. This study aimed to evaluate the performance of colposcopy in estimating the presence of cervical lesion as a secondary test modality to triage women found positive on visual inspection with acetic acid (VIA) test. **Methods:** This prospective cross-sectional study was conducted at Obstetrics and Gynecology clinics of Benha University Hospitals from May 2023 till November 2023 and included 100 women who had a positive VIA test. **Results:** Patients had a mean age of 39.02 years (SD 8.4), and mean BMI of 27.26 kg/m² (SD 4.33). The median gravidity was 2 (IQR 1-3), and median parity was 2 (IQR 1-2). The majority of patients had squamous metaplasia, leukoplakia, condyloma/wart, cervical intraepithelial neoplasia CIN1 (70%), CIN2–3 (7%), and invasive carcinoma (3%). **Conclusion:** VIA and colposcopy are crucial diagnostic tools for identifying cervical pre-malignancy, especially in underdeveloped countries. VIA is commonly used alongside colposcopy for screening purposes, while colposcopy is a highly sensitive test for detecting cervical cancer and serves as a secondary test to triage women who test positive on VIA. **Keywords:** Colposcopic, Biopsy, Positive Visual Inspection, Acetic Acid.

Obstetrics and Gynecology
Department, Faculty of
Medicine Benha University,
Egypt.

Corresponding to:
Dr. Hend A. Mahgoub.
Obstetrics and Gynecology
Department, Faculty of Medicine
Benha University, Egypt.
Email:
hendahmedsoliman@gmail.com

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Introduction

Cervical cancer is the third most common cancer in women, and the seventh overall, with an estimated 530,000 new cases in 2018. More than 85% of the global burden occurs in developing countries, where it accounts for 13% of all female cancers with a mortality rate of almost 250,000 women each year, representing more than 80% of the worldwide deaths due to this preventable cancer ⁽¹⁾.

The highest burden of the disease has been reported in Asian, Latin American, and African countries. In Egypt, the age-specific incidence increases from 18.8 per 100,000 in women aged 20 to 24 years to 373.8 per 100,000 years in women aged 60 to 64 years ⁽²⁾.

Cervical cytology has seen success in the developed world in reducing the incidence and mortality of cervical cancer. However, in the developed world, the sensitivity and specificity of cytology have been shown to vary widely with an average of 53.0% (48.6–57.4%) and 96.3% (96.1–96.5%), respectively. To address the moderate sensitivity, periodic retesting is required ⁽³⁾.

Most developing countries lack the infrastructure, trained personnel, supplies and equipment to enable accurate, reliable and timely testing and reporting of results for one screening visit, much less for periodic retesting. A separate visit is commonly required to investigate abnormal cytology, and this leads to increasing rates of women who are lost to follow-up ⁽⁴⁾.

Visual inspection with 5% acetic acid (VIA) involves washing the cervix with a 3–5% acetic acid (vinegar) solution, and then examining the cervix for acetowhite areas with the naked eye one minute later. Equipment and supply requirements are minimal, test results are immediately available allowing for immediate treatment, and a variety of nonphysician providers can be trained to perform and interpret this test. Moreover, VIA is considered to be cost-effective ⁽⁵⁾.

A randomized trial in India has demonstrated that a VIA-based screening program led to reductions in both cervical cancer incidence and mortality. Previous studies have found that sensitivity and specificity of VIA vary between approximately 60–90% and 43–95%, respectively ⁽⁶⁾.

As the VIA is a test with good sensitivity, low specificity has been its limitation, which would result in excessive referrals and treatment of false-positive lesions subsequently increasing the referral load as well as the cost of unnecessary treatment on the health system. Thus, determining which women with positive VIA-based tests are at risk for significant cervical disease, performing appropriate diagnostic workups, and treating cancer precursors presents a major public health challenge ⁽⁷⁾.

Colposcopic evaluation and guided biopsy remains the critical diagnostic and is the accepted gold standard step used to assess the accuracy of VIA screening methods, especially for women with squamous intraepithelial lesions to identify women who require treatment ⁽⁸⁾.

