Prognostic Factors of Human Papillomavirus Genotypes of Invasive Cervical Carcinoma: An Analytical Cross-Sectional Study in Lower North-East Thailand

Metee Wongsena MD**, Phalakorn Suebsamran MS*,
Pawana Panomket PhD*, Sutthini Tirat MD*,
Parichart Wongsena MD*, Surasak Wanram PhD*

** Clinical Research Unit, College of Medicine and Public Health, Ubon Ratchathani University, Ubon Ratchathani, Thailand*
*** Obstetrics and Gynecology Unit, Ubon Ratchathani Cancer Hospital, Ubon Ratchathani, Thailand*

Background: *Cervical cancer (CXCA) caused by persistent infections by high-risk human papillomavirus (HR-HPV) can lead to multi-step carcinogenesis. The best management strategy and significant prognosis for cervical cancer patients remain unclear.*
Objective: *To investigate the associations of the two most common HR-HPVs with clinical outcomes of progression and recurrence status as well as prognosis outcomes of patients.*
Material and Method: *An analytical cross-sectional study of patients registered at Ubon Ratchathani Cancer Hospital was conducted from 2007 to 2010. Clinical data, histopathological features, and clinical outcomes of progression and recurrence status were recorded. HPV type-specific E6/E7 nested multiplex polymerase chain reaction (NMPCR) was performed to identify HR-HPV16 and 18 using extracted deoxyribonucleic acid (DNA) from embedded paraffin. Clinical findings and HPV genotypes were analyzed using Fisher's exact test. Association studies of crucial factors and HR-HPV genotypes were performed using logistic regression analysis (odds ratio [OR]) and 95% confidence interval [CI]). A p-value of less than 0.05 was considered statistically significant.*
Results: *The study found single HPV16 infection in 57.3%, single HPV18 in 17.3%, mixed HR-HPV16/18 in 13.1%, and non-HPV16, 18, or 16/18 in 12.3%. The findings showed significant association among their genotypes and histopathological types and grading ($p<0.0001$ and $p = 0.014$). Clinical outcomes of progression and recurrence status with increased severity of clinical staging were associated significantly ($p = 0.001$ and $p = 0.002$). HPV18 type-specific was shown as a poor prognostic type with its relevance to the severity of disease higher than that of HPV16.*
Conclusion and Discussion: *HPV16 and 18 remain the major type-specifics especially in relation to invasive CXCA, requiring further therapeutic vaccination study and proper prognosis. HR-HPV type-specific is very important during cervical carcinogenesis but other crucial contributing factors for prognostic outcomes should be further elucidated.*

Keywords: *HPV16, HPV18, HPV16/18, Clinical outcomes of progression, Prognosis, Recurrence, Analytical cross-sectional study*

J Med Assoc Thai 2014; 97 (Suppl. 4): S12-S19
Full text. e-Journal: *http://www.jmatonline.com*

Cervical cancer (CXCA) is the second most common female malignant cancer in Thailand and the world[1,2]. Persistent infection by high risk-human papillomavirus (HR-HPV) was documented as an essential factor of viral pathogens driven through multi-step carcinogenesis[3]. HR-HPV16 and 18 genotypes are the two most frequently found genotypes in CXCA associated with histological types of squamous cell carcinoma[4] and adenocarcinoma[5], and are now the major HR-HPV types used in prophylactic and therapeutic vaccines in the treatment of CXCA. Host immune response is a crucial factor during the development of CXCA but the best management strategy for infected women remains unclear.

The present study investigated the contribution of the two most common HR-HPV genotypes to invasive CXCA and their association with

Correspondence to:
Wanram S, College of Medicine and Public Health, Ubon Ratchathani University, 85 Sathollamark Road, Warinchamrab, Ubon Ratchathani 34190, Thailand.
Phone: 08-1545-7311
E-mail: Mdsurawa@ubu.ac.th

the clinical outcomes of progression and recurrence status, and is significant in the future development of prophylactic and therapeutic vaccinations.

