

## Study of Cervical Cytology and its correlation with Histopathology in diagnosis of Precancerous and Cancerous Lesions of Cervix

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### Abstract

**Background:** The Pap smear is the screening test in which in case of detection of any abnormality, a cervical biopsy is advised for confirming diagnosis and correlation. The present study is carried out to evaluate the pattern of cervical Pap smear cytology at a tertiary care teaching hospital and to correlate it with histopathological findings. **Objectives:** To assess The Bethesda system (TBS 2014) as a reporting system for cervico-vaginal cytology in detection of abnormal cervical cytology diagnostic categories. To compare the results of individual diagnostic cervical cytology categories with histopathological equivalents in available biopsies of cervix. **Material & Methods:** The present study is a Retrospective study conducted in tertiary care rural teaching hospital. The study was conducted in the period from January 2015 to January 2020. A total of 3596 patients underwent cervical cytology by Ayres wooden disposable spatula at Sharda Hospital during this period. The number of Negative for Intra Epithelial Lesion (NILM) and Abnormal cervical cytology (ACC) Cases was noted. The cytomorphology in each diagnostic category for their frequent and common feature was observed. The available biopsies of the cervix from the NILM cases and cases of abnormal cervical cytology of the study were processed by the conventional method of tissue processing. These biopsies were stained by H & E Stain. The assessment of the each category diagnosed by TBS 2014 was compared with the histopathological diagnosis in the available biopsies of the cervix. The values of comparison of TBS 2014 was bought out considering histopathology as a gold standard of diagnosis. **Results & Conclusion:** In a total of 3,596 patients, who underwent the cervical cytology during the study period, total number of cases selected for the study were 199. A total of 99 cases were diagnosed as abnormal cervical cytology and 100 patients were picked up as age match control which were diagnosed as Negative for Intra Epithelial Lesion or Malignancy on Pap smear cytology. The percentile prevalence of abnormal cervical cytology was 2.75% in total of 3,596 cervical cytology performed during the period of study. The categories of TBS 2014 appeared to correlate in all its categories except for ASC-H where in two cases showed the higher lesion. There were no false reporting of the higher lesions (False Positive) when TBS 2014 was applied in the present study. However there were 4 cases in ASC-H category where under reporting of higher lesions has occurred (3 cases of CIN 1, 1 case of CIN 2). No case diagnosed as NILM by TBS 2014 in present study turned to carry higher lesion on the biopsy. The TBS 2014 epithelial cell abnormalities and its categories correlate well with biopsy diagnosis

**Keywords:** The Bethesda system, Negative for Intra Epithelial Lesion Cases, Abnormal cervical cytology Cases

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### Introduction

The humanity at large is thankful to a simple innovation done by Dr. George Papanicolaou who applied the test of collection of uterine cervical cells and studied its stained preparation in diagnosis of precursors of cancer and cancer lesions of uterine cervix. The cervical cytology over the past decades has improved through experiences in regards to collection technique, processing methods and diagnostic cytomorphological reporting[1]. Cancer of the cervix is the second most common cancer among women in the world. It accounts for 12% of all cancers in females[2,3]. The Pap smear is the

screening test which detects Precancerous and Cancerous lesions of Cervix and further a cervical biopsy is advised for confirming diagnosis and correlation[4]. The present study is carried out to evaluate the pattern of cervical Pap smear cytology at a tertiary care teaching hospital and to correlate it with histopathological findings.

#### Objective

- To assess the Bethesda system (TBS 2014) as a reporting system for cervico-vaginal cytology in detection of abnormal cervical cytology diagnostic categories.
- To compare the results of individual diagnostic cervical cytology categories with histopathological equivalents in available biopsies of cervix.

