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Cervical Cancer Prevention by Liquid-Based Cytology in a Low-Resource Setting

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1. Introduction

Cervical cancer is a major public health problem. Worldwide, it is the second most common cancer in women after breast cancer. Approximately 500,000 new cases of invasive cervical cancer have been diagnosed each year with more than 250,000 women dying of the disease. It is the most or second most common cancer among women in developing countries. In Thailand, a low-resource country, cervical cancer is the second most frequent cause of cancer in women, with nearly 10,000 new cases diagnosed and more than 5,200 dying from this disease each year (Ferlay et al., 2010). The incidence and mortality have declined during the last 50 years in developed countries because of increased availability of cervical cancer screening programs (Nieminen et al., 1995). However, cervical cancer continues to be a leading cause of cancer deaths in populations with a low socioeconomic level.

2. Cervical cancer screening

Screening is the use of methods to distinguish apparently unaffected people from those who may have a disease, or may develop it. It is a preliminary process to offer a diagnostic test and if required, treatment. Screening test is usually applied on a large scale and are generally offered to a population of people who have not sought medical attention on account of symptoms of the disease. The purpose of screening is to benefit the individuals being screened. Screening procedures are generally easier to perform and cheaper than diagnostic procedures. Although the screening test is harmless, it can cause anxiety and the subsequent investigations and treatment can be hazardous. Screening methods should provide the most attractive combination of negative predictive value (e.g., reassurance) and false positives that is attainable in a given setting and providers should ensure that a useful remedy is available for all individuals identified as being true positive.

The most widely used screening method for cervical cancer is conventional cytology (conventional Pap smear). Nowadays, conventional cytology is still considered a standard screening method worldwide, especially in developing countries. Conventional cytology has been being used as a standard screening method for cervical cancer in Siriraj Hospital since 1952. This cytological testing involves three major steps, i.e. collecting exfoliated cells from

the cervix, spreading the cells onto a slide, and microscopically examining these cells after staining. Collection of specimens for conventional cytology is performed according to the standard vaginal-cervical-endocervical (VCE) smear technique. A wooden (Ayre's) spatula and a cotton swab are used to collect cells from the posterior fornix, portio vaginalis and endocervix. The collected cells are then spread onto a glass slide and immediately immersed in 95% ethanol for fixation. Despite the proven effectiveness of cervical cytology screening in reducing the incidence of cervical cancer, over the last decade the accuracy of cervical cytology has been questioned. Two large meta-analyses have indicated that although the specificity of conventional cervical cytology is high, its sensitivity is much lower than previously estimated (Fahey et al., 1995; Nanda et al., 2000). Errors due to poor sampling and partial transferring of the collected sample onto a glass slide, consequently producing a nonrepresentative specimen, may account for up to 62% of false negative results (Gay et al., 1985).

3. Siriraj liquid-based cytology

Liquid-based cytology (LBC) was introduced in the mid-1990s as a way to improve performance of the test. LBC can improve specimen quality by providing a standardized method of collecting cervicovaginal material, and dispersing cells in a thin layer with relatively free of inflammation (Austin et al.,1998; Mount et al.,2004; Vassilakos et al.,2002). This results in the decrease in incidence of unsatisfactory smears and increase in detection rate of cytologic abnormality (Bolick et al.,1998; Corkill et al.,1998; Diaz-Rosario et al.,1999; Dupree et al.,1998; Fremont-Smith et al.,2004; Papillo et al.,1998; Roberts et al.,1997). Besides, the leftover specimen for LBC can also be used for HPV DNA testing which is currently incorporated into the management guidelines and post-therapy surveillance of the patient in some institutes. Recently, a number of different LBC techniques are available worldwide. ThinPrep® and SurePath™ are prototypes of LBC technology. They have been approved by the Food and Drug Administration for cervical cancer screening in the USA and are the most commonly used LBC techniques (Bishop et al., 1998; Lee et al., 1997). LBC was introduced as a cervical cancer screening technique in Thailand in 1997. Nowadays there are at least three commercially available LBC in Thailand, e.g. ThinPrep®, SurePath™, and Liqui-Prep®. Despite of their reputation, these LBC are still not in general use because of their cost. In the year 2005, we have developed an alcohol-based preservative solution, Siriraj liquid-based solution, and applied a modified Saccomanno's technique for cells preparation in our institute, and named this technology as the "Siriraj liquid-based cytology" or "Siriraj-LBC". The collection of specimens for Siriraj-LBC is similar to that for conventional cytology, except that a special plastic spatula is used instead of the Ayre's spatula and cotton swab. The plastic spatula has an extended endocervical tip at one end and a rounded rectangular tip at the other end, and scores at 4 cm from each end to facilitate spatula breaking (Figure 1). Immediately after cell collection, both ends of the spatula are manually broken at the scores, and put into a 30 mL plastic bottle containing 10 mL of Siriraj liquid-based solution.

