Original Research Article
Study of Pap Smear with Revised Bethesda System of Reporting, in Screening of Ca Cervix, in Patients Attending in Tertiary Care Hospital at Darbhanga, Bihar

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Abstract
Objective: Carcinoma cervix is the most common carcinoma of female. India is said to be the capital of cancer cervix because of highest incidence rate and mortality rate. It is more prevalent in rural area. In urban area its incidence is somewhat lower where carcinoma breast is emerging as most common cancer in female. The aim of present study was to determine the prevalence of Carcinoma cervix and role of pap smear with revised Bethesda system of reporting as a screening test.

Materials and Method: A total of 78 female patients, of different age groups, presents with various genital complains, attending in obstetrics and gynecology out patients door were included in the study. All the demographic data were noted. From all the patient pap smear were taken and send for cytological evaluation in pathology department. All the fixed smears were stained by papanicolaou stain and seen under oil immersion lens.

Result: Out of 78 patients, 66.67% patients were in 51-60 years of age, 60.26% women were comes from Hindu community. 55.13% patients had low socioeconomic status, multiparity was seen in (62.82%) of cases and 65.38% had rural based origin. Majority of patients, 71.79% had lower abdominal pain, followed by chronic backache in 66.67%, vaginal discharge in 41.02% and 30.76% had Irregular vaginal bleeding. In cytological finding maximum patients (41.02%) were found to have Negative for intraepithelial malignancy (NILM), Atypical squamous cell of undetermined significance (ASCUS) was seen in 33.34% of cases, 15.38% patients had Low grade squamous intraepithelial lesion (LSIL), 7.70% patients showed High grade squamous intraepithelial lesion (HSIL) and 2.56% patients were diagnosed as squamous cell carcinoma. Bacterial vaginosis was the most common infection found in 30.76% of patients followed by fungal (Candida albicans) in 8.98%, Trichomonas in 6.42% of patients and mixed infection in 10.26% of cases. Maximum number of patients had only mild dysplastic charges and were seen in 45-55 year of age groups. In correlation of parity with dysplasia, ASCUS was most common in 1-3 children group, LSIL was most common in 4-5 children group and HSIL was noticed mostly in 4-5 group.

Conclusion: pap smear is a rapid, cost effective and outpatient department procedure which is capable of detecting neoplastic as well as non-neoplastic cervical lesions at the earliest and if applied judiciously to all women it will lead to a fall in incidence and mortality of cervical cancer. Since prevention is always better than cure, we should stress more on education, prevention and treatment of sexually transmitted diseases, family planning measures, widespread use of cytological screening, referral system for patients with dysplasia’s to the good more thoroughly equipped centers and of course, utilization of all possible resources for proper management and follow up of these cases. It will decrease the morbidity as well as mortality associated with invasive cancer of cervix.

Keywords: Pap smear, screening test, Bethesda system of reporting, LSIL, HSIL, Ca Cervix.
Introduction

Cytology is the study of cells. Each cell performs its predestined function and passes away to leave its place to a new cell. This fundamental biological law finds a most typical expression in the epithelial tissues, i.e., the skin and the mucous membranes. These tissues, because of their exposure to the action of extraneous factors, are subjected to an extensive falling or shedding of their superficially located cells. As these cells are lost, their places are promptly taken by new cells growing from undifferentiated basal layer of epithelium, which is endowed with an inherent high potentiality for proliferation and growth.

The Indian Council of Medical Research (ICMR) stated that in 2016 the total number of new cancer cases is expected to be around 14.5 lakh and the figure is likely to reach nearly 17.3 lakh new cases in 2020. Over 7.36 lakh people are expected to succumb to the disease in 2016 while the figure is estimated to shoot up to 8.8 lakh by 2020. Data also revealed that only 12.5 per cent of patients come for treatment in early stages of the disease. Among females, breast cancer topped the list and among males mouth cancer, the study said.

The cervical smear or Pap smear is a routine screening procedure after certain age in female in various foreign countries. But in our country it has not gain its position because of poverty, illiteracy, negligence of female health and lack of government policy. According to ICMR, New Delhi screening test should be done regularly on all women above 30 yrs. The interval between two pap smear screening is 3 years if first smear is in category of LSIL or lower than that. However more frequent screening is needed in HSIL category. Routine pap smear will detect CA cervix at an early stage and hence helpful in reducing morbidity and mortality.