#### Materials and Methods

The present study is a retrospective study conducted in tertiary care rural teaching hospital. The study was conducted in a period of 5 years from 2015 to 2020. A total of 3596 patients underwent cervical cytology by Ayres wooden disposable spatula at Sharda Hospital during this period. The Inclusion criterion in the present study were

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the females who had undergone cervical scrape cytology as screening procedure or indicated because of symptoms, signs and clinical examination findings, related to cervical lesions. The exclusion criterion for the present study were - Women below age of 20 years, Pregnant women, Women with previous hysterectomy performed, Vaginal Vault Cytology, Women who have undergone previous major pelvic surgery, Cases with radiation.

**Ethical Approval:** Ethical Approval was taken from the Institutional Ethical Committee after explaining them the objectives of the Study.

**Informed Consent:** A written Informed Consent in bilingual language ie English/ Hindi was obtained from each patient or their guardian.

Preliminary data was collected as follows- name, MRD No, age, OPD/Ward, complete obstetrical and gynaecological History, per speculum examination of the cervix and vagina, findings of colposcopy whenever available, notation of pelvic sonography finding whenever available and basic haematologic investigations.

Cervical cytology samples were collected by conventional technique. The Ayres disposable wooden spatula was used for collecting sample. The cervical cytology smears were fixed in 95% alcohol for an hour and were stained by established method of Papanicolaou staining[5].

The Bethesda System(TBS) 2014 was adopted as a method of diagnosis, classifying cytomorphology Cervical Smears[6]. The Koilocytes were defined by demonstration of cytopathic effect of HPV virus with characters of koilocytosis, parakeratosis, smudged nuclei and multi nucleation[7].

The available biopsies of the cervix from the NILM cases and cases of abnormal cervical cytology of the study were processed by the conventional method of tissue processing[8]. These biopsies were stained by H & E Stain[9].

The biopsies were reported for their histomorphological diagnosis with reference to standard text book available on the topic[10].

The assessment of the each category diagnosed by TBS 2014 was compared with the histopathological diagnosis in the available

biopsies of the cervix. The assessment of diagnosis in the individual category of the TBS 2014 was done in the light of its equivalent histopathological diagnosis. The values of comparison of TBS 2014 was thought out considering histopathology as a gold standard of diagnosis.

**Observations**

In a total of 3,596 patients, who underwent the cervical cytology during the study period, total number of cases selected for the study were 199. A total of 99 cases were diagnosed as abnormal cervical cytology and 100 patients were picked up as age match control which were diagnosed as Negative for Intra Epithelial Lesion or Malignancy on Pap smear cytology.

The commonest complaint in 99 cases diagnosed as abnormal cervical cytology had no discharge (25 cases) and the symptom of white discharge (58 cases) and blood mixed discharge (16 cases) The 100 NILM cases were negative for intra epithelial lesion cytology had the commonest complaint as the white discharge. No history regarding sexual promiscuity and STD could be retrieved from the patients.

The maximum parity of the patients with abnormal cervical cytology group observed was 4 and control cases had maximum parity recorded also 4.

The per speculum exams of the cervix in 99 cases had the finding of healthy(2 cases), erosion(20 cases), congestion(23 cases), ulceration (42 cases), erythroplakia (5 cases), leukoplakia (2 cases) and growth (5 cases). The per speculum examination of the cervix in 100 cases of control chiefly had healthy (70 cases), congestion (30 cases). The cases recruited as control were not showing ulceration or growth.

The percentile prevalence of abnormal cervical cytology was 2.75% in total of 3,596 cervical cytology performed during the period of study.

The age distribution for the cases of abnormal cervical cytology and of NILM cases is tabulated in Table 1.

**Table 1: Age Distribution Of Abnormal Cervical Cytology(Acc) Cases And Nilm Cases**

Study	Age Distribution (Yrs)				
	21-30	31-40	41-50	51-60	61 & Above
CASES (n -99) (ACC)	03	27	26	25	18
CASES (n – 100) (NILM)	04	31	29	23	13

The maximum number of cases of the abnormal cervical cytology were in age group of 31-40 years. This age group denotes the fertile years of the age in women. The Adequate cellularity could be obtained by Ayres disposable wooden spatula in all cases of abnormal cervical cytology which full filled the adequacy criterion of cellularity of TBS 2014[6].