Material and Method

Sample and data collection

Clinical data for this analytical cross-sectional study were obtained from 150 randomly-selected patients with invasive CXCA registered as hospital cases between 2007 and 2010 at Ubon Ratchathani Cancer Hospital. Clinical data, including clinical outcomes of progression and recurrence status using Response Evaluation Criteria In Solid Tumors (RECIST) criteria[6] were reported by gynecological experts. The recurrence status was classified into partial response, stable disease, progressive disease, and complete response. The histopathological types and grading of tumors were reviewed by reports by a pathologist consultant. A paraffin-embedded tissue block from each case was studied and each block was subjected to hematoxylin-eosin staining and DNA extraction for HPV genotype detection. This project was approved and given the human ethical approval number HE 09/ 2012, Ubon Ratchathani Cancer Hospital, Ubon Ratchathani, Thailand.

Human papillomavirus deoxyribonucleic acid extraction

The HPV DNA was extracted from invasive CXCA paraffin-embedded tissue by the use of the Wizard® SV genomic DNA extraction kit (Promega, USA). In brief, twenty mg of cervical embedded tissue was examined and mixed in A master-mix of digestion solution [Nucleic lysis solution, 0.5 M EDTA (pH 8.0), proteinase K, and RNase A solution]. The sample tubes were then incubated overnight for 16 to 18 hrs at 55°C in a heat block. Finally, the purified genomic HPV DNA was stored at -20°C. Aliquots of these DNA were concentrated and were measured by Ultraspec® 500/ 1100 pro, Amersham Biosciences.

Screening of human papillomavirus L1 using nested polymerase chain reaction

To confirm DNA subjected to the positive HPV infections, a nested polymerase chain reaction (PCR) assay of HPV L1 consensus region including MY11/ MY09 outer primers was completed, yielding an amplicon of 450 bp[7]. This was followed by $GP5^+/GP6^+$ inner primers yielding an amplicon of 150 bp[8]. Briefly, the first PCR amplification was carried out using conventional L1 consensus primers MY11/ MY09 in a total volume of 25 µL containing 2 µL of each HPV DNA, 200 µM of each dNTP, 1X PCR buffer, 500 nM of each primer, 4.0 mM of $MgCl_2$, and 5U of Taq DNA polymerase. The initial conditions were incubation at 95°C for 10 min, followed by 40 cycles of denaturation at 95°C for 1 min, annealing at 55°C for 1 min, extension at 72°C for 1 min, and a final extension at 72°C for 5 min in a Thermal Cycler. The second nested PCR amplification was subsequently performed using $GP5^+/GP6^+$ general primers in a total volume of 25 µL containing 2 µL of each HPV DNA, 200 µM of each dNTP, 1X PCR buffer, 3.5 mM $MgCl_2$, and 5U of Taq DNA polymerase. The initial conditions were 95°C for 10 min, followed by 35 cycles of denaturation at 94°C for 1 min, annealing at 42°C for 1 min, and extension at 72°C for 30 s. The last cycle was followed by a final extension at 72°C for 5 min in a Thermal Cycler.

