RESEARCH ARTICLE

Clinical Factors Associated with Specimen Adequacy for Conventional Cervical Cytology in Thammasat University Hospital, Thailand

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Abstract

    <u>Purpose</u>: To study clinical factors related to adequacy of transformation zone (TZ) components in cervical smears. <u>Materials and Methods</u>: Medical and Papanicolaou (Pap) smear reports from Thammasat University Hospital, Thailand during January to December 2015 were collected. Demographic data was reviewed by attending physicians and impact of clinical factors on TZ adequacy was primary outcome. A total of 3,251 smears were reviewed. Finally, 2,098 smears met The inclusion criteria and enrolled into this study. <u>Results</u>: Average age and bodyweight of participants in this study were 43.0 years and 60.0 kg, respectively. Ninety seven percent of smears were classified as satisfactory for evaluation according to the Bethesda system 2001. Adequacy (group A) and inadequacy (group B) of TZ were equal in percentage (50.9/46.0). Prevalence of abnormal cervical cytology was 4.4%. Percentages of abnormal Pap smears in group A and B were 7.3 and 1.4, respectively (p<0.001). Factors associated with increased adequacy of TZ were old-age (≥ 50 yr), nulliparity, within 3-months postpartum, history of TZ inadequacy and abnormal smears. Sexually transmitted disease (STD), hormonal usage, previous cryotherapy and smears collected by staff were associated with inadequacy of TZ. <u>Conclusions</u>: Collection of cervical specimens should be carefully performed. STD history, hormonal usage and previous cryotherapy are risk factors for TZ inadequate specimens.

Keywords: Adequacy of transformational zone component - Pap smears

Introduction

    Cervical cancer is the second most common malignancy in Thai women with an age-standardized incidence rate of 18.1/100,000 women per year (Khuhaprema et al., 2012). The most widely employed screening method is Papanicolaou (Pap) smears (Ferlay et al., 2015).

    Inadequacy of transformation zone component (TZ) and unsatisfactory smears have a higher risk of progression to cervical cancer or a pre-cancerous lesion than adequacy of TZ (Zhao et al., 2014). Both conditions had a significantly increased likelihood of undergoing cervical biopsy. Barakat's work in the year 2013 showed that cervical smears satisfaction without TZ had a higher probability of obtaining a false-negative finding (Barakat et al., 2013). Inadequacy of TZ is the most common cause of poor quality smears (Siebers et al., 2012; Zhao et al., 2014). Frequent screening with low sensitivity tests or poor quality slide led to increasing healthcare costs (Nandini et al., 2013). Increased TZ adequacy will result in better effectiveness and precision of cervical cancer screening. Therefore, the primary focus of this research was to investigate the factors that resulted in inadequacy of transformation zone on Pap smears.

Materials and Methods

    This retrospective study was conducted between January and December 2015. Data was randomized and selected by computerized program from electronic database of patient records Thammasat University Hospital.

    Sample size of the study was calculated according to Siebers' literature (Siebers et al., 2012). Incidence of TZ inadequacy was 12.47% in the total population. Eight determinants and thirty events per determinants were estimated from the full model. Total of 240 events are needed. Sample size estimation was 1,924.61 cases. Therefore, the sample size of this study should be approximately 2,000.

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Specimen adequacy and cytological diagnoses were classified according to the Bethesda system 2001 (Solomon et al., 2002). The criteria of unsatisfactory smears were scanty epithelial cell (defined as less than an estimated 8,000-12,000 cells per slide) or more than 75% of the obscured epithelial cells by inflammation or blood. TZ inadequacy was defined as the smears without transformation zone component.

Inclusion criteria was all cases of conventional Pap smears done in the study peroid. Exclusion criteria were cervical cancer and hysterectomy history. Demographic data consisted of bodyweight, parity, pregnancy status, within 3-months postpartum, history of vaginal delivery, menopause status, history of abnormal Pap smear, leukorrhea, abnormal vaginal bleeding, history of STD, hormonal usage, history of precancerous lesion treatment, human immunodeficiency virus (HIV) infection, enlarged uterus, collection by trainee or staff and latest Pap smear results.

