



Research Article

A Comparative Study of Ultrafast Papanicolaou Staining with Standard Papanicolaou Staining Technique for the Assessment of Cervical Smear – A Hospital Based Observational Study

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ABSTRACT

This prospective study was conducted to assess the use of ultrafast Papanicolaou stain for cervical smears. Study included 60 cervical smear cases. In each case, two smears were prepared and stained by UFP and standard Papanicolaou stains. The four parameters background, cell morphology, nuclear details and overall staining has been considered. Quality index was calculated from ratio of score achieved to maximum possible score. Comparison is done between the two staining techniques on the basis of these cytomorphological features. It was observed that UFP-stained smears had clear RBC free background, crisp nuclear chromatin, well-stained nucleoli, and transparent cytoplasm. Hence, it was concluded that UFP stain is useful for rapid diagnosis for cervical smear which is time saving and helpful.

Keywords: conventional Papanicolaou stain; ultrafast Papanicolaou stain, Cervical smear, time.

OBJECTIVE OF THE STUDY:

To assess the staining quality and compare the efficacy of ultra-fast Papanicolaou stain with the conventional Papanicolaou stain in cervical smears.

Justification for the study: Among women, cervical cancer is the second most common type of cancer. Screening techniques that identify cervical cancer at an extremely early stage can help avoid it. A straightforward, practical, affordable, and accurate test for quickly identifying cervical abnormalities is the cervical Pap smear. Globally, the incidence and mortality of invasive cervical cancer have drastically decreased since its introduction. For both gynecological and non-gynecological cytology smears, Pap stain is the recommended stain. A routine Pap smear can identify precancerous cells in the cervix.

Standard Papanicolaou stain yields polychromatic transparent staining reaction with crisp and distinct nuclear and cytoplasmic features. It was first developed by Dr. George Nicholas Papanicolaou in 1942 to know the variation in cellular maturity and metabolic activity in vaginal smears. Pap stain clearly distinguish between basophilic and acidophilic cell components and gives the detailed chromatin pattern making the nuclear details very good.

Standard Pap stain is time consuming and takes 20- 30 minutes. As practiced conventionally Standard Pap stain use a substantial quantity of alcohol which hinders its use as a mass screening tool in low resource settings. It takes a long time to complete the staining. Since its evolution, the pap stain has undergone various modifications.

Pap stain has evolved significantly throughout time. With the need for minimal turn around time for assessing the cervical pap smears has encouraged innovations in staining procedures that require lesser staining time with equivocal cell

morphology. Modifications have been developed in pap stain to improve the staining quality and fixation in alcohol formalin. and to minimize staining time

Ultrafast papanicolaou (UFP) stain was introduced by Yang and Alvarez in 1995. UFP stain is a hybrid of the technique by Romanowsky and conventional pap stain to reduce the staining times to less than 2 minutes. It incorporates principle of air drying of cells followed by rehydration in normal saline. Air dried smears stained by Romanowsky offer nuclear opacity and wet fixed (alcohol fixed) offer transparent crisp nuclear features giving accurate nuclear details. Air dried smears make cells appear larger and thus increase the resolution for analysis of cellular details. Normal saline is to rehydrate the cells so that transparency is regained in addition to hemolysis of bloody background . Alcohol formalin ph 5 bring out the vibrant colours in the cells and nucleoli which stains red. The entire procedure is fast enough to permit immediate microscopic assessment of cervical smears.

The main motive for UFP stain is to reduce turnaround time and speed up reporting , which saves time and improves the staining quality. The necessity for Pap stain alterations originates from the requirement to examine the cervical smears in a short period to limit the occurrence of invasive cervical malignancy.

METHODOLOGY:

This is a hospital based study conducting in Nalbari Medical College and hospital over a period of 6 months. Sample size of study is 60 which is statistically calculated . Cervical smears are received in the Department of pathology , Nalbari Medical College and hospital. 2 slides from each sample will be made . One slide is stained with Conventional Pap and the second one is stained by UFP stain.The staining procedure of UFP stain and SP stain is important.

