

## RESEARCH COMMUNICATION

## Risk Factors and Histological Outcome of Abnormal Cervix with Human Papilloma Infection in Northeastern Thai-women

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### Abstract

This study aimed to investigate the histological outcome of cervix with human papillomavirus (HPV) infection and the association of risk factors with cervical intraepithelial neoplasia (CIN) and invasive cervical carcinoma (ICC) development in Northeast Thai women. The study population (n=210) comprised 71 cases of normal cervix, 71 cases of CIN and 68 cases of ICC. The histological outcome of HPV infection was determined for 9.5% of the study population. Increased risk factors for CIN were observed for more than one partner (odds ratio (OR)=3.75, p<0.05), history of sexually transmitted disease (STD) (OR=2.28, p<0.05), menarche under 14 years of age (OR=0.31, p<0.05) and partners' smoking history (OR=3.98, p<0.01). Increased risk for ICC was observed for those with a history of STDs (OR=0.14, p<0.01) and multiparity (OR=2.53, p<0.01). Age at first sexual intercourse was not a risk factor in this study population. Further studies with HPV-DNA tests should more precisely quantify the risks.

**Key Words:** Cervical intraepithelial neoplasia - invasive cervical carcinoma - human papilloma infection - risk factors

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### Introduction

Cervical cancer is the second-most prevalent cancer and the third leading cause of female cancer deaths after breast and lung cancer (Ferlay et al., 2002); thus, an important public health problem worldwide. Cervical cancer remains a leading cause of cancer-related death for women in developing countries. Khon Kaen Northeast Thailand has a moderately high incidence (i.e., the age-standardized rate in 1995-1999 was 16.2/100,000) and cervical cancer is the second-most common cancer of women after liver cancer (Sriamporn et al., 2003).

Molecular and epidemiological studies have shown an association between cervical cancer and sexual activity and indicated that infection with human papilloma virus (HPV) is a precursor event in the genesis of cervical cancer (Schiffman et al., 2003; Tsuda et al., 2003). HPV is a major risk factor for the development of cervical intraepithelial neoplasia (CIN) and/or invasive cervical cancer (ICC) (Burd, 2003). As only some infected women develop cervical cancer, other factors must be important (Franco et al., 2001; Clifford et al., 2003). Several lifestyle factors have been suggested as risk factors for cervical cancer, including early age at first intercourse, multiple partners, coital frequency, high parity, smoking and sexual transmitted infection (Kjaer

et al., 1994; Chichreon et al., 1998). Our previous studies on cervical cancer in Northeast Thailand confirmed that HPV infection and certain risk factors (i.e., sexual behavior and smoking) contribute to the development of cervical cancer (Settheetham-Ishida et al., 2004a; Settheelham-Ishida et al., 2006)

In this study, we report on the histological outcomes of HPV infection and the relationships between abnormal cervix (CIN and ICC) and risk factors in Northeast Thailand.

### Materials and Methods

#### *Study subjects*

A total 210 women were enrolled in this study, 71 cases had confirmed histological diagnosis of a normal cervix, 71 cases of CIN and 68 of ICC. The study subjects were not selected by age. Normal cervix material was collected from cases of leiomyoma, CINs from LEEP conization and ICC from biopsy in Tumor Clinic at the Department of Obstetrics and Gynecology, Faculty of Medicine, Khon Kaen University, Thailand.

The purpose and procedures of this study were explained to all of the subjects. After informed consent was obtained, the participants were asked to complete a questionnaire about the risk factors, including information on: age, sexual

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**Table 1. Histological Outcome of Study Population**

Histological diagnosis	No. patients (n=210)	%
Normal cervix	64	30.5
Normal cervix with HPV	7	3.3
CIN I	5	2.4
CIN I with HPV	3	1.4
CIN II	10	4.8
CIN II with HPV	4	1.9
CIN III	43	20.5
CIN III with HPV	6	2.9
ICC	68	32.4

behavior, history of reproduction, history of sexually transmitted diseases (i.e., STDs: *Neisseria gonorrhoea*, HPV, Herpes simplex virus type 2, *Trichomonas vaginalis*, *Chlamydia* spp and *Candida* spp.) and history of smoking.

The Ethics Committee of Khon Kaen University approved this study.