This study aims to evaluate the performance of colposcopy in estimating the presence of cervical lesion as a secondary test modality to triage women found positive on VIA test.

Patients and methods

This prospective cross-sectional study included 100 patients selected from attendee of Obstetrics and Gynecology clinics of Benha University Hospitals from May 2023 till November 2023.

An informed written consent was obtained from the patients. Every patient received an explanation of the purpose of the study and had a secret code number. The study was done after being approved by the Research Ethics Committee, Faculty of Medicine, Benha University.

Inclusion criteria were non-pregnant women aged 25–55 years with intact

uterus, positive VIA test and no past history of cervical neoplasia

Exclusion criteria were patients who refuse to participate or previously screened for cervical cancer or have previous history of cervical intraepithelial lesions, cervical cancer or hysterectomy.

All studied cases were subjected to the following: Detailed history taking, including [Personal history; Name, age, marital status and number of children, any complaint, obstetric history, menstrual history, past medical and past surgical history and Family history. **Full clinical examination: General examination including** [Vital signs: Blood pressure, temperature, heart rate, and respiratory rate, weight and height measurement for calculation of BMI]. **Routine laboratory investigations** [complete blood count, urine analysis, kidney function tests and liver function tests].

Women first underwent screening by visual inspection with acetic acid (VIA): Acetic acid (5%) was applied to the cervix using a cotton swab and VIA findings were reported 1 min after the application as negative or positive. The result of VIA test was recorded positive when there were sharp, distinct, well-defined, dense acetowhite areas with or without raised margins, closer to the squamocolumnar junction in the transformation zone and not far away from the cervical OS ⁽⁹⁾. The women with negative findings were reassured, counseled, and sent back with an appointment for a next screening date one week later. The women with positive findings were referred to perform colposcope directed biopsy for diagnostic evaluation in the same day.

Colposcopy and biopsy:

At colposcopy, two to three biopsies from observed lesions were collected for all participants in whom acetowhite lesions were present and if the colposcopy was positive with TZ1 or TZ2. Endocervical sampling using cytology or histology (using the cell block technique) was recommended when a TZ3 was observed,

and excision type 2 or 3 was performed when high-grade disease was diagnosed (HSIL+ cytology or CIN2+ histology) ⁽¹⁰⁾. Biopsies were obtained, fixed in formalin, and processed. The colposcopic diagnosis and grading were done on Reid index, which assigns scores 1-8 for the colposcopic appearance of margins, lesion color, vascularity, and iodine staining. The four colposcopic signs were organized into three objective categories, each reflected specific characteristics of premalignant lesions at different severity levels. Each category was assigned a numerical value ranging from 0 to 2: a value of 0 indicated mild dysplasia, 1 indicated mild to moderate dysplasia, and 2 signified the presence of severe or high-grade dysplasia ⁽¹¹⁾.

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Statistical analysis

Statistical analysis was done by SPSS v28 (IBM©, Armonk, NY, USA). Shapiro-Wilks test and histograms were used to evaluate the normality of the distribution of data. Quantitative parametric data were presented as mean and standard deviation (SD) and were analyzed by ANOVA (F) test with post hoc test (Tukey). Qualitative variables were presented as frequency and percentage (%) and were analyzed utilizing the Chi-square test. A two tailed P value < 0.05 was considered statistically significant. Spearman correlation was done to estimate the degree of correlation between two quantitative variables. The overall diagnostic performance of each test was assessed by ROC curve analysis. The area under the curve (AUC) evaluates the overall test performance.