The Papanicolaou stain done in 99 cases of abnormal cervical cytology and 100 cases of control revealed adequately appreciable cytomorphological features to categorize them in TBS 2014.

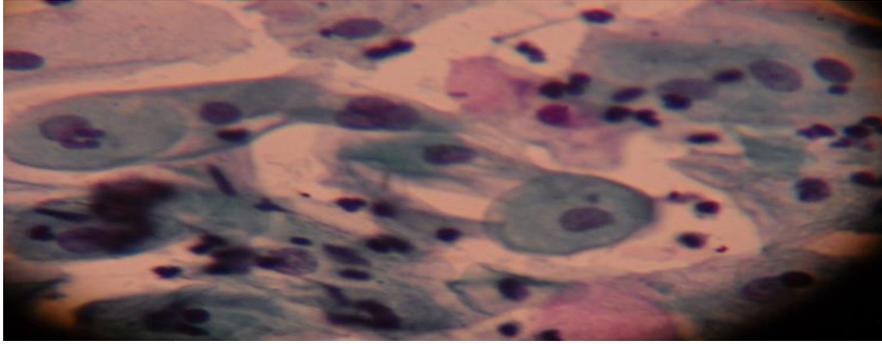
The following is the distribution of categorization and classes of the cases diagnosed as Abnormal Cervical Cytology (Table 2).

**Table 2: Classification Of Abnormal Cervical Cytology Cases According To Tbs 2014**

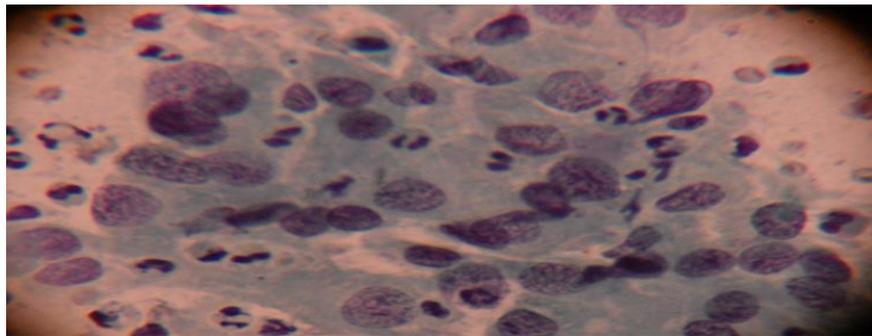
Cytology Diagnosis as Per TBS 2014	Number of Cases	Percentage
Satisfactory for evaluation	99	
Epithelial Cell Abnormality	99	
Atypical squamous cells of undetermined significance (ASC-US)	64	64.64%
Atypical squamous cells cannot exclude HSIL (ASC-H)	05	5.05%
Low-grade squamous intraepithelial lesion (LSIL)	14	14.14%
High-grade squamous intraepithelial lesion (HSIL)	05	5.05%
Squamous cell carcinoma (SCC)	10	10.10%
Atypical glandular cells (AGC) (Endocervical)	01	1.01%

Of 99 cases of Abnormal Cervical Cytology detected in the period of study; 69 were ASC of which ASC-US were 64 while ASC-H were 5. The second frequent abnormal cervical Cytology diagnosed was LSIL which constituted 14 cases, while 5 cases of HSIL were observed. The Squamous Cell Carcinoma was diagnosed in 10 cases