*Statistical analyses*

The study population was grouped for analysis according to the type of cervix material (i.e., normal, CIN or ICC). The association between the histological diagnosis and each of the variables was measured using the Mantel-Haenszel X<sup>2</sup>-test. The odd ratios (ORs) and the 95% confidence intervals (CI) were calculated to evaluate the risk factors. A p-value <0.05 was required for statistical significance.

**Results**

The histological outcomes of the study population are presented in Table 1. Of the 210 women, 9.5% (twenty) had

an HPV infection and the histological diagnoses were: normal epithelium (3.3%), CIN I (1.4%), CIN II (1.9%) and CIN III (2.9%). Of the 9.5%, the 30.5, 2.4, 4.8, 20.5 and 32.4 percent had a normal cervix, CIN I, CIN II, CIN III and ICC, respectively.

Data for risk factors associated with the abnormal cervixes are presented with ORs in Table 2. The number of partners, history of STD, smoking status of partners, and age at menarche increased the risk of CIN. History of STD and number of parity increased the risk of ICC. Women who had more than one sexual partner demonstrated a statistically significant, 3.75-fold higher risk, of CIN (95% CI=1.21-11.53, p-value <0.05) than those who had one. History of STDs, smoking history of partner(s) and age at menarche under 14 years of age showed a statistically significant, 2.28-, 3.98- and 0.31-fold higher risk of CIN (respectively, 95% CI=1.06-4.91, p-value <0.05; 95% CI=1.72-9.16, p-value < 0.01 and 95% CI=0.11-0.88, p-value <0.05). History of STD and 3 or more number of parity elevated the risk 0.14- and 2.53-fold for ICC (95% CI=0.03-0.56, p-value <0.01 and 95% CI=1.20-5.33, p-value <0.01, respectively).

Age of women and age at first sexual intercourse were not associated with any significant increase in the risk of CIN and ICC development.

**Discussion**

In general, the major risk factors for cervical cancer appear to include initiation of sexual activity at an early age, multiple sexual partners, infection with HPV and cigarette smoking. More than 90% of all cervical cancers are squamous cell carcinomas, and evidence suggests that

**Table 2. Association between Risk Factors and Abnormal Cervix (CIN, ICC). OR1=odds ratio from a comparison of subjects with CIN and normal histology. OR2=odds ratio from a comparison of subjects with ICC and normal histology.**

Variable	CIN (n=71)	ICC (n=68)	Normal (n=71)	OR1 (95%CI)	OR2 (95%CI)
Age (years)					
≤35	19 (26.8)#	6 (8.8)	11 (15.5)	0.50 (0.20-1.24)	1.89 (0.68-5.26)
>35	52 (73.2)	62 (91.2)	60 (84.5)		
Number of partners					
1	58 (81.6)	58 (85.29)	67 (94.4)	3.75 (1.21-11.5)*	2.89 (0.90-9.16)
>1 <sup>ab</sup>	13 (18.3)	10 (14.71)	4 (5.6)		
Age at first sexual intercourse (years)					
≤17	65 (91.6)	59 (86.8)	64 (90.1)	0.84 (0.28-2.54)	1.39 (0.54-3.85)
<17 <sup>ab</sup>	6 (8.5)	9 (13.2)	7 (9.9)		
History of STD					
Never	47 (66.2)	66 (97.1)	58 (81.7)	2.28 (1.06-4.91) *	0.14 (0.03-0.56)**
Ever	24 (33.8)	2 (2.9)	13 (18.3)		
Smoking status of partner					
Never	45 (63.4)	55 (80.9)	62 (87.3)	3.98 (1.72-9.16) **	1.63 (0.65-4.02)
Ever <sup>ab</sup>	26 (36.7)	13(19.1)	9 (12.7)		
Number of parity					
≤3	44 (62.0)	23 (33.8)	40 (56.3)	0.79 (0.40-1.54)	2.53 (1.20-5.33)**
>3	27 (38.0)	45 (66.2)	31 (43.7)		
Menarche (years)					
≤14	66 (93.0)	61 (89.7)	57 (80.3)	0.31 (0.11-0.88)*	0.47 (0.18-1.21)
<14 <sup>b</sup>	5 (7.0)	7 (10.3)	14 (19.7)		

#percentage values \*p value <0.05, \*\*p value <0.01, <sup>a</sup>significant for cervical cancer development (Settheetham-Ishida et al., 2004a)

<sup>b</sup>significant for CIN development (Settheetham-Ishida et al., 2006)

cervical carcinoma is related to sexually transmitted organisms. Research indicates that HPV is a cofactor in the development of CIN and ICC (Ngelandgel et al., 1998; Burd 2003; Setheetham-Ishida et al., 2004b).

