Original Article

A Comparative Study of Atypical Squamous Cells of Undetermined Significance in Pap Smears Followed by Biopsy Results for Determination of Reliable Cytologic Parameters

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ABSTRACT

***Background and Objective*:** Microscopic evaluation of cervicovaginal smears (Pap smear) plays an essential role in detection of preneoplastic and neoplastic lesions of uterine cervix. A wide spectrum of changes is seen in Pap smears. Interpretation of intermediate changes which resulting in observation of so- called ASCUS is difficult. ASCUS is defined as a cellular change that is more marked than typical reactive change but lack features diagnostic of intraepithelial neoplasia. The aim of this study was determination of reliable cytological parameters to identifying real nature of ASCUS.

***Patients and Methods*:** In this descriptive, cross sectional study, all Pap smears with presence of ASCUS, which followed by biopsy samples in Urmia, Shahid Matahari hospital during March 1999 to December 2002 were reviewed. In this regard, 17 parameters of cytoplasm, nuclei and smear background were considered and data were analyzed by SPSS and Chi Square test.

***Results*:** Four parameters of irregular nuclear contour, increased nucleo- cytoplasmic ratio, cytoplasmic vacuolization and dirty background are highly suggestive of dysplasia.

***Conclusion*:** It is important to consider that to find out the real nature of ASCUS a group of parameters are more helpful than a single one. In addition, a single Pap smear is not enough for this purpose and further investigation is required.

Keywords: Papanicolaou test, Cervix Dysplasia, Cancer, Iran

Introduction

Microscopic evaluation of cervicovaginal smears (Pap smear) plays an essential role in the detection of the preneoplastic and neoplastic lesions of uterine cervix (1). In this regard, the important problems are 1)Targets of population screening (certain groups of woman who are at high risk for cervical cancer; 2) Correct technique of sampling; 3) Proper staining procedures; and 4) Better understanding of laboratory interpretation by clinicians (1).

A wide spectrum of changes (benign to malignant) is seen in Pap smears. Interpretation of intermediate changes which resulting in observation of cells named as Atypical Squamous Cells of Undetermined Significance (ASCUS) in Bethesda system is difficult

(1). ASCUS is synonymous with Papanicolaou's 'karyomegaly' and defined as a cell with nuclear enlargement 2-3 times the size of nucleous of a normal intermediate cell (1). In 2001, the Bthesda system recommended the term "atypical squamous cell (ASC)" instead of ASCUS and categorized it into two subclasses: ASC-US, which stands for ASC-unknown significance, and ASC-UH, which suggests a high grade SIL (squamous intraepithelial lesion) (2). ASCUS was found in almost 5% of Pap smears done for screening (3, 4). It is no wonder that the biopsies of about one third of patients with ASCUS reveal neoplastic lesions. About 30% of these neoplastic lesions are high grade and about 70% are low grade (1). Really ASCUS is defined as cellular changes that are more marked than typical reactive changes but that qualitatively or quantitatively lack features diagnostic of Squamous Intraepithelial Lesion (SIL) (5). So attempts have been made to divide ASCUS into two groups: one probably benign and the other possibly malignant (1).

Biopsy samples of 104 cases with ASCUS found on Pap smears in Tehran University of Iran revealed 30(28.8%) SIL, 1 invasive carcinoma, and 1 endometrial carcinoma. The repeated Pap smear in 60 women with ASCUS before colposcopy showed ASCUS in 45(75%) cases and SIL in 8 (17.7%) cases. The result was normal in 7 cases. Among these 7 normal cases histological examination showed 2 cases of LSIL (2).

Histological examination of biopsy samples of 17 from 99 patients whose Pap smears showed only atypical cells in Colorado University revealed dysplasia (6). Likewise, dysplasia was seen in 41% of patients with presence of atypical cells in Pap smears in Arkansas University (7). Another study in California revealed SIL in biopsy samples of 29.1% of cases with ASCUS in Pap smears (8).

In one study in Canada, cytological parameters of smears with presence of ASCUS, reviewed in cases whose their biopsies showed Cervical Intraepithelial Neoplasia (CIN).Two parameters of clear perinuclear space and moderate nuclear atypia were associated with CIN I and four parameters of clear perinuclear spaces, hyperchromasia, moderate anisokaryosis and increased nuclear volume of metaplastic cells were predictors of CIN II and CIN III (9).

Another study showed CIN in 50% of nonpregnant women with ASCUS and in 33.3% of obstetric patients who had biopsies within the first 6 months postpartum (10).