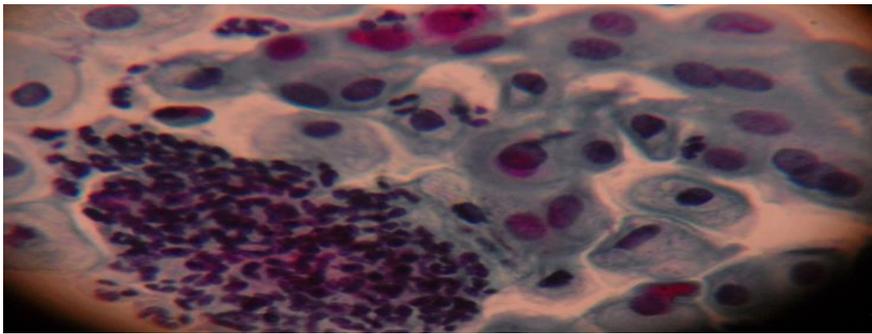
(Keratinizing Variant – 7, Non Keratinizing Variant- 3) . There was 1 case of Atypical Glandular Cells. The common cyto morphological features noted with various categories of epithelial cell abnormalities were as follows-



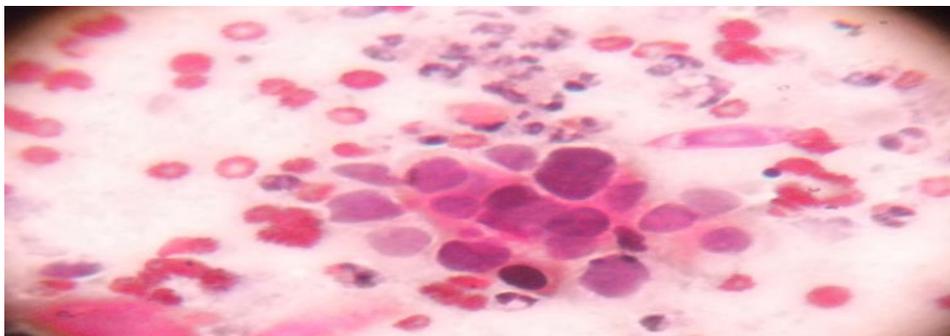
**Photomicrograph 1: Cervical Cytology- Interpretation- ASC-US, Type of Preparation- Conventional (40 X, Pap Stain), Cytomorphology- Slightly enlarged nuclei, multinucleation and small cytoplasmic peri nuclear halo**



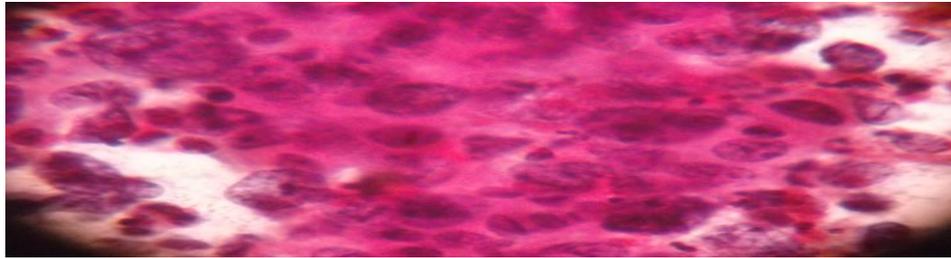
**Photomicrograph 2: Cervical Cytology- Interpretation- ASC-H, Type of Preparation- Conventional (40 X, Pap Stain), Cytomorphology- Loosely cohesive metaplastic cells with increased N/C ratio and chromatin granularity**



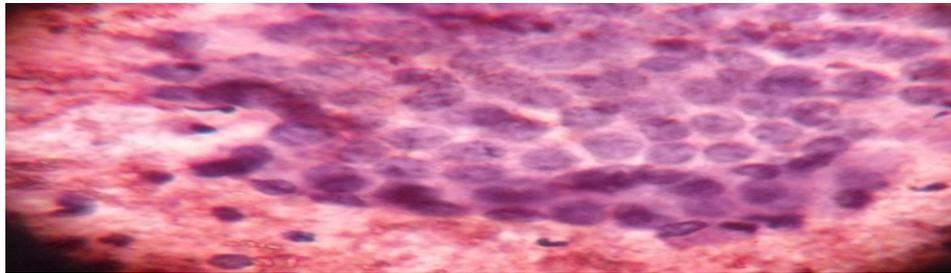
**Photomicrograph 3: Cervical Cytology- Interpretation- LSIL, Type of Preparation- Conventional (40 X, Pap Stain), Cytomorphology- Nucleus shows enlargement, hyperchromasia and koilocytotic change.**



