

**CLINICAL EVALUATION OF CYTOLOGICAL FINDINGS OF CERVICAL PAP SMEARS IN ANMCH OF GAYA, BIHAR, INDIA**

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**Abstract**

**Introduction:**

Some of the cancer control programmes and screening tests have checked the cervical cancer incidence and its related mortality. The incidence and death rate due to cervical cancer is reduced upto 80% in some of the developing countries. Pap smear cytology is useful to detect and evaluate the degree of cellular alterations seen among cervical abnormalities. As Pap smear screening test is simple, rapid and cost effective, it is an ideal tool for mass screening programmes and better reliable results are obtained compared to other tests. Hence based on above findings the present study was planned for Clinical Evaluation of Cytological Findings of Cervical PAP Smears in ANMCH Gaya, Bihar, India.

The present study was planned in Anugrah Narayan Magadh Medical College and Hospital, Gaya, Bihar, India. In the present study 25 cases of the females cervical smears of patients undergone Papanicolaou (Pap) smear testing were enrolled in the present study. Pap smears were obtained from squamocolumnar junction with the help of Ayre’s spatula. The material obtained was quickly smeared on a clean glass slide and the smear was immediately fixed in 95% ethyl alcohol. In the central laboratory, the slides were stained with Papanicolaou stain and examined under light microscope. The cytological interpretation of the smears was made according to The Bethesda System 2014 (TBS).

The data generated from the present study concludes that Pap smear tests are inexpensive and affordable by the patients. This Procedure doesn’t need experts and specialists for collection of smear. Early detection of possibility of malignancy helps in prompt treatment at early stage and prolongation of life expectancy of many women and reduce the mortality and morbidity of cancer cervix. Till today Pap smear test is the most useful screening procedure for cervical cancer.

**Keywords:** cytology, pap smear, screening, squamous intraepithelial lesion, etc.

**Introduction**

Worldwide, approximately 500,000 new cases of cervical cancer and 274,000 deaths are attributable to cervical cancer yearly, making cervical cancer the second most common cause of death from cancer in women. [1] Fortunately, the incidence of cervical cancer has decreased by more than 50% in the past 30+ years, largely due to the increasing use of cervical cancer screening with cervical cytology. [2]

The American Cancer Society estimates about 13,240 new cases of invasive cervical cancer will be diagnosed and about 4170 women will die from cervical cancer in 2018. [3] Although worldwide cervical cancer rates have decreased dramatically with the increase in screening efforts, incidence and prevalence in developing countries remains high due to lack of screening programs, with approximately 80% of all cervical cancer deaths occurring in the developing world. [1]

The mainstay of cervical cancer screening for the last 60+ years has been the Papanicolaou test. The Papanicolaou test, also known as the Pap test or the Pap smear, was

developed in the 1940s by Georgios Papanikolaou. It involves exfoliating cells from the transformation zone of the cervix to enable examination of these cells microscopically for detection of cancerous or precancerous lesions.

In the technique known as liquid-based cytology, these collected cells are released into a vial of liquid preservative that is then used in the cytology lab to produce a slide for microscopic evaluation of the cells. The older, traditional Pap technique involves direct transfer of the cervical cells to a microscope slide for evaluation. Although the traditional method may introduce confounders such as blood and other debris to the slide, which may make interpretation more difficult, both conventional cytology and liquid-based cytology have been shown to have similar sensitivity and specificity for moderate dysplasia or worse lesions when using a threshold of LSIL or higher. In addition, both types of cytological screening are considered acceptable by the American College of Obstetricians and Gynecologists. [2]

When abnormal cells are detected on the Pap Test, diagnostic testing in the form of colposcopy is often indicated. This testing may be followed by diagnosis of dysplasia via colposcopic biopsies. Subsequent cervical cancer may be prevented through the diagnosis and treatment of these cervical cancer precursors.

Evidence shows that approximately 99-100% of cervical cancers are attributable to infection by high-risk types of the human papillomavirus (HPV). HPV represents a family of double-stranded, circular DNA viruses that can infect skin or mucosal cells, including the anogenital region and the oral cavity, and may be transmitted easily via sexual intercourse or direct contact. [4, 5]

More than 100 types of HPV exist, 12 of which can involve the anogenital region and are considered "high risk" or oncogenic in nature. These include HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, and 59. Of these, HPV 16 is responsible for the largest number of CIN 3 and cervical cancer cases, and HPV 16 and 18 combined are thought to be responsible for nearly 70% of all cases of cervical cancer. [4] Although HPV is a necessary factor in the development of cervical dysplasia that can eventually lead to cervical cancer, most women infected with HPV will not develop cervical dysplasia. [6] The presence of high-risk HPV DNA is accompanied by cytologic abnormalities approximately one third of the time. Whether an HPV infection will progress relates to the persistence of the infection and also possibly to the immune response and smoking status of the woman. [7]

The female reproductive organs can be subdivided into the internal and external genitalia. The internal genitalia are those organs that are within the true pelvis. These include the vagina, uterus, cervix, uterine tubes (oviducts or fallopian tubes), and ovaries. The external genitalia lie outside the true pelvis. These include the perineum, mons pubis, clitoris, urethral (urinary) meatus, labia majora and minora, vestibule, greater vestibular (Bartholin) glands, Skene glands, and periurethral area.