3.1 Processing of Siriraj-LBC

The cell specimens are collected in bottles containing Siriraj liquid-based solution and keep at room temperature until processing. Siriraj-LBC slides are prepared according to the following steps (Bales, 2006) : agitate the bottle of specimen on a vortex mixer for 10 sec, and

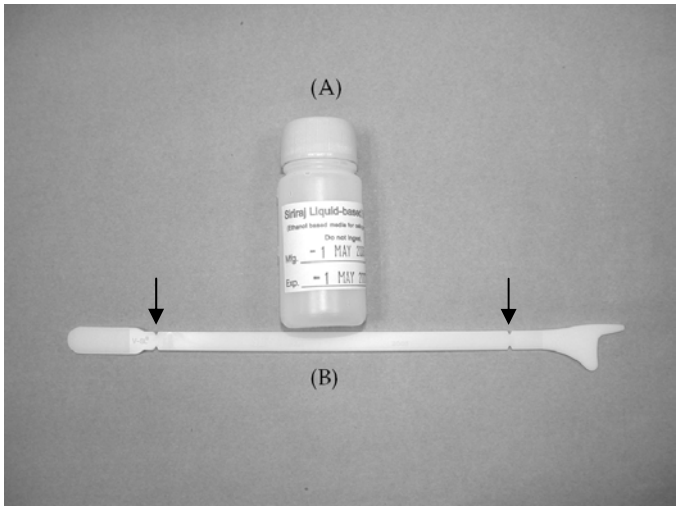

Fig. 1. Collecting devices for Siriraj-LBC: a 30-mL plastic bottle containing 10 mL of Siriraj liquid-based solution (A), and an extended-tip plastic spatula, arrow heads showing scores on the spatula (B)

pour the suspension into a 15-mL centrifuge test tube; centrifuge the specimen at 1000 g. for 10 min, and discard the supernatant; add Siriraj liquid-based solution (approximately 3 times of the sediment volume); agitate the test tube for 10 sec, aspirate 15-20 µL of the sample by using an auto-pipette, drop the sample onto a clean glass slide, smear the droplet to 2 cm in diameter, and let air dry at room temperature for 30 min; fix the slide in 95% ethanol for 20 minutes, and finally stain it with the routine Papanicolaou's staining technique (Figure 2).

3.2 Performance of Siriraj-LBC

Before a new screening or diagnostic tool is introduced into clinical use, it is necessary to evaluate its diagnostic performance. The best way for this evaluation is to compare the result from the new tool with that from the gold standard, revealing the performance parameters including sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), false positive (FP), false negative (FN), and accuracy of the test. However, such parameters cannot be obtained in studies evaluating performance of cervical cytology methods because not all of the study population undergoes the gold standard testing, i.e. colposcopy or cervical histology. The gold standard is mostly lacking in the group of patients who have normal cervical cytology. Therefore, numerous studies have evaluated the comparative performance of the LBC methods and conventional cytology with respect to test positivity, i.e. the detection rate of squamous intraepithelial lesion (SIL). Most studies have utilized one to two types of study design, i.e. "split-sample" or "direct-to-vial" study. With "split-sample" study, it is difficult to ensure that the two cytology specimens are comparable, since the specimen for conventional cytology slide is collected before the specimen for LBC. Therefore, this design seems to lead inherently to bias against LBC. With "direct-to-vial" study, the result of LBC is compared with that of conventional smear from historical data of an identical population; however, it is not certain that the two

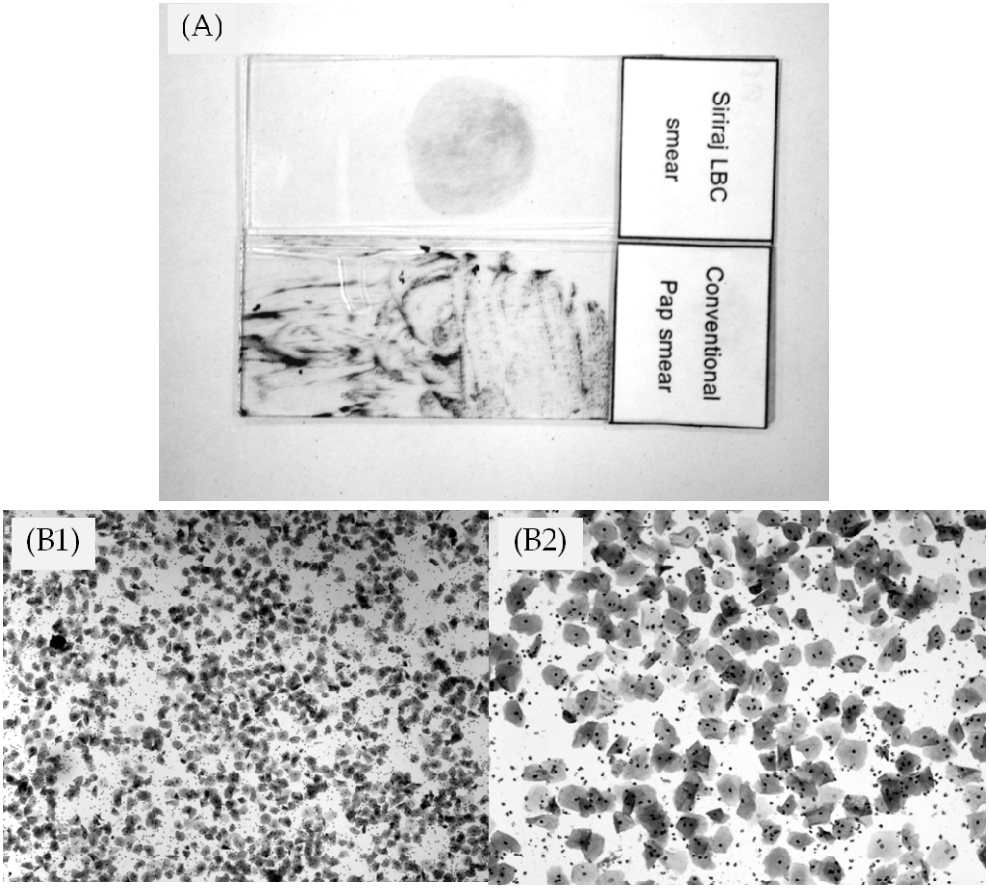

Fig. 2. Stained slides of Siriraj-LBC and conventional smears (A); micrographs of cervical cytology at low magnification: conventional smear (B1), Siriraj-LBC smear (B2)

populations are identical. The diagnostic performance of our home-made LBC has been evaluated in both the "split-sample" study and "direct-to-vial" study.