**Photomicrograph 4: Cervical Cytology- Interpretation- HSIL, Type of Preparation- Conventional (40 X, Pap Stain) Cytomorphology- cells with high nucleus to cytoplasmic ratio, irregular nuclear membrane and hyperchromasia.**



**Photomicrograph 5: Cervical Cytology- Interpretation- SCC, Type of Preparation- Conventional (40 X, Pap Stain) Cytomorphology- Keratinized cells with marked nuclear pleomorphism, coarse chromatin condensation.**



**Photomicrograph 6: Cervical Cytology- Interpretation- AGC, Type of Preparation- Conventional (40 X, Pap Stain) Cytomorphology- loose aggregates of cells with round nuclei with variability in size , granular chromatin, nucleoli and mild nuclear membrane irregularity.**

The Age wise distribution of Abnormal Cervical Cytology is Tabulated in Table 3-

**Table 3: Age Wise Distribution Of Abnormal Cervical Cytology**

Abnormal Cervical Cytology	AGE DISTRIBUTION (Yrs)				
	21-30	31-40	41-50	51-60	61& Above
ASC					
ASC-US	02	18	15	17	12
ASC-H		02	03		
LSIL		03	03	03	05
HSIL		01	03	01	
SCC	01	02	02	04	01
AGC (Endocervical)			01		

The maximum cases of ASC-US were found in age group of 31-40 years. The maximum cases of ASC-H were seen in age group of 41-50 years. The maximum cases of LSIL were seen in age group of 61 years and above. The maximum cases of HSIL were seen in 41-50 years. The maximum cases of SCC were seen in age group of 51-60 years. A single case of AGC (Endocervical ) was seen in age group of 41-50 years.

The Koilocytes with all defined characters were observed in 7 cases amongst 99 cases of Abnormal Cervical Cytology. They were observed in all 5 cases diagnosed as HSIL, 1 case of Squamous Cell carcinoma and 1 case of LSIL. The following table No. 4 shows the distribution of Koilocytes –

**Table 4: Distribution Of Koilocytes Across Abnormal Cervical Cytology Cases**

Total number of Koilocytes	LSIL Category	HSIL Category	SCC Category
07	01	05	01

The biopsies of cervix were available in 23 cases which were diagnosed as abnormal cervical cytology (n- 99) and 18 cases of the Negative for Intra Epithelial Lesion or Malignancy (n-100).The

comparison between Histopathology diagnosis and Cytology diagnosis is tabulated in Table 5

**Table 5: Comparison Between Histopathology And Cytology Diagnosis**

Cytology Diagnosis & Number of Cases	Number of Cases Undergone Biopsy	Histopathology Diagnosis				
		Chronic Cervicitis	CIN 1	CIN 2	CIN 3/ Carcinoma in Situ	SCC
Negative for intraepithelial lesion or malignancy (Total 100 cases)	18	18	---	---	---	---
Abnormal Cervical Cytology (Total 99 cases)	23					
ASC						
ASC-US (64 cases)	01	01	---	---	---	---
ASC-H (5 cases)	05	01	03	01	---	---
LSIL(14 cases)	05	---	05	---	---	---
HSIL(5 cases)	05	---	---	02	03	---
SCC(10 cases)	07	---	---	---	---	07
AGC (1 Case)	---	---	---	---	---	---

The categories of TBS 2014 appeared to correlate in all its categories except for ASC-H where in two cases showed the higher lesion. There were no false reporting of the higher lesions (False Positive) when TBS 2014 was applied in the present study. However there were 4 cases in ASC-H category where under reporting of higher lesions has occurred (3 cases of CIN 1, 1 case of CIN 2). No case diagnosed as NILM by TBS 2014 in present study turned to carry higher lesion on the biopsy. The 100% correlation was observed for categories of LSIL, HSIL, Squamous Cell Carcinoma.