The reporting has been doing using The Bethesda System 2014 of reporting cervical smears. The four parameters background, cell morphology, nuclear details and overall staining has been considered. These findings and interpretation will be noted separately for UFP stain and SP stain . Cytomorphological features is the key taken into account while doing comparison. Data is then entered in Microsoft office excel 2003 ,subsequently analyzed using Statistical Package for the Social Sciences- 25 (SPSS 25)

Inclusion criteria:

All the gynaecological cases considering the eligibility criteria aged 18 to 49 years of age with absence of total hysterectomy and willing to give consent attending Department of Pathology , Nalbari Medical college and hospital to undergo PAP smear test with complaints of bleeding per vagina, passage of discharge and cervical lesions as well as having adequate material in the cervical smears made from each sample..

Exclusion criteria:

- Those who are not willing to give consent
- Cases with smears having inadequate material.

Possible Risks: no risk.

Possible benefits:

Turn around time of test results is characteristically reduced in ULTRA Fast Pap staining procedure. The speed of reporting cervical Pap smears can be improved by rapid assessment of smears. Quick diagnosis plays an important role in efficient medical practice. In an outpatient setting , a quick reporting allows clinician to discuss further lines of action or management options with patience during the very first visit, which is of great benefit for both physicians and the patients.

RESULTS:

The quality of UFP smears were better when compared to conventional PAP for cervical smears and was statistically significant.The data collected are compiled ,tabulated and subjected to statistical analysis.Results are presented in terms of percentage and mean +_SD.The reporting has been done using the Bethesda System 2014 of reporting cervical smears. For assessment of UFP stain, scores were given on four parameters: background of smears, overall staining pattern, cell morphology, and nuclear staining, Diagnosis made by UFP stain was compared with standard PAP in all cases.Data are entered in Microsoft office excel 2003 and then analysed using statistical package for the social sciences -25(SPSS 25). QI of UFP stain for cervical smears was 0.94. Excellent quality of stain was noted in most of the cases allowing easy diagnosis. In very few cases diagnosis was possible with some difficulties. In all cases of smears, UF-PAP stain gave a good score for the background, nuclear staining, cell morphology, and overall staining in comparison with standard PAP method.

Table 1:Age distribution among the study objects

AGE GROUP	NUMBER OF PATIENTS(%)
21-30	30(50%)
31-40	20(33.3%)
41-50	10(16.6%)

TOTAL	60
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Table 2: PRESENTING CLINICAL SYMPTOMS

SYMPTOMS	NUMBER OF PATIENTS(%)
ITCHING	20(33.3%)
WHITISH DISCHARGE	30(50%)
BLEEDING	10(16.6%)
BACKACHE	10(16.6%)

Table 3: Background comparison among both stains

BACKGROUND	ULTRAFASST PAP		STANDARD PAP	
	NO OF CASES	PERCENTAGE	NO OF CASES	PERCENTAGE
HEMORRHAGE	0	0	40	65
NON HEMORRHAGE	60	100	20	35
TOTAL	60	100	60	100

Table 4: OVERALL STAINING OF BOTH THE STAINS

Background	Ultra fast PAP		Standard PAP	
	No of cases	%	No of cases	%
Good	42	70	30	50
Moderately good	28	30	30	50
Bad	0	0	0	0
Total	60	100	60	100

Table 5: Nuclear characteristics comparison among both the stains

	Ultra fast PAP		Standard PAP	
	No of cases	%	No of cases	%
Smudge chromatin	45	75	50	83
Moderately crispy chromatin	10	16.6	5	8.3
Crispy chromatin	5	8.3	5	8.3
Total	60	100	60	100

characteristics	Ultrafast stain(Mean +_SD)	PAP STAIN(Mean +_SD)	P-VALUE
Background of smears	2.01+ 0.00	1.38+ .42	0.00
Staing pattern	2.58+ .42	2.44+ .51	0.00
Cell morphology	2.94+ .22	2.84+ .32	.12
Nuclear staining	2.90+ .22	2.78+ .32	.02
Cumulative score	10.43+ .52	9.44+ .62	0.00
Quality index	.94	.88	0.00

CONCLUSION-

Our study concluded that UFP stain can be applied on a regular basis to offer immediate diagnosis in cervical smears. UFP stain is reliably a rapid and useful diagnostic tool and can be applied on a regular basis in cervical smears to offer ready to go diagnosis.

The findings of this study support the use of UF-PAP method in cytology laboratory with a high emphasis on cervical smears. UFP stain in comparison to routine PAP, provides an excellent and suitable alternative in cytological staining for the study of cervical smears considering all the features studied. UFP is fast, reliable and can be done with locally available reagents with unequivocal morphology which is the crucial for diagnostic set up.

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