3.3 A split-sample study (Laiwejpithaya et al., 2008)

3.3.1 Study population and specimen processing

A cross-sectional study was carried out in the Gynecologic Cytology Unit, Department of Obstetric and Gynecology, Faculty of Medicine Siriraj Hospital, Mahidol University from January to February 2005. Study population were randomly selected from women attending for pelvic examination and cervical cancer screening at the Gynecologic Outpatient Department, Siriraj Hospital during the study period, excluding the women who had previously undergone any surgical procedures of the cervix, were pregnant or suspected of being pregnant, used any kinds of vaginal preparations within previous 24 hours, or denied to participate in the study. Specimens were collected for "split-sample" study by residents

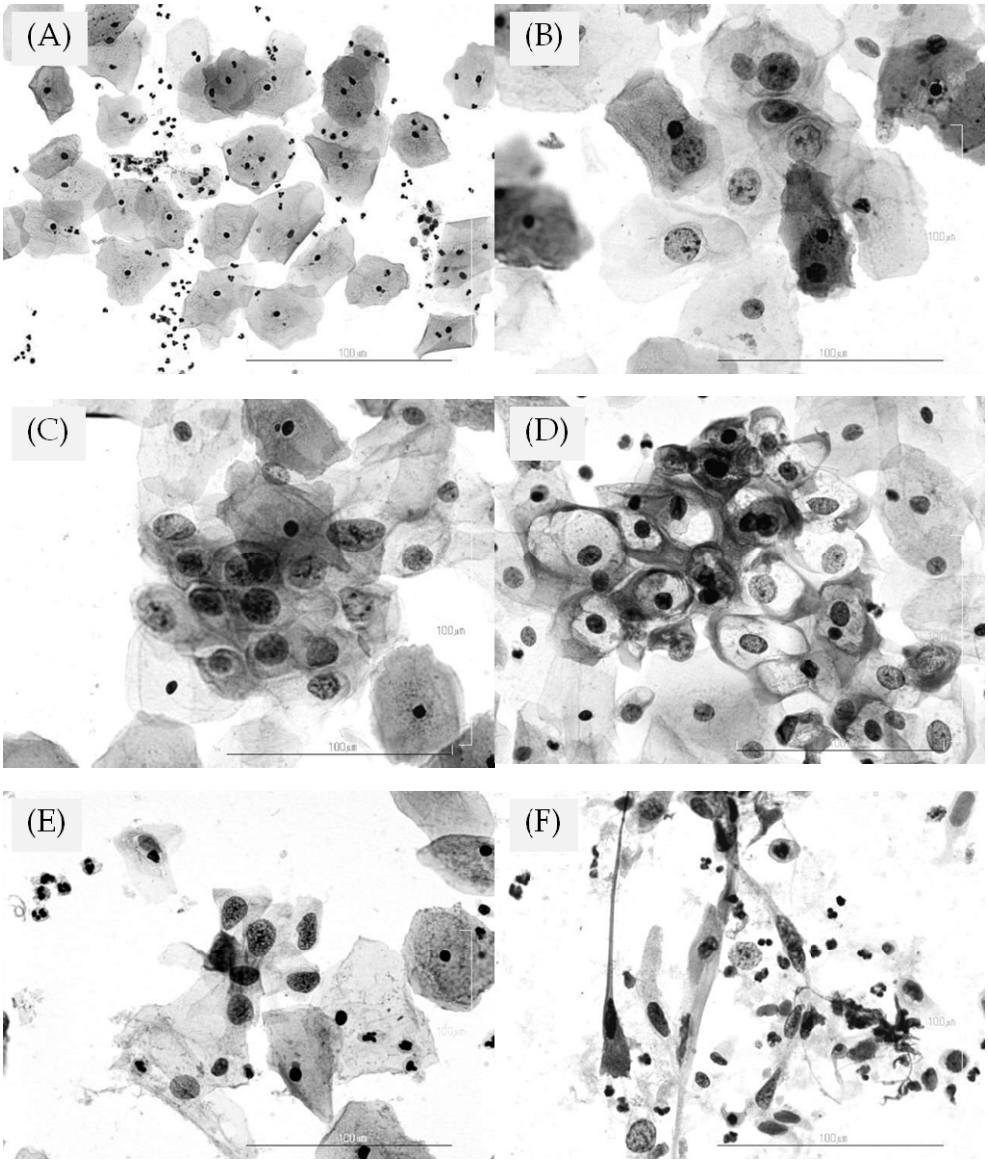

Fig. 3. Micrographs of cervical cytology using Siriraj-LBC at high magnification: normal (A), atypical squamous cells of undetermined significance (B), atypical squamous cells cannot exclude HSIL (C), low-grade squamous intraepithelial lesion with koilocytosis (D), high-grade squamous intraepithelial lesion (E), and squamous cell carcinoma (F)

or gynecologists who were staff members of the Department of Obstetrics and Gynecology. The collected cells were initially prepared for conventional cytology. Leftover cells in the collecting instruments were then collected for Siriraj-LBC by putting the instruments into a