#### Discussion

The common Gynaecological symptoms for which women seek gynaecologic advice is a white discharge, followed by blood mixed discharges. These symptoms are commonly associated with infections and abnormal cervical cytology[11]. The present study also observed the same pattern in which white discharge, followed by the blood tinged discharge was a common symptom for consultation by gynaecologist in the cases with abnormal cervical cytology. The per speculum examination in 99 cases of abnormal cervical cytology in the present study had common abnormal finding of erosion, congestion, ulceration and growth. The higher incidence of the abnormal cervical cytology with the gross lesions on the cervix has also been recorded in a few studies[12]. It has been documented in three studies[13-15] that the abnormal cervical cytology can be detected by cervical scrape cytology in the wide age group of 18 to 70 years and no age group is a limiting factor for abnormal cervical cytology. The observations in the present study for the age in the group of abnormal cervical cytology cases is concurrent to above studies. The two studies[15,16] have located the maximum abnormal cervical cytology on the cervical scrapes in range between 31 to 60 years. Similar observations to above study has been made in our study in regards to occurrence of abnormal cervical cytologic findings in regards to age group distribution. The present study had 99 cases of abnormal cervical cytology that ranged in age group of 31- 60 years. Chheing et al[17] did sampling of cervical cytology by Ayres spatula and recorded the satisfactory sampling of cellularity by its use. These studies did not come across the limitations of Ayres spatula over the brush sampling of the cervix, to meet the cellularity adequacy criterion of TBS. The present study sampled cervix by Ayres disposable wooden spatula and also found no limiting factor in obtaining adequate cellularity that full fills the adequacy criterion of TBS similar to the above studies. The studies of Chheing et al[17] gave the overall distribution of abnormal cervical cytology by TBS as follows- NILM- 80.9%, ASC- 10.32%, AGC- 0.27%, LSIL- 4.54%, HSIL-3.44%. The present study for the distribution of the diagnostic category of abnormal cervical cytology is lower to the findings of the above study. The present study recorded NILM-97.24%, ASC-1.91%, LSIL-0.38%, HSIL-0.13%, SCC-0.27% in total of 3596 cervical cytology cases screened in the period of study. However category of ASC predominated over the distribution of abnormal cervical cytology followed by LSIL, HSIL and SCC in descending order which is similar to observation of Chheing et al[17]. Pan et al[15] reported the presence of Squamous Intra Epithelial Lesions(SIL) and Squamous Cell Carcinoma(SCC) in wide age range of 30 to 65 years and above. The present study observed the similar distribution of the age for the SIL's and SCC. The conclusive age distribution of the lesions could not be established by observations of the study of SIL and SCC for the age similar to above studies. The cases of abnormal cervical cytology in regard to ASC, LSIL, HSIL, SCC were distributed across the age range of 31 to 60 years and above. A specific cytomorphologic feature of HPV infection in the cervical cytology recorded is the presence of Koilocytes in the smear because of this consistent finding of cytopathic effect of HPV virus in the epithelial cells, these cells have been retained to suggest the SIL in TBS. The studies[7,14,18]. have reported high correlation between morphological alteration of Koilocyte and the positive result of HPV detection in the cervical smear. Cramer et al[14] observed the retrieval of HPV DNA in all