Results

According to demographic data, the mean (SD) age of the patients was 39.02 (8.4) years, the mean (SD) weight was 72.2 (10.65) kg, the mean (SD) height was 163.04 (7.01) cm, and the mean (SD) BMI was 27.26 (4.33) kg/m². According to obstetrical history, the median (IQR) gravidity of the patients was 2 (1-3), and

the median (IQR) parity was 2 (1-2). According to history data, 30 (30%) patients had regular menstrual cycles, while 70 (70%) had irregular menstrual cycles. Additionally, 18 (18%) patients experienced bleeding, and 13 (13%) patients reported discharge. **Table 1**

According to vital signs, the mean systolic blood pressure was 123.87 mmHg with a standard deviation (SD) of 9.5, and the mean diastolic blood pressure was 73.03 mmHg with a standard deviation of 8.42. Additionally, the mean heart rate was

92.94 beats per minute with a standard deviation of 12.8, while the mean respiratory rate was 15.26 breaths per minute with a standard deviation of 3.79. In terms of the complete blood count, the mean hemoglobin level among the studied patients was 11.13 g/dL with a standard deviation of 1.03. The mean red blood cell count was 4.76 million cells per microliter with a standard deviation of 0.28, and the mean white blood cell count was 7.77 thousand cells per microliter with a standard deviation of 1.65. **Table 2**

Table 1: Demographic data, obstetrical history and history data in the studied patients.

		Studied patients (n=100)
Age (years)	Mean \pm SD	39.02 \pm 8.4
Weight (kg)	Mean \pm SD	72.2 \pm 10.65
Height (cm)	Mean \pm SD	163.04 \pm 7.01
BMI (kg/m ²)	Mean \pm SD	27.26 \pm 4.33
Obstetrical history		
Gravidity	Median (IQR)	2(1-3)
Parity	Median (IQR)	2(1-2)
History data		
Menstrual history		
	Regular	30 (30%)
	Irregular	70 (70%)
Bleeding		
	Yes	18 (18%)
	No	82 (82%)
Discharge		
	Yes	13 (13%)
	No	87 (87%)

Table 2: Vital signs and complete blood count (CBC) in the studied patients.

		Studied patients (n=100)
SBP (mmHg)	Mean \pm SD	123.87 \pm 9.5
DBP (mmHg)	Mean \pm SD	73.03 \pm 8.42
HR (bpm)	Mean \pm SD	92.49 \pm 12.8
RR (breaths per minute)	Mean \pm SD	15.26 \pm 3.79
Complete Blood Cell (CBC)		
Hb (g/dl)	Mean \pm SD	11.13\pm1.03
RBCs (million/mm ³)	Mean \pm SD	4.76\pm0.28
WBCs (10 ³ /mm ³)	Mean \pm SD	7.77\pm1.65

SBP: systolic blood pressure, DBP: diastolic blood pressure, HR: heart rate, RR: respiratory rate, Hb: hemoglobin, RBCs: red blood cells count, WBCs: white blood cells count

According to Reid index, 0 (0%) of patients had a Reid score of 0-2, 90 (90%) of patients had a Reid score of 3-5, and 10 (10%) of patients had a Reid score greater than 6. **Table 3**

According to colposcopy findings, 8 patients (8%) were diagnosed with

squamous metaplasia, 7 patients (7%) had leukoplakia, 5 patients (5%) presented with condyloma/warts, 70 patients (70%) were diagnosed with CIN1, 7 patients (7%) had CIN2-3, and 3 patients (3%) were found to have invasive carcinoma.

Table 4

Table 3: Reid index in the studied patients.

		Studied patients (n=100)
Reid score 0-2	Yes	0
	NO	100(100%)
Reid score 3-5	Yes	90(90%)
	NO	10(10%)
Reid score >6	Yes	10(10%)
	NO	30(90%)

Table 4: Shows colposcopy findings in the studied patients.

Squamous metaplasia	8 (8%)
Leukoplakia	7 (7%)
Condyloma/wart	5 (5%)
CIN1	70 (70%)
CIN2-3	7 (7%)
Invasive carcinoma	3 (3%)

Discussion

This prospective cross-sectional study was conducted at the Obstetrics and Gynecology clinics of Benha University Hospitals from May 2023 to November 2023. The study was carried out on 100 women who tested positive on the VIA test.

The main aim of this study was to evaluate the performance of colposcopy in estimating the presence of cervical lesions as a secondary test modality to triage women found positive on the VIA test.

The present study showed that, based on demographic data, the mean (SD) age of the patients was 39.02 (8.4) years, the mean (SD) weight was 72.2 (10.65) kg, the

mean (SD) height was 163.04 (7.01) cm, and the mean (SD) BMI was 27.26 (4.33) kg/m².