30 mL plastic bottle containing 10 mL of Siriraj liquid-based solution. Specimens for both techniques were transported to the Gynecologic Cytology Unit, and processed by experienced technicians. All of the slides were screened by a team of cytotechnologists. The abnormal slides were reviewed and diagnosis made by an experienced cytopathologist. Examples of various abnormal cervical cells are shown in Figure 3. Evaluation of slides for conventional cytology and Siriraj-LBC was made in a blind fashion, i.e. the interpretations of the results from one technique were made without knowledge of those from the other technique. The cytological interpretation was made according to the Bethesda system 2001 as followed: negative for intraepithelial lesion or malignancy, reactive or reparative change, atypical squamous cells of undetermined significance (ASCUS), low-grade squamous intraepithelial lesion (LSIL), high grade squamous intraepithelial lesion (HSIL), atypical squamous cells cannot exclude HSIL (ASC-H), squamous cell carcinoma (SCC), atypical glandular cells (AGC), or adenocarcinoma (Solomon et al., 2002). Clinical management of abnormal cytology includes referral to colposcopy and treatment according to the guideline of Siriraj Hospital. The patients with cytology results of ASCUS or LSIL were suggested to have either colposcopy or repeat cervical cytology testing, and those with ASC-H, HSIL or more aggressive ones were referred to colposcopy.

3.3.2 Performance of cytology as a screening test

The performance of cytology was evaluated from detection rate of abnormal cervicovaginal cytology, and predictive values using colposcopy and/or histology as the gold standard. The data for calculating positive predictive value (PPV) were obtained from the patients who had abnormal results of Siriraj-LBC, and underwent operative procedures revealing histology. Those procedures included colposcopic directed cervical biopsy, loop electrosurgical excision procedure (LEEP), cold-knife conization, and hysterectomy. The data were presented in n (%), or odds ratio (OR) and 95% confidence interval (CI), as appropriate. Percentage of agreement was used to determine the diagnostic agreement between pairs of specimens evaluated by conventional cytology and Siriraj-LBC. Kappa and Spearman rho correlation coefficient were used to determine correlation of result between pairs of specimens. Chi-square test was used to compare frequency between the two cytology techniques. All tests were 2-sided, and a P value of less than 0.05 was considered statistically significant.

3.3.3 Results

There were 479 participants recruited during the study period. Their mean age was 41.6$\pm$12.6 years. The Siriraj-LBC significantly increased overall detection rate of abnormal cytology compared to the conventional method, i.e. from 1.67% to 11.1%, $P < 0.001$ (Table 1). The data yielded a complete diagnostic agreement of 430 of 479 pairs of specimens (89.8%). Among these, HSIL was detected in both specimens in 2 cases and cancer in 1 case. There were 49 cases whose Siriraj-LBC revealed higher cytologic grading than the conventional cytology did; whereas, none of the conventional cytology showed the vise versa result. The highest disagreement was found in 18 cases which were interpreted as normal by conventional cytology, but as ASCUS by Siriraj-LBC. As a result, these two cytology techniques had minimal to fair correlation with a Kappa of 0.128 ($P < 0.001$) and a Spearman rho correlation coefficient of 0.394 ($P < 0.001$) (Table 2).

Table 3 reveals final diagnoses by colposcopy or histology in 45 patients with abnormal cytological diagnoses by Siriraj-LBC. In this specific group, the Siriraj-LBC had a positive

Finding	Conventional	Siriraj-LBC	P-value
Overall	8 (1.67)	53 (11.1)	<0.001
ASCUS	2 (0.42)	18 (3.76)	
ASC-H	1 (0.21)	7 (1.46)	
LSIL	2 (0.42)	15 (3.13)	
HSIL	2 (0.42)	10 (2.09)	
Cancer	1 (0.21)	3 (0.63)	

[Data are n (%). The data were analyzed using Fisher's exact test.]
From Laiwejpithaya, S.; Rattanachaiyanont, M.; Benjapibal, M.; Khuakoonratt, N.; Boriboonhirunsarn, D.; Laiwejpithaya, S.; Sangkarat, S.; & Wongtiraporn, W. (2008). Comparison between Siriraj liquid-based and conventional cytology for detection of abnormal cervicovaginal smears: A split-sample study. Asian Pacific J Cancer Prev, Vol.9, No.4, pp. 575-580.

Table 1. Detection Rates of Abnormal Cervical Cytology by Conventional Cytology and Siriraj-LBC

Siriraj-LBC	Conventional cytology					
	Negative	ASCUS	ASC-H	LSIL	HSIL	Cancer
Negative	**426**	0	0	0	0	0
ASCUS	18	**0**	0	0	0	0
ASC-H	7	0	**0**	0	0	0
LSIL	12	2	0	**1**	0	0
HSIL	6	0	1	1	**2**	0
Cancer	2	0	0	0	0	**1**

(Data are number of cases. Identical diagnoses are shown in bold; cases with negative diagnosis by conventional cytology but abnormal diagnoses by Siriraj-LBC are in red; Spearman rho correlation = 0.394, Kappa = 0.128, P <0.001.)
From Laiwejpithaya, S.; Rattanachaiyanont, M.; Benjapibal, M.; Khuakoonratt, N.; Boriboonhirunsarn, D.; Laiwejpithaya, S.; Sangkarat, S.; & Wongtiraporn, W. (2008). Comparison between Siriraj liquid-based and conventional cytology for detection of abnormal cervicovaginal smears: A split-sample study. Asian Pacific J Cancer Prev, Vol.9, No.4, pp. 575-580.

Table 2. Comparison of Cytological Diagnoses between Conventional Cytology and Siriraj-LBC in 479 Pairs of Samples

predictive value (PPV) of 71.1% whereas conventional cytology had PPV and negative predictive value (NPV) of 85.7% and 31.6%, respectively. None of the ASCUS had cervical lesions beyond CIN1. Seven cases of ASC-H was found in Siriraj-LBC but only one in conventional cytology; one in seven (14.28%) of ASC-H had cervical lesion of CIN2/3. The Siriraj-LBC did not miss any high grade cervical lesions (CIN2/3 or cancer) but over-diagnosed cancer in one case; whereas conventional cytology failed to detect 3/6 (50%) cases of CIN2/3 and 1/2 (50%) cases of cancer.

