RESEARCH ARTICLE

Lack of Significant Effects of *Chlamydia trachomatis* Infection on Cervical Cancer Risk in a Nested Case-Control Study in North-East Thailand

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Abstract

Cervical cancer continues to be an important public health problem in Thailand. While the high risk human papillomavirus (HPV) types have been established as the principle causative agent of both malignancies and the precursor lesions, cervical intraepithelial neoplasia (CIN), other factors may also be involved like other sexually transmitted diseases, as well as smoking. *Chlamydia trachomatis* is an obligate intracellular Gram-negative bacterium which has a tendency to cause chronic infection featuring inflammation and therefore might be expected to increase the risk of cervical cancer. In the present nested case-control study, 61 cases of cervical cancer and 288 matched controls with original serum samples were identified from the Khon Kaen Cohort, established in the North-East of Thailand, by linkage to the Khon Kaen population based cancer registry. *C. trachomatis* specific IgG antibodies at recruitment were measured by microimmunofluorescence and assessed for association with cervical cancer using STATA release10. No significant link was noted either with all cancers or after removal of adenocarcinomas. The results suggest no association between Chlamydia infection and cervical cancer development in North-East Thailand, but possible influencing factors must be considered in any future research on this topic.

Keywords: Cervical cancer - nested case-control study - *Chlamydia* infection - Northeast Thailand

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Introduction

Cancer of the cervix is one of the most serious public health problems among Thai women (National Cancer Institute et al., 2010). It is now widely accepted that high risk types of human papillomavirus (HPV), particularly HPV 16 and 18, play an important role in the genesis of cervical carcinoma (zur Hausen, 1991). Most HPV infections in the cervix spontaneously resolve and only very few (HPV-infected females develop cervical cancer. The HPV types in Thailand HPV-16, -52, -18, -11, -51, -31 and -33, in that order (Natphosuk et al., 2013; Swangvaree et al., 2013).

Risk factors other than HPV in Thailand include tobacco and sexual behaviour (Settheetham-Ishida et al., 2006, Natphopsuk et al., 2012). An increased risk of cervical cancer associated with tobacco smoking has been found in many studies (Settheetham-Ishida et al., 2006; Yetimalar et al., 2011).

Other possible risk factor for cervical cancer are sexually transmitted diseases, like infection with *Chlamydia trachomatis*. For example serum antibody levels have provided evidence of a strong increase in associated risk of cervical cancer (Dahlstrom et al., 2011).

Since there has been no report of *C. trachomatis* infection effects by longitudinal study in Northeastern Thai women, the present investigation was conducted with the purpose of examining the risk of cervical cancer using a nested case-control approach within the cohort established in Khon Kaen University (Sriamporn et al., 2005).

Materials and Methods

Cases and Controls

Seventy two cases of cervical cancer and two hundred eighty eight matched controls were identified from the Khon Kaen Cohort Study by linkage to the population based cancer registries using personal identification numbers. The descriptive and inferential statistics were analyzed by STATA release10. This study was approved by the Ethical Review Boards in Khon Kaen University.

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C. trachomatis specific IgG antibodies were measured by microimmunofluorescence (Anttila et al., 2001).

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Table 1. Association of *Chlamydia trachomatis* Infection with Cervical Cancer

C. trachomatis test	Cases (n %)		Controls (n %)		OR	(95%CI)	X2 test	P-value
All cases								
IgG positive	11	18.0	36	14.6	1.28	(0.55:2.80)	0.44	0.51
IgG negative	50	82.0	210	85.4	Ref.			
Adenocarcinomas excluded								
IgG positive	10	17.9	35	15.4	1.20	(0.55:2.60)	0.21	0.64
IgG negative	46	82.1	193	84.7	Ref.			

Statistical analysis

The genotypic frequencies between the case and controls were compared using the χ^2-test. The association between selected variables and risk for SCCA were studied using uni- and multi-variate logistic regression analyses (using 800-STATA on PC) and the odds ratio (OR) at the 95% confidence interval (CI) was calculated. Differences were considered statistically significant when the p-value was <0.05.