Cervical cytology is done for various diseases affecting the female genital tract. It can be Inflammatory (Bacterial, Viral, Fungal and Parasitic), degenerative or retrogressive, cytohormonal changes and neoplastic

Previously there was no unanimous reporting system but later on a lot of reporting method came in vogue. The most appreciable one was WHO classification (1956). After that Richard (2001) classified cervical lesion as CIN I, CIN II, CIN III and invasive cancer cervix. Now a days, reporting system for cervical smear is Bethesda system (2001, Revised 2006) this system takes into account the adequacy of specimen. Four elements contribute to adequacy like Patient and specimen identification, Pertinent clinical information, Technical interpretability, cellular composition and sampling of the transformation zone. A satisfactory smear should therefore have appropriate labeling and identifying information, relevant clinical information, adequate numbers of well preserved, well visualized Squamous epithelial cells and an adequate endocervical or transformation zone component i.e. a minimum of two clusters of endocervical cells, each containing at least five cells or squamous meta plastic cells. With the advent of this reporting system it is possible to recognize Ca Cervix at an early stage and reduce false positive cases.

Material & Method

Present study was carried out in the Department of pathology, Darbhanga Medical College, Laheriasarai, with the help of Obstetrics and Gynecology Department, during the period of January 2016 to January 2018. A total of 78 female patients, of different age groups, presents with various genito-urinary complains attending in obstetrics and gynecology out patients door ,were included in study. All the demographic data (regarding age, religion. Socioeconomic status, parity, locality and presenting complains of the patients) were noted. From all the patient cervical smear (pap smear) were taken by the help of Ayre's spatula. The material obtained was smeared on grease free dry slide, wet fixed and send for cytological evaluation in our department. All the fixed smears were stained by papanicolaou stain and seen under oil immersion lens.
Results

Table shows Pap smear taken from different age group of female patients.

| Age Group of female patients in years | Total No. female patients included in study | Percentage |
|---------------------------------------|--------------------------------------------|------------|
| 30-40                                 | 4                                          | 5.13       |
| 41-50                                 | 12                                         | 15.38      |
| 51-60                                 | 52                                         | 66.67      |
| 61-65                                 | 7                                          | 8.98       |
| 66-70                                 | 2                                          | 2.56       |
| >70                                   | 1                                          | 1.28       |
| Total patients                        | 78                                         |            |

Out of 78 female, undergone pap smear examination, maximum no. of patients’66.67% was belonged to 51 - 60 years of age.

Out of 78 female patients undergone Pap smear, maximum number 47 (60.26%) belonged to Hindu community followed by Muslims comprising 28(35.90%) of cases, Christian 2 (2.56%) cases and Sikhs contributed to 1 (1.28%).

Out of 78 patients Maximum no. of patients 43 (55.13%) comes from low socio economic status followed by 28 (35.90%) patients were from middle income group and only 7 (8.97%) patients belonged to high income group.
Out of 78 patients 2(2.56%) patients had no any children’s, 26(33.33%) females had 1 to 3 children’s, 49(62.82%) patients bears 4-5 children’s and only one female had more than 6 children.

Out of 78 patients, 51 patients (65.38%) resides in rural area and 27(34.62%) were urban residents.

Out of 78 patients, 32(41.02%) patients had Different types of vaginal discharge, 24(30.76%) patients had Irregular vaginal bleeding, 8(10.25%) patients had Bleeding or pain on coitus, 52(66.67%) patients had Chronic backache, 56(71.79) patients had Lower abdominal pain and 30(38.46%) patients had Dysuria.
Out of 78 patients, 34 (43.58%) had No any pathogenic organism seen, 24 (30.76%) patients had Bacterial Vaginosis, 7 (8.89%) patients had Candida albicans, 5 (6.42%) patients had Trichomoniasis and 8 (10.26%) patients had Mixed infection.