3.3.4 Discussion

The present study used the data of the year 2005 when the Siriraj-LBC was undergoing the development process. We found that the detection rate of abnormal cells in Siriraj-LBC (11.06%) was much higher than that in the conventional cytology (1.67%). Even though the "split-sample" study has potential bias against LBC, we were not encountered with this problem. It was possible that the specimen processing in the Siriraj-LBC, especially the

Cytology	Diagnoses by Colposcopy or Histology			
	Normal	CIN1/HPV	CIN2/3	Cancer
Conventional				
Negative	12	22	3	1[b]
ASCUS	1	1	0	0
ASC-H	0	0	1	0
LSIL	0	0	1	0
HSIL	0	1	1	0
Cancer	0	0	0	1[c]
Siriraj-LBC				
Negative	NA	NA	NA	NA
ASCUS	7	9	0	0
ASC-H	3	3	1	0
LSIL	1	8	0	0
HSIL	1	4	5	0
Cancer	1[a]	0	0	2[b,c]

(Cytological vs. histological diagnoses: [a]adenocarcinoma of endometrium vs normal, [b]adenocarcinoma of unknown origin vs. adenocarcinoma of peritoneum, and [c]adenocarcinoma of endometrium vs. squamous cell carcinoma. CIN = cervical intraepithelial neoplasia, HPV = human papilloma virus infection, NA = not applicable)
From Laiwejpithaya, S.; Rattanachaiyanont, M.; Benjapibal, M.; Khuakoonratt, N.; Boriboonhirunsarn, D.; Laiwejpithaya, S.; Sangkarat, S.; & Wongtiraporn, W. (2008). Comparison between Siriraj liquid-based and conventional cytology for detection of abnormal cervicovaginal smears: A split-sample study. Asian Pacific J Cancer Prev, Vol.9, No.4, pp. 575-580.

Table 3. Final Diagnoses by Colposcopy or Histology in 45 patients with Abnormal Cytological Diagnoses

agitation of collecting devices in the liquid-based solution would elude the entrapped cells into the solution, therefore more cells were collected into the solution, and then evenly sampling to put onto a glass slide. We found that the complete diagnostic agreement between our LBC and the standard cytology was in high level (89.77%). However, the Kappa and the correlation coefficient were not in that high level; this was due to the interesting fact that detection rate of abnormal cells was much higher in the Siriraj-LBC than in the conventional cytology. Our result was comparable with that of Park et al (2001) showing that the results of conventional cytology and LBC exactly agreed in 91.4% of cases.

The increase in detection rate raised the concern of increase in false positive cytology. In the present study, 45 cases of the abnormal cytology detected by Siriraj- LBC undergone gold standard testing. We found that the overall PPV in the Siriraj-LBC was less than that in the conventional cytology (71.1% vs. 85.7%). This implied that the Siriraj-LBC increase false positive result from 7.7% to 100.0%. In this specific group of patients with positive result by Siriraj-LBC, the conventional cytology had an astounding high false negative result of 81.2%. The false negative result of Siriraj-LBC was unknown because none of the patients with negative cytology undergone gold standard testing, despites, we assumed that the Siriraj-LBC would have less false negative result since none of the negative Siriraj-LBC had abnormal conventional cytology. However, we are aware that these numbers are not the real

false negative and false positive rates, as the actual rates cannot be obtained due to the limitation of this kind of study.

The high false positive result by Siriraj-LBC caused only little concern. Considering that only HSIL or cancer needs further invasive investigation, e.g. conization and/or diagnostic curettage, two in 45 cases had risk of unnecessary further investigation if the Siriraj-LBC was used in place of conventional cytology; this risk returned with the benefit of detecting three more cases of CIN2/3 and one case of cancer which were cases with false negative result in the conventional cytology. Noteworthy, the missing cancer in conventional cytology was a case of peritoneal adenocarcinoma without lesion at the uterine cervix.

Our limited data showed that the conventional cytology had high false negative result where as the Siriraj-LBC had high false positive result. It is estimated that approximately two thirds of false negative result in the conventional cytology are caused by sampling error due to limited transfer of cells from the collecting device onto the slide (Gay et al., 1985). The false positive result in the Siriraj-LBC was due to the increase in all types of abnormal epithelial cells. This may be due to the misinterpretation of immature squamous metaplastic or atrophic cells to be abnormal cells because the morphologies of these cells are alike. Moreover these cells could be more easily detected in the Siriraj-LBC than in the conventional cytology because of the better quality of slide. However, we could not disregard the fact that our novice in the LBC field also contributed to this false positive. We expect to get better results of this technique in the future.

Our result was compatible with many previous reports. Nanda et al (2000) reviewed the accuracy of conventional and new methods of Papanicolaou (Pap) testing to detect cervical cancer and its precursors. Ninety-four studies of the conventional Pap test showed that, estimates of sensitivity and specificity varied greatly in individual studies. In the 12 studies with the least biased, estimates sensitivity ranged from 30-80% and specificity ranged from 86-100%. Guo et al (2005) evaluated the accuracy of a LBC test, ThinPrep®, by comparing concurrent LBC and cervical biopsy results of 782 patients who were referred for colposcopy because of previously abnormal conventional cytology. They found that concurrent LBC has high diagnostic accuracy for SIL. Besides, several studies showed that the detection rates of LSIL and HSIL are improved by LBC but the effect of LBC on the detection of ASC-US is uncertain (Limaye et al., 2003; Mount et al., 2004).