Type specific E6/E7 nested multiplex polymerase chain reaction

A PCR-based approach for the sensitive and type-specific detection of HR-HPV infections was developed and modified[9]. The first round of the DNA of all HR-HPV generated products at approximately 600-630 bp. The second round amplifications of NMPCR for HPV genotyping were modified by a reagent mix in two cocktails containing 13 fragments of HR-HPV specific primers by NMPCR. The first PCR amplification was performed using the degenerate GP-E6/E7 primers consisting of GP-E6-3F, GP-E6-5B, and GP-E6-5B in a total volume of 25 µL containing 5 µL of each DNA clinical sample, 200 µM of each dNTP, 1X PCR buffer, 500 nM of each primer, 4.0 mM of $MgCl_2$, and 5U of Taq DNA polymerase. The conditions used for the first round amplifications of NMPCR with E6/E7 consensus primers had the cyclic profile of incubation at 94°C for 4 min followed by 40 cycles of denaturation at 94°C for 1 min, annealing at 40°C for 1 min, and extension at 72°C for 2 min. The last cycle was followed by a final extension step of 10 min at 72°C. The second nested PCR amplification was subsequently performed using a mixture in one cocktail of HR-HPV specific primers for the HR-HPV16 and 18. The total volume was 25 µL containing 5 µL of optimal diluted PCR product of the first round amplification, 200 µM of each dNTP, 1X PCR buffer, 2.5 mM of $MgCl_2$, and 2.5U of Taq DNA polymerase. The second round of NMPCR with type specific primers was performed under the conditions of incubation at 94°C for 4 min followed by 35 cycles of denaturation at 94°C for 30 s, annealing at 58°C for 30 s, and extension at 72°C for 45 s. The last cycle was

followed by a final extension step of 4 min at 72°C. The E6/E7 primers of HPV16 and 18, amplicon size, and their sequences were examined using HPV16E6E7F: 5'-AAAGCCACTGTGTCCTGAAG-3', HPV16E6E7R: 5'-ATTTCATCACCCTCCTCCTC-3', product size 285 bp and HPV18E6E7F: 5'-TATTAATAAGGTGCCTG CGG-3', HPV18E6E7R: 5'- TGTCTTGCAATGTTGC CTTA-3', product size 215 bp, respectively. SiHa and CasKi DNA were used as positive controls for HPV16 and 18 respectively.

Statistical analysis

Overall HPV distribution and average age were determined as percentages of all cases. The clinical findings and histopathological features of all patients with invasive CXCA were compared using Fisher's exact test. The occurrence of the two most common HR-HPV genotypes, the clinical outcomes of progression and recurrence status, and prognostic factors were analyzed using Logistic regression analysis (odds ratio [OR] and 95% confidence interval [CI]). A p-value of less than 0.05 was considered statistically significant. All statistical analyses were performed using the SPSS package for Windows, version 15.0 (SPSS, Inc., Chicago, IL, USA).

Results

Prevalence of the two most common high risk human papillomavirus genotypes

The prevalence of the two most common HR-HPV genotypes in females with invasive CXCA in lower northeast Thailand was determined using E6E7 nested multiplex PCR and is summarized in Table 1. The results showed the median ages of the females at the diagnosis of infections were 57.38±11.7 in the 86 out of 150 (57.3%) cases due to HPV16 and 54.12±13.3 in the 26 (17.3%) cases due to HPV18. The median ages of the females in the 20 cases (13.1%) of infection due to mixed HR-HPV16/18 were 51.35±10.4, and 51.35±10.4 in the 18 (12.3%) cases due to non-HPV16, 18 or 16/18.

Baseline characteristics of patients

One hundred and fifty female patients with invasive CXCA in lower northeastThailand were examined in this analytical cross-sectional study. The patients' ages ranged from 21 to 92 years (mean 55.5±11.7) (Table 1). The use of the International Federation of Gynecology and Obstetrics (FIGO) clinical staging identified early stages of invasiveness in 105 cases and advanced stages in 42 cases (three cases of unknown/not mentioned were classified as missing data). An analysis of the clinical management and histopathological features of the two most common HPV genotypes is shown in Table 2. Other epidemiological data relevant to the clinical treatment of the patients, including co-factors and metastatic involvements, were reported but are not shown.

Human papillomavirus16 and 18 genotypes associated with histopathological features

According to the analytical cross-sectional study, respective significant associations of $p<0.0001$ and $p = 0.014$ and Fisher's exact test between the HR-HPV genotypes and histopathological features, including types and grading, were found (Table 2). The significant associations of HR-HPV and histopathological types were observed with high percentages of 95.3% (82 out of 86 cases) squamous cell carcinoma for HPV16 and 96.2% (25 out of 26 cases) adenocarcinoma for HPV18. Mixed HPV16 and 18 were predominately found in adenocarcinoma.