smears which had koilocytes being present. Matsuura et al [7] observed this consistency of koilocytosis, parakeratosis, smudged nuclei, multi nucleation in the cells associated with HPV infection. The present study is in concordance with the studies of the aforesaid authors and observed Koilocyte in 7 cases amongst 99 Abnormal Cervical Cytology cases. The present study in NILM category had biopsy available in 18 of 100 cases which did not show any high grade lesion and corresponded well with histopathologic diagnosis (100%). There were 6 biopsies available in category of ASC (ASCUS=1, ASC-H= 5). The ASC-H category showed 1 case on biopsy diagnosed as CIN 2 and three cases as CIN-1. Though the biopsy follow up for this category was not high but the observations pertaining to ASC-H category of present study are similar to Chieng et al[17] as ASC-H is the category of TBS which on biopsy follow up has resulted in SIL. The present study had 14 cases of LSIL of which 5 were available for biopsy correlation and all were corresponding to histopathological diagnosis of CIN-1 (100%). There were 5 biopsies of 5 HSIL cases available for correlation, all 5 cases had a consistent findings of CIN 2 (2 cases), CIN 3(2 cases), CIS (1 cases). There were 7 cases of SCC who were followed up by biopsy revealed the 100% correlation. The observations in the present study for biopsy follow up in categories of LSIL, HSIL, SCC are in concordance with many studies[17-24]

The present study observed a positive impact of TBS in respect to its utility as the criteria of satisfactory cellularity, uniformity of nomenclature, clarity of categorization, reduction in equivocality of cytomorphologic classes and corresponding equivalents of histopathology. These observations in general have also been seen by many studies [17,20,22,23]

#### Conclusion

- The TBS 2014 system for reporting cervical cytologic diagnosis is of high value in detecting abnormal cervical cytology and is helpful in suitable categorization of epithelial cell abnormality.
- The TBS 2014 epithelial cell abnormalities and its categories correlate well with biopsy diagnosis done by CIN (Cervical intra epithelial neoplasia) grading.
- Koilocytes as a cytopathic change of HPV is diagnostic of HPV infection and is found on Cervical cytology smears and its identification should prompt further ancillary studies. Findings of these studies will help in the work up of cervical cancer screening programme carried out at various levels of medical services.

#### References

1. Koss, Leopold G, Melamed, Myron R. Squamous Carcinoma of the Uterine Cervix and Its Precursors, IN Koss' Diagnostic Cytology and Its Histopathologic Bases. (Ed) Koss, Leopold G; Melamed, Myron R, Lippincott Williams & Wilkins, 5th Edition, 2006, 283-312.
2. Urasa M, Darj E. Knowledge of cervical cancer and screening practices of nurses at a regional hospital in Tanzania. Afr Health Sci. 2011;11:48-57.
3. Ferlay J, Shin HR, Bray F, Forman D, Mathers C, Parkin DM. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. Int J Cancer. 2010;127:2893-917
4. Likhari KS, Saluja A, Gupta SG, Hazari RA, Likhari SK. Precancerous and cancerous lesions of cervix diagnosed by Pap's smear – A hospital based study. J Evol Med and Dent Sci. 2014;3:1899-904.
5. Bales C. Laboratory Techniques IN Koss' Diagnostic Cytology and Its Histopathologic Bases. (Ed) Koss, Leopold G; Melamed, Myron R, Lippincott Williams & Wilkins, 5th Edition. 2006, 1592-5.
6. Nayar R, Wilbur DC. The Pap Test and Bethesda 2014. J Low Genit Tract Dis. 2015;19(3):175-84.