The cervix is the inferior portion of the uterus, separating the body of the uterus from the vagina. The cervix is cylindrical in shape, with an endocervical canal located in the midline, allowing passage of semen into the uterus. The external opening into the vagina is termed the external os, and the internal opening into the endometrial cavity is termed the internal os. The internal os is the portion of a female cervix that dilates to allow delivery of the fetus during labor. The average length of the cervix is 3-5 cm.

The Pap test is indicated to screen for malignant and premalignant lesions of the cervix. The recommended age at initiation of cervical cancer screening has

undergone significant revision over time as the natural history of HPV infection and subsequent cervical dysplasia has been elucidated. Although former guidelines recommended starting Pap smear screening at age 18 or the onset of sexual activity, these guidelines were revised in 2006 to recommend initiation 3 years after the onset of sexual activity or age 21, whichever comes first. In 2009, these were further revised to recommend that cervical cancer screening begin at age 21, regardless of sexual history. This recommendation was confirmed in 2012 and again in January 2016. [2]

Abnormal cervical cytology is very common in young women, and most abnormal cytology resolves without treatment in adolescents. In addition, women under the age of 21 account for only 0.1% of all cervical cancers, and no evidence exists that cervical cancer screening in this age group reduces cervical cancer incidence, morbidity, or mortality.

Recognizing these facts and the likelihood of cervical cancer screening leading to unnecessary and potentially harmful evaluation and treatment in women at very low risk for malignancy, the 2009 ACOG guideline revision recommended cervical cancer screening beginning at age 21 years of age, regardless of sexual history, and that recommendation remains unchanged. [2]

Both the U.S. Preventive Services Task Force (USPSTF) and the American Cancer Society (ACS) agreed with this recommendation and issued age-appropriate screening strategies for cytology (Pap tests) and HPV testing for cervical cancer screening in 2012. [8, 9]

Note that HPV DNA testing for the purpose of primary cervical cancer screening is currently not yet recommended by major societies in women younger than age 30 years due to the high prevalence of HPV infections in this age group as well as the transient nature of these infections. One HPV test was approved by the FDA in 2014 for primary HPV screening in women age 25 years and older. This test may be used as an alternative to cytology in women in this age group; however, appropriate algorithms should be followed according to the interim guidance by the ASCCP and Society for Gynecologic Oncologists (SGO). [10] HPV DNA testing is not recommended in women younger than 21 years of age for any reason or following any cytology result (and if performed should be ignored). In addition, testing for low-risk HPV types is never appropriate and should not be performed under any circumstances.

Reviews conducted for the US Preventive Services Task Force support these indications. Vesco et al confirmed the low incidence of cervical cancer among women younger than 20 years and underscored the difficulties of

detection and the high frequency of false-positives in this age group. [11] Further, incidence of and mortality rates from cervical cancer in women aged 65 years and older who have had a Pap smear within 3 years have decreased since 2000, and available evidence reinforces discontinuation of cervical cancer screening among these women who have had satisfactory screening and are not otherwise at high risk.

In a systematic review, Whitlock et al found evidence supporting the use of liquid-based or conventional cytology for cervical cancer screening, but cautioned that more evidence is needed before adopting HPV-enhanced primary screening for women aged 30 years and older. [12]

A metal or plastic speculum is placed in the vagina to examine the cervix. If necessary, lukewarm water may be used to lubricate and warm the speculum for patient comfort. In situations in which a lubricant must be used, only a small amount should be applied to the outer portion of the speculum, with caution to avoid the tip. According to Hologic Inc, maker of ThinPrep pap test, the following lubricants do not contain substances that interfere with the liquid-based Pap tests: Surgilube, Astroglide, and Crystelle. [13]

To ensure an adequate sample is collected, the surface anatomy of the cervix must be fully visualized, including the squamous epithelium of the ectocervix, squamocolumnar junction, and the external os. The transformation zone of the cervix is the region where squamous epithelium replaces glandular epithelium in a process called squamous metaplasia. Because HPV has a predilection for this region, [14] screening must focus on sampling the cells at the transformation zone to adequately detect the presence of dysplasia. Discharge covering the cervix may be removed carefully using a large swab, ensuring that the cervix is minimally traumatized.