Clinical outcomes of patients and recurrence status associated with tumor stage

Findings showed significant association among clinical outcomes of progression and CXCA staging at $p = 0.001$ (Table 3). The clinical outcomes of progression increased associated with tumor staging from invasive CXCA stage I to IV (OR up to 21.33). Results showed significant association among recurrence status and CXCA staging at $p = 0.002$ (Table

Table 1. Prevalence of the two most common HPVs and average age of patients with invasive cervical cancer in lower North-East Thailand

HPV genotypes	Prevalence (%)	Age, year (mean ± SD)
Single and mixed infections (n = 150)		
Single HPV16	86 (57.3)	57.38±11.7
Single HPV18	26 (17.3)	54.12±13.3
Mixed HPV16/18	20 (13.1)	51.35±10.4
Non-HPV16, 18 or 16/18	18 (12.3)	53.28±10.4

Table 2. Clinical management and histopathological features of the two most common HPV genotypes in patients with invasive cervical cancer in lower North-East Thailand (TAH = total abdominal hysterectomy, RHND = radical hysterectomy and pelvic lymph node dissection, NA = not mentioned)

HPV genotypes clinical information	HPV16	HPV18	HPV16 and 18	p-value (Fisher's exact test)
Surgery for primary (n = 129)				0.137
No.	53 (62.4)	11 (44.0)	8 (42.1)	
Primary surgery	22 (25.9)	11 (44.0)	10 (52.6)	
Adjuvant surgery	10 (11.8)	3 (12.0)	1 (5.3)	
Procedure (n = 132)				0.222
TAH	9 (10.5)	4 (15.4)	1 (5.0)	
RHND	23 (26.7)	10 (38.5)	10 (50.0)	
Unknown	54 (62.8)	12 (46.2)	9 (45.0)	
Pathology (margin) (n = 132)				0.255
Free margin	24 (27.9)	13 (50.0)	8 (40.0)	
Not free margin	7 (8.1)	1 (3.8)	2 (10.0)	
Unknown	55 (64.0)	12 (46.2)	10 (50.0)	
Radiotherapy (n = 132)				0.192
No.	18 (20.9)	10 (35.8)	7 (35.0)	
Primary RT	60 (69.8)	15 (57.7)	10 (50.0)	
Post-op RT	8 (9.3)	1 (3.8)	3 (15.0)	
Chemo regimen (n = 132)				0.442
No.	29 (34.1)	13 (52.0)	11 (55.0)	
Concurrence	43 (50.6)	9 (36.0)	6 (30.0)	
Neo-adjuvant	10 (11.8)	2 (8.0)	2 (10.0)	
Adjuvant	3 (3.5)	1 (4.0)	1 (5.0)	
Histopathological types (n = 132)				<0.0001
Squamous cell carcinoma	82 (95.3)	1 (3.8)	5 (25.0)	
Adenocarcinoma	4 (4.7)	25 (96.2)	15 (75.0)	
Histopathological grading (n = 132)				0.014
Well	5 (5.8)	7 (26.9)	6 (30.0)	
Moderately	15 (17.4)	6 (23.1)	2 (10.0)	
Poorly	9 (10.5)	3 (11.5)	3 (15.0)	
NA	57 (66.3)	10 (38.5)	9 (45.0)	

Table 3. Clinical outcomes of progression among stages in patients with invasive cervical cancer in lower North-East Thailand (*Reference group, Logistic regression analysis (odds ratio [OR] and 95% confidence interval [CI])

Results FIGO staging	Clinical outcome of progression		OR	95% CI	p-value
	Progression (%)	Non-progression (%)			
Invasive CXCA (n = 147)					0.001
Stage I*	6 (18.8)	64 (55.7)	1.00		
Stage II	11 (34.4)	24 (20.9)	4.88	1.62-14.68	
Stage III	13 (40.6)	26 (22.6)	5.33	1.83-15.53	
Stage IV	2 (6.2)	1 (0.9)	21.33	1.67-271.0	