The results from Siriraj-LBC and conventional cytology have high diagnostic agreement and minimal to fair correlation. The Siriraj-LBC increases detection rate of abnormal cervicovaginal cells with probably decrease false negative but increase false positive from the baseline values by conventional cytology. Therefore the screening performance of Siriraj-LBC is not inferior to the conventional cytology and may be used as an alternative screening method for cervical cancer.

3.4 A direct-to-vial study (Laiwejpithaya et al., 2009)

3.4.1 Study population and specimen processing

The study was carried out in the Gynecologic Cytology Unit, Department of Obstetrics and Gynecology, Faculty of Medicine Siriraj Hospital, Mahidol University. Data were retrieved from the database of the Gynecologic Cytology Unit. The data of conventional cytology and

LBC were recruited from records of the years 2004 and 2006, respectively. The specimens for either conventional cytology or LBC were obtained from patients who had pelvic examination at the Gynecologic Outpatient Department of Siriraj Hospital during the relevant study periods. Specimens were collected by residents and staff members of the Department of Obstetrics and Gynecology. Collection of specimens for conventional cytology was performed according to the standard VCE smear technique. The collection of specimens for Siriraj-LBC was similar to that for conventional cytology, except that a special plastic spatula was used instead of the Ayre's spatula and cotton swab. All specimens were transported to the Gynecologic Cytology Unit, and processed by experienced technicians. All of the slides were screened by a team of cytotechnologists and the abnormal slides were reviewed and diagnosis made by an experienced cytopathologist. Clinical management of abnormal cytology includes referral to colposcopy and treatment according to the guideline of Siriraj Hospital as mentioned above.

3.4.2 Performance of cytology as a screening test

The performance of Siriraj-LBC was evaluated from the detection rate of abnormal cervical cytology, and predictive values using cervical histology as the gold standard. The data for calculating negative predictive value (NPV) were obtained from the patients who had normal results of pre-hysterectomy screening cervical cytology. The data for calculating positive predictive value (PPV) were obtained from the patients who had abnormal results of cervical cytology and underwent operative procedures revealing cervical histology. Those procedures included colposcopic directed cervical biopsy, loop electrosurgical excision procedure (LEEP), cold-knife conization, and hysterectomy. Normal cervical histology was considered when the cervical mucosa was clear from any neoplastic lesion, ignoring the histopathological result of endometrium. The data were presented as mean$\pm$standard deviation (SD), n (%), or % change, as appropriate. Data were analyzed using the t-test for continuous data or Chi-square test for categorical data. All tests were 2-sided, and a P-value of < 0.05 was considered statistically significant.

3.4.3 Results

There were 23,676 records of conventional Papanicolaou's smear and 25,510 records of Siriraj-LBC in the years 2004 and 2006, respectively. Almost all of the specimens came from Thai women. The mean age$\pm$SD of women in the years 2004 and 2006 were 40.67$\pm$12.54 and 42.66$\pm$12.21 years, respectively, which were not statistically different. Compared with the conventional smear, the Siriraj-LBC significantly increased overall detection rate of abnormal cytology by 110.23% (from 1.76% to 3.70%, $P < 0.001$), as it increased the detection rate of ASCUS, LSIL, HSIL, ASC-H, and malignant cells, but it did not significantly increase the detection rate of atypical glandular cells (AGC). The Siriraj-LBC significantly reduced the number of smears that were deemed poor quality by 73.44% (from 18.60% to 4.94%, $P < 0.001$), as it markedly decreased the smears with obscuring blood and inflammatory cells, thick smears (Fig. 1b and c), and the smears without transformation zone component. However, the Siriraj-LBC had a marked increase in scant cellular smears (Table 4).

The predictive value of Siriraj-LBC was better than that of conventional smear. The NPV of Siriraj-LBC was apparently higher than that of conventional cytology (96.33% vs. 92.74%, $P = 0.001$). The PPV of both methods was > 80%, which did not demonstrate any significant

	Siriraj-LBC (N= 25,510)	Conventional (N= 23,676)	% change	P
All abnormal cervical cells	944 (3.70)	417 (1.76)	+110.23	<0.001
ASCUS	251 (0.98)	100 (0.42)	+133.33	<0.001
LSIL	278 (1.09)	132 (0.56)	+94.64	<0.001
HSIL	213 (0.83)	94 (0.40)	+107.50	<0.001
ASC-H	117 (0.46)	41 (0.17)	+170.59	<0.001
AGC	25 (0.10)	16 (0.07)	+42.86	0.243
Malignant	60 (0.24)	34 (0.14)	+71.43	0.020
All poor quality slides	1261 (4.94)	4404 (18.60)	-73.44	<0.001
No transformation zone component	1094 (4.29)	3766 (15.91)	-73.04	<0.001
Scant cellular smears	153 (0.60)	51 (0.22)	+172.73	<0.001
Thick smears	2 (0.01)	212 (0.90)	-98.89	<0.001
Obscuring blood and inflammatory cells	12 (0.05)	375 (1.58)	-96.84	<0.001

[Note: Data are n (%). Percent change was the incremental (+) or decremental (_) rate of Siriraj-LBC comparing to the baseline rate of conventional cytology. The data were analyzed using Chi-square test.] From Laiwejpithaya, S.; Benjapibal, M.; Laiwejpithaya, S.; Wongtiraporn, W.; Sangkarat, S.; & Rattanachaiyanont, M. (2009). Performance and cost analysis of Siriraj liquid-based cytology: a direct-to-vial study. Eur J Obstet Gynecol Reprod Biol, Vol. 147, No.2, pp. 201-205.