To obtain the specimen, a cervical broom or cervical spatula is applied to the surface of the cervix and turned in a single direction to achieve an adequate sample for cytology, making sure to rotate it at least 360° for the spatula and 5 rotations for the broom. If the spatula is used, a cytobrush is additionally needed and must be inserted into the cervix so that the outermost bristles are still visible at the external os. The brush is then rotated one half turn in a single direction to achieve an adequate cytology sample.

The specific protocol for specimen transfer varies depending on the test used. For SurePath, after the cervical broom or cervical spatula and cytobrush are removed from the cervix, they are placed specimen side

down into the liquid cytology vial, each removable head is snapped off, and the vial is labeled and sent to pathology. For ThinPrep, the spatula and brush are to be swirled vigorously in the vial 10 times to release the specimen and then discarded. Similarly, if the broom is used, it is to be pushed into the bottom of the vial 10 times and then swirled vigorously and discarded. When conventional cytology is to be performed, the specimens are smeared on a glass slide and subsequently sprayed with fixative or placed in 90% alcohol solution.

Although the FDA-approved protocol for the cervical broom does not require use of the cytobrush, some practitioners use the cytobrush following the broom in an attempt to improve the likelihood of obtaining an endocervical component in the sample. Small studies show no significant difference in acquiring endocervical cells between the broom and spatula plus cytobrush; however, other studies have shown the spatula/cytobrush method to be better at sampling endocervical cells than the broom alone. [15]

In addition, 2 much larger studies found that the broom/cytobrush combination improves sampling of the endocervix compared to the broom alone. [16] Whether these potential sampling differences affect the sensitivity of cervical cytology for detecting moderate or severe dysplasia or cancer is unclear; however, based on the available data, using the cytobrush to obtain endocervical cells in addition to the spatula or the broom is reasonable

Some of the cancer control programmes and screening tests have checked the cervical cancer incidence and its related mortality. The incidence and death rate due to cervical cancer is reduced upto 80% in some of the developing countries. Pap smear cytology is useful to detect and evaluate the degree of cellular alterations seen among cervical abnormalities. As Pap smear screening test is simple, rapid and cost effective, it is an ideal tool for mass screening programmes and better reliable results are obtained compared to other tests. [17] Hence based on above findings the present study was planned for Clinical Evaluation of Cytological Findings of Cervical PAP Smears in a Tertiary Care Hospital of Gaya, Bihar, India.

#### **Methodology:**

The present study was planned in Anugrah Narayan Magadh Medical College and Hospital, Gaya, Bihar, India. In the present study 25 cases of the females cervical smears of patients undergone Papanicolaou (Pap) smear testing were enrolled in the present study.

Pap smears were obtained from squamocolumnar junction with the help of Ayre's spatula. The material obtained was quickly smeared on a clean glass slide and the smear was immediately fixed in 95% ethyl alcohol. In the central laboratory, the slides were stained with Papanicolaou stain and examined under light microscope. The cytological interpretation of the smears was made according to The Bethesda System 2014 (TBS).

All the patients were informed consents. The aim and the objective of the present study were conveyed to them. Approval of the institutional ethical committee was taken prior to conduct of this study.

Following was the inclusion and exclusion criteria for the present study.

Inclusion criteria: • Women between 25 to 70 years of age with sexual history.

Exclusion criteria: • Women below 25 years. • Women without sexual exposure. • Women above 70 years.

### Results & Discussion:

Cervical cancer is an increasing health problem, comprising approximately 12% of all cancers among women worldwide. [18] According to the world cancer statistics, developing and low resource countries have more than 80% of all the cervical cancers due to lack of awareness and difficulty in running cytology-based screening programmes. [19] India has the highest age standardized incidence of cervical cancers in South Asia. [20] By simple pap screening test cervical cancer and its precursor lesions can be detected and treated early. Pap smear is a routine screening test with sensitivity of 70-80% in detecting HSIL. [21] Usually Pap smear screening test is recommended starting around 21 years of age upto 65 years. Repeated examination is recommended after every three years interval and in case of abnormal Pap smear report follow up is advisable six monthly. [22]

The differences in prognosis of cancer according to stage have encouraged physicians and the public in belief that if only cancer could be found early enough then it could almost invariably be cured. We recognized that the majority of the cases of cancer of cervix pass through an in situ phase, when they are detectable by means of cervical smear and it is believed that this is preceded by a phase of dysplasia. We know that an appreciable proportion of in situ cases and even greater proportion of dysplasia cases regress. However as we can not identify which precursor lesion will progress to invasive cancer, if left untreated. All patients with detected lesion must be treated and passed on special surveillance. The screening test appear to identify cases in an in situ and early invasive phase, that have a long survival, yet the

long survival appears due to the additional observation period gained by earlier diagnosis possible with a screening test (Lead time).