Table Shows PAP smear reporting according to Bethesda system

| Cytological finding                        | No. of patients | Percentage |
|--------------------------------------------|-----------------|------------|
| Negative for intraepithelial malignancy(NILM)| 32              | 41.02      |
| Atypical squamous cell of undetermined significance (ASCUS) | 26 | 33.34 |
| Low grade squamous epithelial lesion(LSIL)  | 12              | 15.38      |
| High grade squamous epithelial lesion(HSIL) | 6               | 7.70       |
| Squamous Cell Carcinoma                    | 2               | 2.56       |

Maximum patients, 41.02% were found to have Negative for intraepithelial malignancy (NILM), Atypical squamous cell of undetermined significance (ASCUS) was seen in 33.34% of cases, 15.38% of patients had Low grade squamous epithelial lesion (LSIL), 7.70% had High grade squamous epithelial lesion (HSIL) and 2.56% patients had Squamous Cell Carcinoma.

Table Shows Comparative study of Dysplasia according to age groups

| Age Group       | Number of Patients |
|-----------------|--------------------|
|                 | Atypical squamous cell of undetermined significance. | Low grade squamous epithelial lesion. | High grade squamous epithelial lesion. |
| 45-55 Years     | 23                 | 9                   | 4                      |
| 56-65 Years     | 2                  | 2                   | 2                      |
| 66 – 75 Years   | 1                  | 1                   | 0                      |

Maximum number of patients had only mild dysplastic changes and maximum number of dysplastic changes were seen in 45-55 years age group. 58.53% of patients showed different grades of dysplasia, out of them 29.26% had ASCUS, 19.51% showed LSIL, and 9.57% showed HSIL.

Table Shows Comparative study of parity with Dysplasia

| Age Group       | Number of Patients |
|-----------------|--------------------|
|                 | Atypical squamous cell of undetermined significance. | Low grade squamous epithelial lesion. | High grade squamous epithelial lesion. |
| No Children     | 1                  | 0                   | 0                      |
| 1-3 Children    | 16                 | 2                   | 1                      |
| 4-5 Children    | 9                  | 10                  | 4                      |
| More than 5 Children | 0              | 0                   | 1                      |
Discussion
The term cervical intra epithelial neoplasia denotes a continuum of disorders, from mild through moderate to severe dysplasia and carcinoma in situ. Mild dysplasia is often seen in inflammatory conditions like trichomoniasis, HPV and is reversible following treatment whereas severe varieties may progress to invasive cancer in about 10-30% of cases in 5-10 yrs time. The ICMR reports an incidence of dysplasia to be 15:1000 women cytological screened and incidence of severe dysplasia is reported to be about 5: 1000. According to Abuja from Hyderabad the risk of dysplasia progressing to frank invasive carcinoma was 0.5% in mild dysplasia as compared to 9.6% for severe dysplasia.

The term mild dysplasia or CIN I is equivalent to LSIL of Bethesda system of reporting whereas moderate dysplasia or CIN II and severe dysplasia or CIN III or carcinoma in situ are merged into HSIL of Bethesda system. In LSIL category the nucleus occupies less than 50% of total epithelial thickness. The nucleus is enlarged with irregular out line.

HSIL category includes CIN II and CIN III. CIN II is labeled when undifferentiated cells occupy the lower 50-75% epithelial thickness. In CIN III the entire thickness is occupied by abnormal cells. There is no cornification and stratification is lost. In CIN II cells are mostly intermediate with moderate nuclear enlargement, hyper chromatic and irregular chromatin. In CIN III the cells are mostly parabasal with increased N:C ratio, irregular nuclei with coarse chromatin material.

The Squamous cell carcinoma in a cervical smear is identified by the presence of discohesive smear with numerous, single abnormal squamous cell and marked anisocytosis and anisonucleosis. Besides parakeratotic cells containing atypical nuclei, tadpole and caudate cells, spindle keratinized cells and keratinized cells with bizarre cytoplasm forms are also present
In our study maximum number of cases (66.67% ) were seen in the age group of 51-60 years. Ambedkar Raj Kuldandai Velu, Banushree C Srinivasamurthy, M Balamurugan in their study of "Clinicopathologic significance of Papanicolaou smear study of postmenopausal women in a rural tertiary care center" reported maximum number of cases in the age group of 50-59 years.280 patient were in the age group of 50-59 years out of total 590 smears.

In our study maximum number (60.26%) of female comes from Hindu community, followed by Muslims 35.90 %, Sikhs 1.28% and Christians contributed to 2.56 %.

55.13%of female patients were comes from lower socioeconomic status, 35.90% patients were from middle income group and 8.97% patients belonged from high income group. As expected lower income group comprised the maximum because of multiple pregnancy, repeated abortion, severe malnutrition, PID, lack of proper antenatal check up and lack of safe sexual practice.