Table 4. Detection rates of abnormal cervical cells and quality of slides using Siriraj liquid-based cytology (Siriraj-LBC, year 2006) and conventional Papanicolaou's smear (year 2004)

difference between the methods. The PPV for SCC was the highest; whereas that for abnormal glandular cell types was the lowest (Table 5). The cost of Siriraj-LBC was higher than that of the conventional cytology used in Siriraj Hospital but lower than that of the commercially available LBC in Thailand. When cost was estimated from the laboratory charge, it was found that the cost of Siriraj-LBC was 1.67, 0.50, and 0.30 times of those of the conventional cytology, Liqui-Prep®, and ThinPrep®, respectively (Table 6).

Cervical cytology	N	Cervical histology, n (%)		P
		Normal	Abnormal	
Normal				
Siriraj-LBC	1012	975 (96.33)[(a)]	37 (3.67)	0.001
Conventional	744	690 (92.74) [(a)]	54 (7.26)	
Overall abnormalitie				
Siriraj-LBC	277	47 (16.97)	230 (83.03)[(b)]	0.285
Conventional	167	22 (13.17)	145 (86.83)[(b)]	
ASCUS				
Siriraj-LBC	13	7 (53.85)	6 (46.15)[(b)]	0.342
Conventional	9	3 (33.33)	6 (66.67)[(b)]	

Cervical cytology	N	Cervical histology, n (%)		P
		Normal	Abnormal	
LSIL				
Siriraj-LBC	41	6 (14.63)	35 (85.37)[(b)]	0.597
Conventional	29	3 (10.34)	26 (89.66)[(b)]	
HSIL				
Siriraj-LBC	124	10 (8.06)	114 (91.94)[(b)]	0.301
Conventional	71	3 (4.23)	68 (95.77)[(b)]	
ASC-H				
Siriraj-LBC	33	4 (12.12)	29 (87.88)[(b)]	0.579
Conventional	23	4 (17.39)	19 (82.61)[(b)]	
SCC				
Siriraj-LBC	26	0 (0.00)	26 (100.00)[(b)]	0.183
Conventional	15	1 (6.67)	14 (93.33)[(b)]	
Abnormal glandular cell types (AGC+AIS+Adenocarcinoma)				
Siriraj-LBC	40	20 (50.00)	20 (50.00)(b)	0.464
Conventional	20	8 (40.00)	12 (60.00)(b)	

[Note: Data are n (% of the corresponding row). The data were analyzed using Chisquare test. Abnormal cervical histology meant cervical tissue specimen showing intraepithelial neoplasia, squamous cell carcinoma, or adenocarcinoma. (a) Negative predictive value; (b) positive predictive value; AGC, atypical glandular cells; AIS, adenocarcinoma in situ; ASCUS, atypical squamous cells of undetermined significance; ASC-H, atypical squamous cells cannot exclude HSIL; CI, confidence interval; LSIL, low grade squamous intraepithelial lesion; HSIL, high-grade squamous intraepithelial lesion; SCC, squamous cell carcino]
From Laiwejpithaya, S.; Benjapibal, M.; Laiwejpithaya, S.; Wongtiraporn, W.; Sangkarat, S.; & Rattanachaiyanont, M. (2009). Performance and cost analysis of Siriraj liquid-based cytology: a direct-to-vial study. Eur J Obstet Gynecol Reprod Biol, Vol. 147, No.2, pp. 201-205.

Table 5. Predictive values of cervical cytology using Siriraj liquid-based cytology (Siriraj-LBC, year 2006) and conventional Papanicolaou's smear (year 2004)

Costs	Thai baht	US dollars
Siriraj liquid-based cytology	150.00	4.55
Siriraj conventional cytology	90.00	2.72
Commercially available liquid-based cytology techniques		
Liqui-Prep®	300.00	9.09
ThinPrep®	500.00	15.15

From Laiwejpithaya, S.; Benjapibal, M.; Laiwejpithaya, S.; Wongtiraporn, W.; Sangkarat, S.; & Rattanachaiyanont, M. (2009). Performance and cost analysis of Siriraj liquid-based cytology: a direct-to-vial study. Eur J Obstet Gynecol Reprod Biol, Vol. 147, No.2, pp. 201-205.

Table 6. Costs of Siriraj liquid-based cytology, conventional cytology at Siriraj Hospital, and commercially available liquid-based cytology techniques in Thailand

3.4.4 Discussion

In the present direct-to-vial study, we compared the data of patients undergoing specimen collection for cervical cytology by the same physician group in two 12-month periods, targeting conventional Papanicolaou's smear in 2004 and Siriraj-LBC in 2006. As expected, the Siriraj-LBC increased detection of precancerous lesions of the uterine cervix. From our previous split-sample study conducted during the development of this technology in 2005, the Siriraj-LBC showed a tremendously increased detection rate of abnormal cytology of 565%, from 1.67% to 11.10%. However, the increase in the detection rate decreased to 100% in 2006 when the Siriraj-LBC was routinely used. The huge increase in 2005 was probably due to rigorous examination of slides during the development of the new technology. The increase in 2006 was less pronounced but still impressive even though the slides were routinely examined in the same manner as they were in 2004.