7. Matsuura Y, Kawagoe T, Toki N, Sugihara K, Kashimura M. Low Grade Cervical Intraepithelial Neoplasia Associated with Human Papillomavirus Infection, Long-Term Follow-up, ActaCytol. 1998;42(3):625-30.
8. Gamble M. The Hematoxylin and Eosin in Theory and Practice of Histological Technique.(Ed) John. D.Bancroft, Marilyn Gamble, Churchill Livingstone Elsevier, 6th Edition, 2008, 121-34.
9. Rosai J. Uterus-Cervix in Female Reproductive System, Rosai and Ackerman's Surgical Pathology, (Ed) Juan Rosai, Elsevier, Tenth Edition. 2011; 2:1436-76.
10. Ellenson L, Pirog E. The Female Genital Tract in Robbins and Cotran Pathologic Basis of Disease.(Ed) Kumar V, Abbas A K, Fausto N, Aster J, Saunders Elsevier, Philadelphia, 8th Edition, 2010, 1017-24.
11. Heider A, Austin R, Zhao C. HPV Test Results Stratify Risk for Histopathologic Follow-Up Findings of High-Grade Cervical Intra-Epithelial Neoplasia in Women with Low-Grade Squamous Intra-Epithelial Lesion Pap Results, ActaCytologica. 2011;55:48-53
12. Howell L, Zhou H, Davis R. Significance of Subclassifying High grade Squamous Intraepithelial Lesions into Moderate dysplasia/CIN II Versus severe dysplasia/ CINIII/CIS in the Bethesda System terminology, Diagn Cytopathol. 2004;30(5):362-66.
13. Nasuti J, Fleisher S, Gupta P. Atypical Glandular cells of undetermined significance(AGUS): Clinical Considerations and Cytohistologic correlation. Diagnostic Cytopathol. 2002;26: 186-90.
14. Cramer H, Skinner-Wannemuehler S, Brown D, Katz B, Fife K. Cytomorphologic Correlates of Human Papillomavirus Infection in the "Normal" Cervicovaginal Smear, ActaCytol. 1997;41:261-68.
15. Pan Q, Belinson J, Li L, Pretorius R, Qiao Y, Zhang W, Zhang X. A Thin-Layer. Liquid-Based Pap Test for Mass Screening in an Area of China with a High Incidence of Cervical Carcinoma: A Cross-Sectional, Comparative Study, ActaCytol. 2003;47(1): 45-50.
16. Kitchener HC, Almonte M, Wheeler P, Desai M, Gilham C, Bailey A, Sargent A, Peto J on behalf of the ARTISTIC Trial Study Group. HPV testing in routine cervical screening: cross sectional data from the ARTISTIC trial, British Journal of Cancer. 2006; 95:56-61.
17. Chhieng D, Roberson J, Gidley J, Eltoum I. Bethesda 2001, Impact on the Reporting of Gynecologic Cytology, Acta Cytol. 2004;48(3):355-62.
18. Basu S, Mitra PK, Roy A, Chatterjee R. Detection of human papillomavirus in cervical swabs from Indian women by cytological and immunocytochemical technique, Neoplasma. 1991; 38(6):639-44.
19. Emerson R, Puzanov A, Brunner C, Younger C, Cramer H. Long term follow up of women with Atypical Squamous Cells of Undetermined Significance, Diagn Cytopathol. 2002;27(3): 153-57.
20. Howell L, Zhou H, Wu W, Davis R. Significance of Subclassifying High grade Squamous Intraepithelial Lesions into Moderate dysplasia/CIN II Versus severe dysplasia/ CINIII/CIS in the Bethesda System terminology, Diagn Cytopathol. 2004;30(5):362-66.
21. Nieh S, Fu E, Gau C. Is p16IN K4A expression more useful than human papillomavirus test to determine the outcome of atypical Squamous cells of undetermined significance-categorized Pap smear? A comparative analysis using abnormal cervical smears with follow up biopsies, Gynaecologic oncology. 2005; 97:35-40.
22. Kaveri SB, Khandelwal S. Role of Pap smear in cervical biopsy in unhealthy cervix. J SciInnov Res. 2015;4:4-9.
23. Susic MG, Babic G, Dimitrijevic A, Mitrovic S, Varjadic M. Correlation between cervical cytology and histopathological cervical biopsy findings according to the Bethesda system. Ser J Exp Clin Res. 2014;15:205-16
24. Kulkarni Padmaja R, Hephzibah R. Cytohistological Correlation Study of Conventional Papanicolaou Smears in Cervical Neoplasia. Journal of Research in Medical Education & Ethics. 2013;3(2):172-79.

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