4). The recurrence status increased and was associated with tumor staging. Findings showed invasive CXCA stages II and III had higher recurrence status than stage I by OR equal to 6.78 and 4.48, respectively. Results did not show significant associations among HR-HPV genotypes and clinical outcomes of progression as

Table 4. Recurrence statuses among stages in patients with invasive cervical cancer in lower North-East Thailand (*Reference group, **Recurrence status: recurrence as partial response, stable disease and progressive disease; non-recurrence as complete response; [d] some patients died or did not come to see the doctor, so we could not find the recurrence status)

Results FIGO staging	Recurrence statuses**		OR	95% CI	p-value
	Recurrence (%)	Non-recurrence (%)			
Invasive CXCA (n = 147)					0.002
Stage I*	5 (18.5)	65 (54.2)	1.00		
Stage II	12 (44.4)	23 (19.2)	6.78	2.15-21.34	
Stage III	10 (37.1)	29 (24.2)	4.48	1.40-14.29	
Stage IV	- (-)[d]	3 (2.5)	0.00	0.00-0.00	

well as recurrence status, but increased progression and recurrence status of single HPV18 were higher than single HPV16 and mixed HPV16/18 by OR equal to 1.84, 1.32, and 1.25, respectively (data not shown).

Discussion

Persistent infection by HR-HPV was characterized as the most significant risk factor for CXCA[10,11]. The majority of HR-HPV-induced cervical cancer development arises in a multi-step carcinogenesis[12,13]. The results of this study showed similar prevalence to other studies of HR-HPV16 and 18[14-16], especially the high prevalence of HPV18 and increase in Thailand as indicated by Siriaunkgul S et al[17]. The results of the study that is the subject of this paper strongly suggested that both single and mixed HPV16 and HPV18 were required for invasive CXCA development, meaning that the use of therapeutic vaccine independently and in combination between HPV16 and HPV18-E6E7 should be elucidated for cervical therapy.

The present study's findings regarding the two most common genotypes HR-HPV16 and 18 showed single HPV16 had the highest prevalence of HR-HPV and associated CXCA, especially for the squamous cell carcinoma antigen (SCCA) type, followed by single HPV18 associated with the adenocarcinoma type. This is similar to the findings of Bulk et al[18] that revealed that HPV18 was mainly a risk factor for the development of adenocarcinoma whereas HPV16 was associated with both SCCA and adenocarcinoma. HPV18 was shown to increase poor prognosis more than other HR-HPVs, a finding similar to that of Kang et al 2011 and Intaraphet et al, 2013[19,20]. The results of the study that is the subject of this paper did not generally show the association of clinical outcomes of progression and recurrence status with HPV occurrence, but OR did show approximately 50% of progression and 70% of recurrence by single HPV18 occurrence. The researchers noticed that the HPV18 genotype is a reliable prognostic factor of early-stage invasive CXCA recurrence. With the increased severity of invasive CXCA results, significant associations of the clinical outcomes with clinical staging were displayed but the researchers were unable to explain the reasons for this. Because of this, not only epidemiologic clinical observation and HPV genotypes-specific experimental association but also physical state with it load of specific HPV, such as HPV16[21] and host immune responses including molecular approaches and epigenetics alteration, are important. Also, significant prognostic clinical outcomes followed by procedures of treatments and analysis of disease-free survival due to various crucial contributing factors of association with enhancement of cervical cancer carcinogenesis should be further investigated.

Conclusion

The two most common genotypes HR-HPV16 and 18 are very important as type-specific of invasive CXCA. HPV18 is a poorer prognostic type than 16 with relevance to the severity of disease. These genotypes can be used in the prediction of prognostic clinical outcomes, but further efficient prognostic factors, such as contributing oncogenic HPV and host immune response including genetics and epigenetics alteration, are required.