Most (90 to 95 percent) of squamous cell carcinomas of the cervix contain HPV DNA (Kjaer et al., 1994; Green et al., 2003; Setheetham-Ishida et al., 2004a). The virus infects the tissue of the lower genital tract and may produce obvious genital warts or mild, moderate or severe dysplasia and in situ carcinoma. Genital warts are associated with cervical dysplasia and ICC (in 25% of all cases). Although infection with HPV increases the risk of cervical cancer, many women infected by HPV do not develop cervical cancer. Co-factors--such as immune system functioning, nutrition and diet, hormones, smoking and other STDs have been studied (Luesley et al., 1994; Blomfield et al., 1998; Chichreon et al., 1998; Ho et al., 1998; Ngelandgel et al., 1998; Franco et al., 2001; Giuliano et al., 2002;)

Results of HPV-DNA testing of the specimens of cervical tissue from the subjects in this study were not available. We therefore only report on the histological diagnosis of the study population with HPV infection; that is, 9.5% of the studied population. Previous reports about the histological outcomes of HPV infection varied between 10.1 and 89.2 percent in the United Kingdom (Luesley et al., 1994; Blomfield et al., 1998) and 62.5% in Venezuela (Nunez et al., 2004). The prevalence of oncogenic HPV DNA in the normal cervixes of Thai women was 5% in Bangkok (Siritantikorn et al., 1997), between 4.8 and 16 percent in Songkla (Chichreon et al., 1998; Sukvirach et al., 2003), and 11% in Khon Kaen (Sriamporn et al., 2006); comprising 61.5% low-grade squamous intraepithelial lesions (LSIL) to 100% high-grade squamous intraepithelial lesions (HSIL) and ICC in Songkla (Sukvirach et al., 2003) and 87% ICC in Khon Kaen (Setheetham-Ishida et al., 2005).

Several previous studies on cervical cancer in Thailand concentrated on sexual behavior risk factors in women or on the presence of HPV DNA in cases vs. control subjects (Chichreon et al., 1998; Thomas et al., 2001; Nunez et al., 2004; Setheetham et al., 2004a; Setheetham et al., 2006). These reports demonstrated that the number of partners was a determinant of risk, and that age at first sexual intercourse was not an independent risk factor.

In our study, the majority of women had only one sexual partner (94.4% of normal women), and age at first sexual intercourse was late (i.e., 90.1, 91.6 and 86.8 percent of normal, CIN and ICC women reporting age over 17). An increase in the risk of both CIN and ICC with age at first sexual intercourse was observed, but the associations were not significant (Table 2). History of STDs was a strong risk factor for an abnormal cervix and may be related to transmission--in this population--of other sexually transmitted agents that can act as co-factors for HPV. This hypothesis is supported by the high frequency of reported histories of venereal diseases.

There was a positive association of cervical cancer risk and parity. Previous studies, both in Thailand (Chichreon et

al., 1998; Thomas et al., 2001) and elsewhere (Ngelandgel et al., 1998) suggest that high parity contributes to the risk, perhaps because of cervical trauma during parturition and/or the hormonal and nutritional influences of pregnancy.

We have confirmed the increased ORs of CIN when the partner(s) had smoking habits either at present or in the past. The harmful effects of smoking are well-known. Detection of tobacco-specific carcinogens in the semen of smoking males (Zenzes et al., 1999) indicates another vector for cervical exposure to carcinogens through sexual contact. Moreover, smoking reduces cervical immunity, which would enhance the persistence of HPV infection (Giuliano et al., 2002). Persistent HPV infection predicts a higher risk for progression, host conditions that compromise immunity, such as multiparity and environmental factors such as smoking. Various gynecologic factors, including age of menarche, number of sexual partners, were suggested in our previous report as risks for HPV infection and CIN or cervical carcinoma with high ORs (Setheetham-Ishida et al., 2004a; Setheetham-Ishida et al., 2005; Setheetham-Ishida et al., 2006).

In conclusion, special attention should be paid to the risk factors of sexual behavior, STDs and smoking as these may function as cofactors which increase risk of cervical cancer in the presence of HPV. Further study involving HPV-DNA testing need to be done.

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