Results of an investigation in Chicago indicated that the degree of agreement between cytological abnormalities reported using the Bethesda system for cervicovaginal diagnosis and underlying cervical lesions is statistically significant but weak. This study showed false positive rates decrease with severity of the lesions have been reported in Pap smears. Finally, these results suggested that the reliability of a single smear is poor and serial testing is required to exclude abnormality (11, 12). Therefore, follow up of the patients with ASCUS is as serious as SIL (13).

The goal of our study was determining the reliable cytological parameters to identifying real nature of ASCUS.

Material and Methods

This study was a descriptive, cross sectional one. All Pap smears with presence of ASCUS, which their following biopsy samples were available, were reviewed during an almost four year's period (March 1999 to December 2002). The result of Pap smear report was compared with histopathology diagnosis. In this regard, 17 parameters of cytoplasm, nuclei and smear background were considered for each case and the obtained data were analyzed by SPSS and Chi square test. Sensitivity, specificity, positive and negative predictive values were determined for each parameter with consideration of its histopathology diagnosis which was Reactive changes, dysplasia or malignancy.

It is important to note that only some of these criteria had been considered in each of previous studies (1, 6, 9).

Results

The results obtained from statistical analysis are seen in Table 1. Accordingly irregular nuclear countor, increased nuclear to cytoplasmic ratio, cytoplasmic vacuolization and dirty background are important in differentiating reactive nature of atypical cells from dysplasia.

It has to be noted that we divided the number of atypical cells and number of nucleoli into small groups (rare, occasional, intermediate, and plenty) and (none, 1-2, 2-3, more than 3), respectively and because the number of cases located in each one were so few, therefore statistical evaluation was not significant and not carried out.

Table1: Sensitivity, specificity, positive and negative predictive values of parameters

parameter	Sensitivity (%)	Specificity (%)	Positive predictive value(%)	Negative predictive value(%)
Nuclear size	62.5	80.6	62.5	80.6
N/C ratio	87.5	16.1	35	71.4
Thick nuclear Membrane	18.8	93.5	60	69
Irregular nuclear Shape	56	96.8	90	81
Coarse chromatin	37.5	90.3	66.7	73.7
Multinucleation	56.3	80.6	60	78.1
Nuclear molding	12.5	93.5	50	67.4
Perinuclear halo	43.8	90.3	70	75.7
Nuclear vacuolization	50	77.4	53.3	75
Cellular adhesion	50	25.8	25.8	50
Cytoplasmic color	56.3	74.2	52.9	76.7
Cytoplasmic vacuolization	81.3	64.5	54.2	87
Hyperkeratosis	12.5	90.3	40	66.7
Parakeratosis	31.3	87.1	55.6	71.7
Dirty background	25	100	100	72.1

Discussion

In this study, biopsy samples of Pap smears with ASCUS showed reactive changes in 31 cases (65.9%), dysplasia in 12 cases (25.53%) and malignancy in 4 cases (8.51%) . Likewise, different academic studies showed dysplasia in 30% to 40% of biopsy samples with presence of ASCUS in Pap smears (1, 7, 8).

The most considerable aspects of this study are:

Irregularity of nuclear shape, increased nuclear to cytoplasmic (N/C) ratio, cytoplasmic vacuolization and dirty background are suggestive of dysplasia and /or malignancy with high sensitivity.

Rarity of atypical cells, enlargement of atypical nuclei up to 3 times of nuclei of intermediate cells, lower nuclear to cytoplasmic ratio, lack of thick nuclear membrane, absence of vacuolization, fine chromatin and lack of hyperkeratosis are suggestive of reactive lesions.

No acceptable statistical results obtained for nuclear molding, number of nucleoli, nuclear vacuolization, cellular adhesion and also parakeratosis.

The limitation of this study was relatively small number of cases. However, this study similar to study in Canada (9) indicated that the consideration of a group of parameters is more helpful than a single one.

However, since distinction between ASCUS and SIL can not be possible by cytopathology examination lonely, it seems that further investigation such as colposcopy or repeated Pap smear are necessary for managing cases with ASCUS (2). Some investigation advised colposcopic evaluation after a repeated Pap smear with ASCUS (14). Other studies recommended directed biopsy immediately because according to their research the differences in histological findings between the two groups (with repeated Pap smear before colposcopic biopsy and without it) were not significant (2, 4).

Conclusion

Following up of the patients with ASCUS whose smears show four criteria of irregular nuclear shape, increased N/C ratio, cytoplasmic vacuolization and dirty background are recommended emphatically.

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