Multivariate binary logistic regression analysis was used to determine the association factors with TZ adequacy. A p value of less than 0.05 was considered statistically significant. Analyses were performed using the SPSS statistics version 17 (SPSS Inc., Chicago, USA).

Results

During the period of this study, 3,251 conventional smears were enrolled. A total of 1,153 smears were rejected from the study. Finally, 2,098 cases were selected for data analysis.

Average age and bodyweight of participants in this study were 43.0 years and 60.0 kg, respectively. Ninety seven percent of smears were classified as satisfactory for evaluation according to the Bethesda system 2001. Adequacy (group A) and inadequacy (group B) of TZ were similar in percentage (50.9/46.0). Prevalence of abnormal cervical cytology was 4.4%. Percentage of abnormal Pap smears in group A and B were 7.3 and 1.45, respectively (p<0.001). Detail of demographic data was presented in Table 1.

Factors that associated with increased adequacy of TZ were old-age ($\geq$ 50 yr), nulliparity, history of TZ inadequacy and abnormal smears. Sexually transmitted disease (STD), hormonal usage, previous cryotherapy and smears collected by staffs were reported with inadequacy of TZ.

Percentage of abnormal smears detection in group A was more than that of group B (7.3/1.45) with statistical difference (p<0.001). ASC-US smears was more frequently reported in group A. There was no abnormal cervical smears in group B subjects who were younger than 30 years old. The relationship between cytological reports and adequacy of TZ were shown in Table 2.

The relationship between clinical factors and adequacy of TZ smears were analyzed and shown in Table 3. Factors that associated with increased adequacy of TZ were old-age ($\geq$ 50 yr), nulliparity, within 3-months postpartum, history of TZ inadequacy and abnormal smears. STD,

Table 1. Demographic Data

Demographic data	Results
Age *	43.0 ± 15
BW *	60.0 ± 10.6
Result of Pap smears (N, %)	2098 (100)
NILM	2006 (95.6)
Abnormal Pap smears	92 (4.4)
Atypical	
ASC-US	62 (2.96)
ASC-H	5 (0.24)
Atypical glandular cells	2 (0.10)
LSIL	6 (0.29)
HSIL	3 (0.14)
Cancer (N, %)	
Squamous cell carcinoma	11 (0.52)
Adenocarcinoma	3 (0.14)
Adequacy of Pap smears (N, %)	2098 (100)
Unsatisfactory	65 (3.10)
Satisfactory	
with TZ	1068 (50.91)
without TZ	965 (46.00)

*mean + standard deviation, Pap: Papanicolaou, NILM: negative for intraepithelial lesion or malignancy, ASC-US: atypical squamous cells of undetermined significance, ASC-H: atypical squamous cells cannot exclude HSIL, LSIL: low grade squamous intraepithelial lesion, HSIL: high grade squamous intraepithelial lesion, TZ: transformation zone component, BW: bodyweight

Table 2. Comparison of Cytological Results Between Absent and Present TZ Smears

Results of smears	p value	Satisfactory With TZ	Satisfactory Without TZ	With TZ smears Age ≥ 30 yr	With TZ smears Age < 30 yr	Without TZ smears Age ≥30 yr	Without TZ smears Age < 30 yr
NILM	<0.001	990 (92.10)	951(98.55)	783 (92.66)	207 (92.83)	795 (97.9)	156 (100)
Abnormal Pap smears	<0.001	78 (7.30)	14 (1.45)	62 (7.34)	16 (7.17)	14 (2.09)	0
Atypical							
ASC-US	<0.001	57 (5.34)	5 (0.52)	45 (5.33)	12 (5.38)	5 (0.60)	0
ASC-H	0.734	3 (0.28)	2 (0.21)	3 (0.34)	0	2 (0.20)	0
AGC	0.178	2 (0.19)	0	2 (0.24)	0	0	0
LSIL	0.221	5 (0.47)	1 (0.10)	3 (0.34)	2 (0.90)	1 (0.10)	0
HSIL	0.607	1 (0.09)	2 (0.21)	1 (0.12)	0	2 (0.20)	0
Cancer (N, %)							
SCC	0.231	8 (0.75)	3 (0.31)	7 (0.83)	1 (0.45)	3 (0.40)	0
Adenocarcinoma	0.621	2 (0.19)	1 (0.10)	1 (0.12)	1 (0.45)	1 (0.10)	0
Total		1068	965	845	223	812 (100)	153 (100)