Our study revealed that 2.56% of patients were nulliparous, 62.82% had 4-5 children and 33.33% had 1-3 children. Incidence of dysplasia and even squamous cell carcinoma were highest in 4-5 children group. Luthra et al, Mali et al , Singh et al, stated that repeated pregnancy caused lacerations of the cervix, after healing by secondary intention, the cervical canal become more patulous and may cause the infection the cervical canal. This supports the theory of an infective agent in the causation of dysplasia and carcinoma cervix. Munoz N et al, International Agency for Research on Cancer, Multicentre Cervical Cancer Study Group, (2002), concluded that high parity increases the risk of squamous cell carcinoma of the cervix among the Human Papilloma Virus positive women.

According to my study 65.38% of patients were resides rural areas are more risk of cervical cancer due to most of them are deprived of basic health facilities. Pooja H. Khakhla et al in their study observed that 67.5 % of cases were from the urban areas while 32.5% were from rural areas.

In my study 71.79% patients complained of lower abdominal pain or discomfort and 66.67%
complained of backache. The preponderance of these symptoms may be due to the associated parametritis. Vaginal discharge were found in 41.02 % of cases indicating associated cervicitis. Dysuria (38.46%), indicated urinary tract infection and probably the same organism causing cervicitis. Mithila Bisht, Shweta Agarwal, Deepak Upadhyay et al in their study showed that most of the women had multiple symptoms. Vaginal discharge was the most common presentation (56.37%) followed by pruritus (36.31%), burning micturition (24.85%), pelvic pain (20%) and backache (17.98%).

In my cytological study 41.02% were found to have NILM (Negative for intra epithelial lesion or malignancy). Atypical Squamous cells of undetermined Significance (ASCUS) was seen in 33.34 % of cases.15.38% of cases showed Low grade squamous intraepithelial lesion (LSIL), 7.70% showed High grade squamous intraepithelial lesion (HSIL) and 2.56% showed Squamous cell carcinoma. Mithila Bisht, Shweta Agarwal, Deepak Upadhyay et al in their study showed that 85.16% slides were found to be negative for intraepithelial abnormalities. Among the epithelial cell abnormalities, most frequent was Atypical Squamous Cells of Undetermined Significance (ASCUS) reported in 10.08% cases followed by LSIL reported in 1.10% patients. Out of all smears examined, 0.56% Pap smears revealed high grade lesions and malignancy (0.45% HSIL and 0.09% malignancy).

Out of 78 cases, infection was found in 34 cases. Bacterial vaginosis was the most common infection found in 30.76 % of patients, followed by Fungal (Candida) on 8.98% of patients. The interpretation of inflammatory condition in pap smear varies with different investigators. Singh et al reported 35.2 % cases with infection and Sinha et al reported 11.1% cases.

Maximum number of patients had only mild dysplastic changes and maximum number of dysplastic changes were seen in 45-55 years age group. In our study 58.53% of patients showed different grades of dysplasia, out of them 29.26 % had ASCUS, 19.51 % showed LSIL, and 9.57 % showed HSIL. Stern and Nelly (1963) reported incidence of dysplasia to be around 0.54%.

According to ICMR, 2.3% of incidence of dysplasia was seen in New Delhi. Manjit Singh Bal, Rishu Goyal, Anil Kumar Suri, Manjit Kaur Mohi (2012) reported the incidence of dysplasia to be 5% in Patiala, Punjab.

Low socioeconomic status, early marriage, early sexual exposure and early child birth, leads to a grim situation in our country especially in Bihar, leading and factors are high risk for cervical infection and their consequences.

**Conclusion**

Women especially from the rural area and low socioeconomic status suffers disproportionately from precancerous and cancerous lesions of the female genital tract. To reduce this burden it is mandatory to motivate and screen asymptomatic and symptomatic older women for the pap smear after 35 years of age. The easy accessibility of the cervix and the propensity of the cancer cells to exfoliate from its surface enables us to study the process of malignant transformation in the cervix in very early stage. It can be concluded that pap smear screening is very helpful in easy detection of carcinoma because it can detect cases of LSIL and HSIL which can progress to carcinoma. So pap smear should be made mandatory for women after a certain age.

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