Funding

Financial supports were provided by grants from the National Research Council of Thailand year 2011-2012 and Anandamahidol Foundation Thailand Affairs.

Ethical approval

Ethical clearance was provided by Ubon Ratchathani Cancer Hospital HE No. 09/2012.

Authors' contributions

All the authors contributed to the design of the paper. Dr. Metee Wongsena carried out the clinical work as the gynecologist, Dr. Parichart Wongsena acted as the pathologist, and Assistant Professor Dr. Pawana Panomket, Dr. Sutthini Thirat and Mr. Phalakorn Suebsamran were responsible for the laboratory tests and statistical analysis respectively. All authors contributed to the writing-up of the paper led by Assistant Professor Dr. Surasak Wanram.

Acknowledgement

The authors are grateful for the participation and data of the patients with cervical cancer at Ubon Ratchathani Cancer Hospital and to the College of Medicine and Public Health, Ubon Ratchathani University, Ubon Ratchathani, Thailand. Appreciation is expressed to the staff of the Office of International Relations at Ubon Ratchathani University for assistance with English.

Potential conflicts of interest

None.

References

1. Ferlay J, Bray F, Pissani P, Parkin DM. GLOBOCAN 2002: cancer incidence, mortality and prevalence worldwide. IARC Cancer Base No. 5, version 2.0. Lyon, France: IARC Press, 2004.
2. Sriamporn S, Snijders PJ, Pientong C, Pisani P, Ekalaksananan T, Meijer CJ, et al. Human papillomavirus and cervical cancer from a prospective study in Khon Kaen, Northeast Thailand. Int J Gynecol Cancer 2006; 16: 266-9.
3. Berek JS, Hacker NF. Gynecologic oncology. Philadelphia: Lippincott Williams & Wilkins; 2010.
4. Ferlay J, Shin HR, Bray F, Forman D, Mathers C, Parkin DM. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. Int J Cancer 2010; 127: 2893-917.
5. Wang SS, Sherman ME, Hildesheim A, Lacey JV Jr, Devesa S. Cervical adenocarcinoma and squamous cell carcinoma incidence trends among white women and black women in the United States for 1976-2000. Cancer 2004; 100: 1035-44.
6. Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, et al. New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. J Natl Cancer Inst 2000; 92: 205-16.
7. Gravitt PE, Peyton CL, Apple RJ, Wheeler CM. Genotyping of 27 human papillomavirus types by using L1 consensus PCR products by a single-hybridization, reverse line blot detection method. J Clin Microbiol 1998; 36: 3020-7.
8. Roda Husman AM, Walboomers JM, Hopman E, Bleker OP, Helmerhorst TM, Rozendaal L, et al. HPV prevalence in cytomorphologically normal cervical scrapes of pregnant women as determined by PCR: the age-related pattern. J Med Virol 1995; 46: 97-102.
9. Sotlar K, Diemer D, Dethleffs A, Hack Y, Stubner A, Vollmer N, et al. Detection and typing of human papillomavirus by e6 nested multiplex PCR. J Clin Microbiol 2004; 42: 3176-84.
10. Meijer CJ, van den Brule AJ, Snijders PJ, Helmerhorst T, Kenemans P, Walboomers JM. Detection of human papillomavirus in cervical scrapes by the polymerase chain reaction in relation to cytology: possible implications for cervical cancer screening. IARC Sci Publ 1992; 271-81.
11. Bosch FX, Munoz N, de Sanjose S, Franco EL, Lowy DR, Schiffman M, et al. Re: Cervical carcinoma and human papillomavirus: on the road to preventing a major human cancer. J Natl Cancer Inst 2001; 93: 1349-50.
12. Ostor AG. Studies on 200 cases of early squamous cell carcinoma of the cervix. Int J Gynecol Pathol 1993; 12: 193-207.
13. Arends MJ, Buckley CH, Wells M. Aetiology, pathogenesis, and pathology of cervical neoplasia. J Clin Pathol 1998; 51: 96-103.
14. Clifford GM, Smith JS, Aguado T, Franceschi S. Comparison of HPV type distribution in high-grade cervical lesions and cervical cancer: a meta-analysis. Br J Cancer 2003; 89: 101-5.
15. Munoz N, Bosch FX, de Sanjose S, Herrero R, Castellsague X, Shah KV, et al. Epidemiologic classification of human papillomavirus types associated with cervical cancer. N Engl J Med 2003; 348: 518-27.
16. Suthipintawong C, Siriaunkgul S, Tungsinmunkong K, Pientong C, Ekalaksananan T, Karalak A, et al. Human papilloma virus prevalence, genotype distribution, and pattern of