NILM: negative for intraepithelial lesion or malignancy, ASC-US: atypical squamous cells of undetermined significance, ASC-H: atypical squamous cells cannot exclude HSIL, LSIL: low grade squamous intraepithelial lesion, HSIL: high-grade squamous intraepithelial lesion, AGC: atypical glandular cells, SCC: squamous cell carcinoma, TZ: transformation zone component

Table 3. Relationship between Clinical Factors and Satisfactory without TZ Smears by Multivariate Binary Logistic Regression Analysis

Factors that influence upon TZ adequacy	P value	Odd ratio	
		(95 % confidence interval)	
Decreased TZ adequacy			
STD	0.002	1.65	(1.20-2.27)
Previous cryotherapy	0.030	3.76	(1.15-12.45)
Smear collected by OB-GYN staff	0.001	1.48	(1.17-1.89)
Hormonal usage	0.009	1.68	(1.14-2.47)
Increased TZ adequacy			
Age ≥50 yr	0.022	0.73	(0.56-0.96)
Nulliparity	0.001	0.70	(0.56-0.86)
Within 3-months postpartum	0.048	0.78	(0.58-1.05)
History of TZ inadequacy	<0.001	0.13	(0.10-0.17)
History of abnormal smears	<0.001	0.20	(0.09-0.42)
No change			
BW ≥ 70 kg	0.058	1.28	(0.99-1.65)
Menopause	0.495	1.10	(0.83-1.47)
Leukorrhea	0.139	0.32	(0.07-1.44)
Abnormal vaginal bleeding	0.354	0.32	(0.02-3.52)
Pregnancy	0.645	0.78	(0.27-2.25)
HIV infection	0.456	0.77	(0.39-1.53)
Myoma uteri/adenomyosis (enlarged uterus)	0.216	1.38	(0.83-2.31)
Previous LEEP	0.185	1.79	(0.75-4.25)
Smear collected by OB-GYN resident	0.635	0.94	(0.74-1.20)

BW: bodyweight, STD: sexually transmitted disease, TZ: transformation zone component, HIV: human immunodeficiency virus, LEEP: loop electrosurgical excision procedure, OB-GYN: obstetrics and gynaecology.

Table 4. Comparison of Factors Influencing TZ Collection between OB-GYN Staff and Trainees

		Staff	Trainee	p value
Decreased TZ group				
STD	Yes	67	165	0.194
	No	618	1248	
Previous cryotherapy	Yes	28	14	<0.001
	No	657	1399	
Hormonal usage	Yes	41	96	0.511
	No	644	1317	
Increased TZ group				
Nulliparity	Yes	505	801	<0.001
	No	180	612	
Age ≥ 50 yr	Yes	226	449	0.576
	No	459	964	
Within 3-months postpartum	Yes	91	272	0.001
	No	594	1141	
History of abnormal smears	Yes	59	90	0.061
	No	626	1323	
History of TZ inadequacy	Yes	175	390	0.282
	No	511	1022	

STD: sexually transmitted disease, TZ: transformation zone component, LEEP: loop electrosurgical excision procedure, OB-GYN: obstetrics and gynaecology.

hormonal usage, previous cryotherapy and smears collected by staffs were associated with inadequacy of TZ. The underlying disease such as enlarged uterus and HIV infection were not associated with TZ adequacy.