**Table 1:** Age Groups

Age	No. of Cases
25 – 35 years	2
36 – 45 years	8
46 – 55 years	9
56 – 65 years	4
66 & above years	2
Total	25

**Table 2:** Different cytological findings

Different cytological findings	No. of Cases
Unsatisfactory smears :	
Inadequate sample	2
Obscured with blood	1
Normal smear	4
Abnormal smears	18
Total	25

**Table 3:** Cyto diagnosis

Cyto diagnosis	No. of Cases
A) NILM: Negative for Intra epithelial Lesion or malignancy	23
Normal	4
Inflammatory:	
Non-specific	18
Candida	1
Trichomonas	0
B) ASCUS: Atypical Squamous Cell of Undermined Significance	2
SIL: Squamous Intraepithelial Lesion	
LSIL: Low grade Squamous Intraepithelial Lesion	1
HSIL: Low Grade Squamous Intraepithelial Lesion	0
Carcinoma:	
SCC: Squamous Cell Carcinoma	1
ADC: Adenocarcinoma	0
Inadequate:	0
Total	25

A recent study conducted in Ningen Dock, Japan aimed to determine the gynaecological status of asymptomatic women who attended the hospital for health check-up, showed low prevalence cervical cell abnormalities of 1.2%. The explanation behind this result is mostly because of their cultural traditions and great concern regarding their health check-ups and less likelihood of having multiple sexual partners. [23]

The age distribution pattern of invasive cancer shows a rise that starts in the early 20s continues up sharply in the 30s, and plateaus at 40 to 50 years of age. Data cancer registers in developing countries indicate that more than 80 to –90% cervical cancer cases develop in women 35 years or older. Most of studies in developing countries revealed that average age of women with CIS was between 35-44 years.

Intensive screening programmes in various countries show a striking reduction in mortality from cancer of cervix. In general, in countries where Pap smear screening is routine, it is recommended that females who have had sex should seek regular Pap smear testing. Guidelines on frequency vary from every three to five years. If results are abnormal, and depending on the nature of the abnormality, the test may need to be repeated in six to twelve months. [24] In 1988, the Bethesda system of terminology has been introduced to sub-classify the lesions into grades: high grade and low grade Squamous Intraepithelial Lesions (SIL) for Pap smear reporting and some studies reported comparison of various terminologies. [25-26] The Bethesda System (TBS) for reporting the results of cervical cytology was developed as a uniform system of terminology that could provide clear guidance for clinical management. [27]

According to National Cancer Registry Program of India, cancers of uterine cervix and breast are the leading malignancies seen in females of India. There should be an effective mass screening program aimed at specific age group for detecting precancerous condition before they progress to invasive cancers. [28] It is a well-known fact that the burden of cervical cancer has been reduced dramatically after the introduction of screening programmes. [29] Prevention of cervical cancer can be primary or secondary. Primary prevention modalities include changes in sexual behaviour and Human Papilloma Virus (HPV) vaccination. Secondary prevention of cervical cancer includes visual inspection of cervix, cervicoscopy, HPV testing and cytology. Pap smear test is a secondary preventive method aimed at identification of premalignant and malignant lesions, which may need follow-up and/or treatment. [30]

Cervical cancer is one of the most common malignancies in women of developing country like India. Pap smear is a simple, cheap, safe and practical diagnostic tool for early detection of cervical cancer in high risk group population; so it should be established as a routine screening procedure. It also has a greater role in diagnosis of inflammatory lesions including the identification of causative organism, atrophic changes, changes of radiation therapy and some rare tumors.

As we all know that cervical cancer is one of the leading cause of mortality in India and its precursor lesions usually occur 5-10 years earlier. Henceforth, Pap smear examination is an important and fundamental tool for the screening, prevention and early diagnosis of cervical cancers.

#### Conclusion:

The data generated from the present study concludes that Pap smear tests are inexpensive and affordable by the patients. This Procedure doesn't need experts and specialists for collection of smear. Early detection of possibility of malignancy helps in prompt treatment at early stage and prolongation of life expectancy of many women and reduce the mortality and morbidity of cancer cervix. Till today Pap smear test is the most useful screening procedure for cervical cancer.

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