Results

The total 307 subjects serum available, there are 61 cases and 246 controls. The presence of serum antibodies against *C. trachomatis* was higher among cases than among controls at 18.0% and 14.6% but the difference was not significant (see Table 1). When prevalences were transformed into odds ratio, serum antibodies to *C. trachomatis* were not significantly associated with cervical cancer risk overall or after exclusion of adenocarcinomas.

Discussion

The current nested case-control study did not provide any evidence of a link between *C. trachomatis* infection and later development of cervical cancer. While earlier studies found that a history of *C. trachomatis* infection was associated with cervical cancer (Anttila et al., 2001; Wallin et al., 2002; Smith et al., 2004; Madeleine et al., 2007), no statistically significant link was found with *C.trachomatis* IgG, IgA and IgM seropositivity in a recent study in Turkey (Onel et al., 2013) and there was no association between the presence of *C. trachomatis* DNA in cervical specimens and cervical cancer in Iran (Farivar and Johari, 2012). While the situation might be complicated by the possibility of a squamous cell limitation, *C. trachomatis* DNA being reported as absent in cervical adenocarcinomas (Quint et al., 2009), here we also did not find any association when such lesions were omitted from the analysis.

It may be that *Chlamydia* is only associated with early lesions in the cervix. Thus, although infection did not seem related to cervical cancer in one study in Brazil, women with abnormal cytology had a significant high rate of *C. trachomatis* (de Abreu et al., 2012). A positive *Chlamydia* test was also significantly associated with the risk of having an abnormal cervical cytology in Puerto Rican pregnant women (Seda et al., 2011) and in Greece (Peitsidis et al., 2012). However, hazard ratios were only elevated for development of any CIN grade 2 and not CIN grade 3 or worse (Lehtinen et al., 2011) and a recent case–control study found that *C. trachomatis* did not affect the risk of progression to high grade CIN among HPV-positive women (Safaeian et al., 2010). Furthermore, data are not consistent and in India infection was not significantly associated with CIN, and most of its risk factors, including HPV infection, in symptomatic women (Bhatla et al., 2013). Chlamydia infection as an independent etiologic factor was not significantly associated with CIN relapse (Zivadinovic et al., 2011). However, virtually concomitant HPV18/45 and *C. trachomatis* infections were reported to be linked to very high CIN3 risk (Luostarinen et al., 2013) and it may be that the parasite could act via effects on HPV. Seropositivity was associated with high grade neoplasia in women infected with HPV, mainly when the types 16 and 18 were involved (da Silva-Barros et al., 2011)

Relationships with positive HPV infection have in fact been reported in northeastern Argentina (Deluca et al., 2011), Paraguay (Medoza et al., 2013), Tanzania (Lazenby et al., 2014) and Brazil (Tavares et al., 2014) and there is evidence that *C. trachomatis* infection might increase the probability that infections with high-risk HPV types will become persistent (Silins et al., 2005; Insinga et al., 2011). A role of *C. trachomatis* and number of recent sexual partners was noted for type-specific HPV redetection (Shew et al., 2013) and a causal association between HPV and infection in young women has been proposed, with the bacterium as a predisposing factor for subsequent infection with HPV, or vice versa (Silva et al., 2013).

It is of interest that prospective studies of *C. trachomatis* DNA in archival smears showed risk of cervical cancer be only increased in smears taken more than 6 years before the cancer diagnosis (Wallin et al., 2002). These data suggest that an effect early in carcinogenesis. This is also supported by a study of HPV DNA-positive women that had HPV-persistence as the endpoint, where C. trachomatis history was the only positively associated risk factor for HPV persistence (Silins et al., 2005). Other studies of the possible role of *C. trachomatis* in the etiology of cervical cancer are consistent not with the progression of cervical neoplasia, but possibly with persistence of cervical atypia and persistence of HPV infection (Castle et al., 2003; Golijow et al., 2005; Lehtinen et al., 2011).

With our present nested cancer control design it is not possible to look into these questions. Furthermore, links with other markers of increasing malignancy like p16INK4a (Calil et al., 2011) and effects on host DNA damage and proliferation as well as DNA damage responses (Chumduri et al., 2013) could not be addressed in the present work. Future studies should take these possibilities into consideration to allow a more comprehensive picture of the influence of *C. trachomatis* infection on cervical cancer development in Thailand to be obtained.

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