Cervical collection in nulliparity and within 3-months postpartum subjects were performed by resident trainee more often than faculty staffs. The subjects who had history of cryotherapy were usually assigned to get their screening performed by faculty staff members rather than resident trainee. Table 4 showed the factors that explained why the inadequacy of TZ smears was more commonly found by staff.

Discussion

Based on theory that abnormal finding should be found at endocervical cylindrical cells or TZ. The present study showed the significantly increased prevalence of abnormal cytology in adequacy of TZ group. The result was similar to that of the previous study that the prevalence of positive cytological results and histological diagnosed lesions were greater among cases with TZ adequacy group (Zhao et al., 2014).

No abnormal Pap smears were found in any subjects who were younger than 30 years old. This finding was in the same direction to The American Society for Colposcopy and Cervical Patholgy (ASCCP) guideline. It was recommended that the women who were younger than 30 years old with TZ inadequacy could be safely monitored through a follow up program (Massad et al., 2013). Women who were older than 30 years old should be immediately further investigated.

Many studies reported that unsatisfactory results were associated with many factors such as old age, vaginal estrogen cream (Richards et al., 2014), vaginal bleeding, endocervical polyps, and performing within 3-month postpartum (Lu et al., 2010) but no research reported satisfaction with absence of TZ.

Nulliparity was associated with increasing TZ adequacy. Cervix in nulliparity presented small, well form transformation zone and thus it was easy to thoroughly collect the cell from TZ.

Subjects with history of TZ inadequacy and abnormal smears were associated with increasing in TZ adequacy collection. This may be due to the more care of the physicians spending time scraping and carefully collecting the cells from cervix in these patients.

STD was associated with TZ inadequacy collection. This may be due to too much abnormal vaginal discharge which obscured transformation zone.

Postpartum women have many changes in their physiologic hormone. The cervical ectropion during pregnancy usually has not regressed (Cunningham et al., 2014). It was associated with increased unsatisfactory cervical smears (Lu et al., 2010). In this study, postpartum women showed improvement in collection of TZ. This finding may be from the change of cervix which had adequate mucous production with well visualized transformation zone.

Menopausal subjects in this investigation reported association with decreased adequacy of TZ smears with no statistical significance which comparable to the study of Lu (Lu et al., 2010).

Underlying disease such as HIV infection has high risk to get sexually transmitted human papillomavirus infection. A recent systemic review confirmed that women with HIV infection had high incidence of abnormal Pap smears (Denslow et al., 2014). However HIV infection and enlarged uterus were not associated with prevalence of specimen adequacy in this study.

Our finding reported that hormonal usage was associated with increased prevalence of TZ inadequacy. Prolong usage of progestin contraceptive will decrease the size of transformation zone which may effected in more difficulty for cell collection in this area (Barakat et al., 2013).

Women with abnormal vaginal bleeding were not found to be associated with the number of TZ adequacy. However the number of abnormal vaginal bleeding was only 4 of 2,098 women. Patients who had abnormal vaginal bleeding usually underwent different types of smears or were sent for fractional and curettage instead.

Previous LEEP was found to decrease the prevalence of TZ adequacy with no statistical significance. However, previous cryotherapy had decreased prevalence of TZ adequacy with statistical significance. Both LEEP and cryotherapy are known to cause structural change of cervix after healing and may have cervical stenosis in some women which did not allow spatula to pass through.

Smears collected by OB-GYN staff were associated with increased TZ inadequacy smears. OB-GYN staff performed more cases of high chance for decreased TZ component like previous cryotherapy and the number of cases done by staffs which had high chance to collect TZ component such as in nulliparious and postpartum women were less than residents or trainee.

Collection of cervical specimens should be carefully performed by experienced practitioner or change to liquid base cytology technique (Nandini et al., 2013) in the patients who have high chance of TZ inadequacy such as STD history, hormonal usage and previous cryotherapy.

Acknowledgements

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