The increase in detection rate of Siriraj-LBC was probably due to the benefit of improved slide quality. Whereas 18.60% of our conventional smears had limited quality due to no endocervical or transformation zone component, obscuring blood and inflammatory cells, thick smear, or scant cellular smears, only 4.94% of the Siriraj-LBC had smears limited by these factors. Our results were in line with the experience of others (Bolick et al.,1998; Corkill et al.,1998; Diaz-Rosario et al.,1999; Dupree et al.,1998; Fremont-Smith et al.,2004; Papillo et al.,1998; Roberts et al.,1997). The improvement in the quality of slides in 2006 is likely to be due to the property of Siriraj- LBC itself rather than other factors, since the detection rate and quality of slides before 2005 had never reached those of the slides in 2006. Our 3-year retrospective data from 2002 to 2004 showed that our conventional smears had detection rates varying from 1.7% to 2.1%, and the poor quality smears varying from 18.6% to 32.6%.

The better detection rate of intraepithelial lesions is a common thread in studies on liquid-based cervical cytology. The increase in detection rates in previous studies varied from 12.0% to 106.8% (Bolick et al.,1998; Corkill et al.,1998; Diaz-Rosario et al.,1999; Dupree et al.,1998; Fremont-Smith et al.,2004; Papillo et al.,1998; Roberts et al.,1997). The reason for the wide range of this effect is not clear; but a wide range in detection rates of intraepithelial lesions by the conventional smear is also noted in the same studies, i.e. 1.1–2.7%. The increment seems higher in the studies with a low baseline detection rate using the conventional smear. In addition, this variation may represent regional or population-based differences, or different practice patterns. In our direct-to-vial study, Siriraj-LBC had a 100% increment for the detection of SIL from a baseline rate of 0.96% (LSIL = 0.56% and HSIL = 0.40%). The overwhelming diagnostic improvement of Siriraj-LBC, especially with respect to HSIL detection, would enhance the success of cervical cancer screening in our institute. Owing to the fact that the LBC slide makes it easier for a cytologist to find small numbers of abnormal cells, the percentages of ASCUS and ASC-H were also higher with Siriraj-LBC compared with the conventional method. The results were welcomed by the gynecologists who were chronically in fear of false negative results.

The increase in detection rate came with the concern of an increase in false positive cytology. From our cytology-histology paired data, Siriraj-LBC increased NPV without compromising PPV. This information implied that the Siriraj-LBC decreased false negative results, i.e. from 7.26% to 3.67% (P = 0.001) without affecting the overall false positive result, i.e. 13.17% vs.

16.97% (P = 0.285). However, we were aware that these numbers were not the real false negative and false positive rates, as the actual rates could not be obtained due to the limitations of this kind of study. The Siriraj-LBC displayed a higher false positive HSIL than the conventional method did, probably due to the morphological resemblance of immature squamous metaplastic or atrophic cells to HSIL. As these cells were easier to detect in the Siriraj-LBC than in the conventional smear, the higher false positive HSIL was not unexpected. However, as discussed in the split-sample study, we could not disregard the fact that our being novices in the LBC field could contribute to this false positivity. We expect to get better results using this technique in the future.

When the laboratory cost of the various cervical cytology techniques was calculated, we found that the cost of Siriraj-LBC was only 67% higher than that of the conventional cytology technique used in Siriraj Hospital and was lower than that of the commercially available LBC techniques in Thailand. However, we did not directly compare the diagnostic performance among the LBC techniques; therefore we did not know which LBC technology was the most cost-effective for our population. Nevertheless, because of its low cost and better performance than conventional cytology, Siriraj-LBC would be an accessible LBC technology for women of low-socioeconomic level. As a result, Siriraj-LBC has replaced the conventional cytology in Siriraj Hospital since 2006.

The Siriraj-LBC shows an impressive improvement in the detection rate of abnormal cervical cells. The Siriraj-LBC has an acceptable performance and quality which are within the same range as other previously reported LBC techniques. Therefore, the Siriraj-LBC may be considered a better option for cervical cancer screening than the conventional method and more economical than the commercially available LBC.

4. Conclusion

The diagnostic performance of our home-made LBC has been evaluated in both the "split-sample" study and "direct-to-vial" study and we have found that the screening performance of Siriraj-LBC was superior to that of conventional cytology. Our LBC does not require any expensive equipment; therefore its cost is much less than that of the commercial ones. Siriraj-LBC may be considered a better option for cervical cancer screening than the conventional method. For centers where conventional Pap smear does not perform well, the introduction of a low cost Siriraj-LBC may help to improve performance. We believe that the Siriraj-LBC will make a significant change in cervical cancer screening and patient management in our country.

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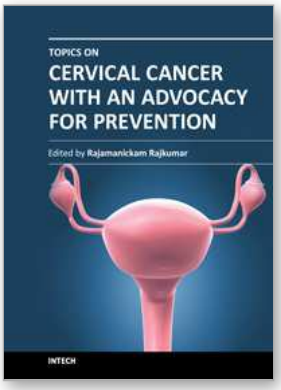

Topics on Cervical Cancer With an Advocacy for Prevention

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Cervical Cancer is one of the leading cancers among women, especially in developing countries. Prevention and control are the most important public health strategies. Empowerment of women, education, "earlier" screening by affordable technologies like visual inspection, and treatment of precancers by cryotherapy/ LEEP are the most promising interventions to reduce the burden of cervical cancer.Dr Rajamanickam Rajkumar had the privilege of establishing a rural population based cancer registry in South India in 1996, as well as planning and implementing a large scale screening program for cervical cancer in 2000. The program was able to show a reduction in the incidence rate of cervical cancer by 25%, and reduction in mortality rate by 35%. This was the greatest inspiration for him to work on cerrvical cancer prevention, and he edited this book to inspire others to initiate such programs in developing countries. InTech - Open Access Publisher plays a major role in this crusade against cancer, and the authors have contributed to it very well.

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