infection in Thai women. Asian Pac J Cancer Prev 2011; 12: 853-6.
17. Siriaunkgul S, Utaipat U, Suthipintawong C, Tungsinmunkong K, Triratanachat S, Khunamornpong S. HPV genotyping in adenocarcinoma of the uterine cervix in Thailand. Int J Gynaecol Obstet 2013; 123: 226-30.
18. Castellsague X, Diaz M, de Sanjose S, Munoz N, Herrero R, Franceschi S, et al. Worldwide human papillomavirus etiology of cervical adenocarcinoma and its cofactors: implications for screening and prevention. J Natl Cancer Inst 2006; 98: 303-15.
19. Kang WD, Kim CH, Cho MK, Kim JW, Cho HY, Kim YH, et al. HPV-18 is a poor prognostic factor, unlike the HPV viral load, in patients with stage IB-IIA cervical cancer undergoing radical hysterectomy. Gynecol Oncol 2011; 121: 546-50.
20. Intaraphet S, Kasatpibal N, Siriaunkgul S, Sogaard M, Patumanond J, Khunamornpong S, et al. Prognostic impact of histology in patients with cervical squamous cell carcinoma, adenocarcinoma and small cell neuroendocrine carcinoma. Asian Pac J Cancer Prev 2013; 14: 5355-60.
21. Wanram S, Limpaiboon T, Leelayuwat C, Yuenyao P, Guiney DG, Lulitanond V, et al. The use of viral load as a surrogate marker in predicting disease progression for patients with early invasive cervical cancer with integrated human papillomavirus type 16. Am J Obstet Gynecol 2009; 201: 79-7.

ปัจจัยพยากรณ์โรคกับไวรัสฮิวแมนแปบปิลโลมาชนิดที่มีความเสี่ยงสูงของผู้ป่วยมะเร็งปากมดลูกในภาคตะวันออกเฉียงเหนือตอนล่าง: การศึกษาเชิงวิเคราะห์แบบตัดขวาง

เมธี วงศ์เสนา, พลากร สืบสำราญ, ภาวนา พนมเขต, ศุทธินี ธิราช, ปาริชาติ วงศ์เสนา, สุรศักดิ์ แว่นรัมย์

ภูมิหลัง: *มะเร็งปากมดลูกมีสาเหตุสำคัญจากการติดเชื้อไวรัสฮิวแมนแปบปิลโลมา ชนิดที่มีความเสี่ยงสูงนำมาซึ่งกระบวนการเกิดมะเร็งที่หลากหลาย อย่างไรก็ตามกลยุทธ์ที่ดีที่สุดในการดูแลรักษาผู้ป่วยที่ติดเชื้อจนพัฒนาเป็นมะเร็งดังกล่าว โดยเฉพาะต่อพยากรณ์โรคที่มีนัยสำคัญยังไม่กระจ่าง*