Our results are consistent with, Shrestha & co-workers⁽¹²⁾ who aimed to compare the accuracy of visual inspection with acetic acid (VIA) with Liquid-based cytology (LBC) as a method of cancer screening. One hundred and forty-four women were included in this study. They found that the mean age of the study population was 38.93 ± 9.28 years with a range from 17 to 70 years.

Furthermore, Islam and colleagues⁽¹³⁾ who aimed to evaluate the VIA-positive cases by colposcopy, colposcopy guided biopsy, and histopathological examination. This

was a cross-sectional observational study conducted among 100 VIA-positive cases attended at the General Out-patient Department (GOPD) of Obstetrics and Gynaecology of Dhaka Medical College Hospital from July 2014 to December 2014. Colposcopy was performed in all VIA-positive cases. They revealed that according to demographic data, the mean age of the studied patients was 37.9 with $SD \pm 9.3$ years.

Moreover, Aggarwal and other researchers⁽¹⁴⁾ who aimed to compare the sensitivity, specificity, positive and negative predictive values and accuracy of visual inspection with acetic acid under magnification (VIAM) and colposcopy for detecting cervical intraepithelial neoplasia (CIN) to determine if VIAM can substitute colposcopy in identifying the biopsy site in screen-positive cases. A prospective cross-sectional study was carried out on 408 symptomatic multiparous women in the reproductive age group (15–49 years).

In our study, according to obstetric history, we found that the median (IQR) gravidity of the patients was 2 (1-3), and the median (IQR) parity was 2 (1-2).

In agreement with our results Biswas and co-workers⁽¹⁵⁾ who aimed to assess the correlation between colposcopy findings and histopathological results in cases where visual inspection of the cervix with acetic acid (VIA) was positive. This cross-sectional study involved 95 women who tested positive for the Visual Inspection with Acetic Acid (VIA) method. The findings indicated that the mean parity among the participants was 2.52, with a standard deviation of 1.26. The range of para values varied from 0 to 7.

In our study, we found that based on history data, 30 (30%) patients had regular menstrual cycles, while 70 (70%) had irregular menstrual cycles. Additionally, 18 (18%) patients experienced bleeding, and 13 (13%) patients reported discharge.

Our results are consistent with Naz and other researchers⁽¹⁶⁾ who aimed to determine the degree of agreement

between visual inspection with acetic acid (VIA) and Papanicolaou's (Pap) smear as screening methods for cervical cancer. Two hundred and fifty women in the reproductive age group presenting with various gynecological complaints were included in the study. They reported that according to history data, (46.2%) of patients had menstrual irregularities, 28 women complained of white discharge per vaginum (11.2%), and 1.6% (4) of women had a previous history of Pap smear.

In contrast with our results Akter and colleagues⁽¹⁷⁾ who aimed to find out the role of VIA in the detection of precancerous and early cancerous lesions of the cervix with co-relation of colposcopic findings and ultimate. 100 VIA-positive cases were considered for this study. They found that menstruation continued in 91% of cases and history revealed that 31 cases (34.0%) had irregular menstruation, and the rest 60 cases (65.93%) had regular menstruation with the period ranging from 2-7 days. Among the cases having regular menstruation, 35 (38.40%) had a 4-5 days duration of the cycle, which is the highest group within the regular cycle group. In 69.3% of cases had average menstrual flow.

The present study showed the following vital signs for the patients under examination: the mean systolic blood pressure was 123.87 mmHg with a standard deviation (SD) of 9.5, and the mean diastolic blood pressure was 73.03 mmHg with a standard deviation of 8.42. Additionally, the mean heart rate was 92.94 beats per minute with a standard deviation of 12.8, while the mean respiratory rate was 15.26 breaths per minute with a standard deviation of 3.79. In terms of the complete blood count, the mean hemoglobin level among the studied patients was 11.13 g/dL with a standard deviation of 1.03. The mean red blood cell count was 4.76 million cells per microliter with a standard deviation of 0.28, and the mean white blood cell count was 7.77

thousand cells per microliter with a standard deviation of 1.65.

VIA is a less expensive test and does not require a sophisticated laboratory infrastructure for testing and reporting. The negative predictive value of these tests is sufficiently high to assure screen negative women. The immediate availability of test results provides an enormous logistic advantage, facilitating diagnosis (colposcopy/biopsy) and treatment to be carried out in the same visit as the screening or the provision of treatment in a single visit approach⁽¹⁸⁾.

In our study, we found that according to the Reid index, 0 (0%) of patients had a Reid score of 0-2, 90 (90%) of patients had a Reid score of 3-5, and 10 (10%) of patients had a Reid score greater than 6.

In contrast with our results, Saleh and colleagues⁽⁹⁾ who aimed to evaluate the value of visual inspection with acetic acid (VIA) in screening cervical cancer in comparison to a Pap smear. 200 women attending the obstetrics and gynecology Department at Zagazig University Hospital from December 2011 to November 2012 were included. 24/200 (12%) women were found to be acetowhite-positive on primary screening test (VIA). They revealed that according to the Reid index, 18 (51.4%) of patients had Reid score 0-2 and considered negative, 17 (48.6%) of patients was Reid score 3-8 had a positive colposcopy.

Also, in contrast with our results, Aggarwal and co-workers⁽¹⁴⁾ who reported that according to the Reid index, 68 (68.7%) of patients had a Reid score of 0-2, 21 (21.2%) of patients had Reid scores of 3-5 and 10 (10.1%) of patients had a Reid score >6.

In our study, we observed the following results based on colposcopy findings: 8 patients (8%) were diagnosed with squamous metaplasia, 7 patients (7%) had leukoplakia, 5 patients (5%) presented with condyloma/warts, 70 patients (70%) were diagnosed with CIN1, 7 patients

(7%) had CIN2-3, and 3 patients (3%) were found to have invasive carcinoma.

Our results are consistent with Akter and other researchers⁽¹⁷⁾ who reported that a colposcopically healthy cervix was in 10 cases. Suspected inflammation, CIN I, CIN II, and CIN III were noted in 22%, 34%, 17%, and 14% of cases respectively. Invasive carcinoma was found in 3% of cases.

Our results are consistent also with Begum and colleagues⁽¹⁹⁾ who aimed to evaluate the role of colposcopy in the evaluation of VIA-positive cases. This was a cross-sectional study done among married female patients who had VIA-positive reports attending the colposcopy clinic. A total number of 97 women were included in this study having an age range between 22 and 65 years. The findings on colposcopy showed out of 97 VIA-positive patients, 3 (3.09%) had normal colposcopic findings. Colposcopy showed inflammation of the cervix in 20 (20.62%) cases, CIN I in 45 (46.39%) cases, CIN II in 12 (12.37%) cases, CIN III in 4 (4.12%) cases, and invasive cervical carcinoma in 13 (13.40%) cases.

Similarly, Islam and other researchers⁽¹³⁾ who reported that according to colposcopy findings, there were three patients with leukoplakia (3%), two patients with Condyloma/wart (2%), 10 patients with CIN1 (10%), seven patients with CIN2-3 (7%) and one patient with invasive carcinoma (1%).

Limitation: Single center study and small sample size

Conclusion

VIA and colposcopy are crucial diagnostic tools for cervical pre-malignancy, particularly in underdeveloped countries. VIA is often used alongside colposcopy for screening, while colposcopy is a sensitive test for detecting cervical cancer and can be used as a secondary test to triage women positive on VIA. It is recommended that future studies be conducted using well-designed

randomized controlled trials or large, comparative observational studies, inclusion a representative sample of patients with similar age, gender, and disease severity, Data collection using standardized tools and protocols, at regular intervals postoperatively, The sample size of future studies should be large enough to provide meaningful conclusions and to control for confounding factors, To accurately assess long-term outcomes, studies should have a longer follow-up period and we recommended that future research should include multicenter studies to validate our findings.

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Author contribution

Authors contributed equally to the study.

Conflicts of interest

No conflicts of interest

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