วัตถุประสงค์: *ศึกษาความสัมพันธ์ระหว่างไวรัสฮิวแมนแปบปิลโลมาชนิดที่มีความเสี่ยงสูงที่พบมากที่สุดสองอันดับแรก กับผลการรักษาทางคลินิกต่อพยากรณ์โรคในแง่ของการดำเนินโรคและการกลับเป็นซ้ำ ของผู้ป่วยมะเร็งปากมดลูก ภาคตะวันออกเฉียงเหนือตอนล่างของประเทศไทย*

วัสดุและวิธีการ: *ศึกษาเชิงวิเคราะห์แบบตัดขวาง ในกลุ่มผู้ป่วยที่มารับการรักษาที่โรงพยาบาลศูนย์มะเร็ง อุบลราชธานี ในระหว่าง ปี พ.ศ. 2550-2553 โดยข้อมูลทางคลินิก, พยาธิวิทยาเนื้อเยื่อและผลการรักษา ที่ประกอบด้วยการดำเนินโรคและสถานะปัจจุบันต่อการกลับเป็นซ้ำของผู้ป่วยได้รับการบันทึกไวรัสฮิวแมนแปบปิลโลมาชนิดที่มีความเสี่ยงสูงถูกตรวจหาแบบจำเพาะจาก E6/E7 ด้วยวิธีการเพิ่มปริมาณดีเอ็นเอแบบ NMPCR จากบล็อกชิ้นเนื้อตัวอย่าง ข้อมูลทั้งหมดถูกวิเคราะห์ทางสถิติตามลักษณะของข้อมูล ทั้งเชิงพรรณนา ด้วยการหาร้อยละ ค่าเฉลี่ย และเชิงวิเคราะห์เพื่อหาความสัมพันธ์และความแตกต่างด้วย Fisher's exact test กับ Logistic regression analysis ซึ่งพิจารณาค่าความแตกต่างอย่างมีนัยทางสถิติที่ค่า p-value น้อยกว่า 0.05*

ผลการศึกษา: *การศึกษาใน 150 ตัวอย่างผู้ป่วย ตรวจพบ single HPV16 และ HPV18 ร้อยละ 57.3 และ 17.3 ตามลำดับ และในกรณี mixed 16/18 พบร้อยละ 13.1 ในขณะที่ไม่ใช่สองชนิดดังกล่าวพบร้อยละ 12.3 ในการศึกษาครั้งนี้พบความสัมพันธ์ระหว่างชนิด ของไวรัสที่มีความเสี่ยงสูงกับผลพยาธิวิทยาเนื้อเยื่อ และระดับของพยาธิวิทยามะเร็งอย่างมีนัยสำคัญทางสถิติ ที่ค่า p<0.0001 และ p = 0.014 ตามลำดับ พบความสัมพันธ์ระหว่างข้อมูลทางคลินิกในการดำเนินโรคและสถานการณ์กลับเป็นซ้ำของผู้ป่วย กับระยะของโรคที่รุนแรงมากขึ้นอย่างมีนัยสำคัญทางสถิติ ที่ค่า p = 0.001 และ p = 0.002 การศึกษาได้แสดงให้เห็นว่า HPV18 เป็นเสมือนชนิดที่บ่งชี้พยากรณ์โรคที่รุนแรงมากกว่า HPV16 เมื่อพิจารณาจากค่า Odds ratio*

สรุป: *HPV16 และ HPV18 เป็นสองชนิดที่พบมากและมีความสำคัญต่อพัฒนาการของโรคของผู้ป่วยมะเร็งปากมดลูกระยะลุกลาม ซึ่งมีความสำคัญต่อการศึกษาวัคซีนที่ใช้บำบัดรักษาในอนาคต และมีความสำคัญต่อพยากรณ์โรคอย่างมีประสิทธิภาพถึงแม้ว่าชนิดของไวรัสที่มีความเสี่ยงสูงมีความสำคัญดังกล่าวแต่ปัจจัยอื่นๆ ที่มีความสำคัญ ซึ่งมีผลต่อข้อมูลพยากรณ์โรคของผู้ป่วยควรได้รับการศึกษาในอนาคต*