RESEARCH COMMUNICATION

Histopathological Findings for Cervical Lesions in Malaysian Women

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Abstract

   <u>Objective</u>: The objective of this study was to determine the histopathological features and cell morphology of various cervical lesions observed among Malaysian women. <u>Methodology</u>: A retrospective study was conducted to evaluate 77 cervical cases collected from the histopathology laboratory of Ipoh hospital from 1st January, 2005, to 31st December, 2006. <u>Results</u>: Cervical intraepithelial neoplasia (CIN) was found in 33 (42%) cases, CIN III accounting for 27%, and CIN I, CIN II and CIN II-III 5% each. The highest rate for CIN cases was 43% in the 41-50 year age group and the lowest rate was 6% in the group aged 61-70 years . Non-keratinizing and metastatic squamous cell carcinomas (SCCs) accounted for 16% and 13%, respectively, the combination being second in majority (29%), followed by adenocarcinoma (17%). The histopathological results showed CIN I to be characterized by mild papillary projections of the epithelium with some degree of nuclear enlargement, pleomorphism, mild koilocytosis, bionucleated cells and a low nucleo-cytoplasmic ratio. CIN II demonstrated typical squamous epithelium with disorganization of the lower part of the epithelium accompanied by nuclear hyperchromatism, an increased nucleo-cytoplasmic ratio, and scanty mitotic figures. CIN III was characterized by pleomorphic nuclei, atypical cells with mitotic figures, nucleo-cytoplasmic ratio, anisokaryosis and hyperchromasia. <u>Conclusion</u>: Lesions related to cervical cancer showed tumor progression correlating with histopathological changes in cell morphology.

Key words: Cervical cancer - cancer cell morphology - Malaysia

Introduction

Cervical cancer (CC) is one of the most common cancers in women, being the second top cancer affecting females in Malaysia after breast cancers (Ferlay et al., 2004). The standardized incidence rate of cervical cancer among Malaysian women has been reported to be 19.7/100,000 population (National Cancer Registry, 2003, Efren et al., 2008). A Pap smear coverage of only around 30-40% of women in Malaysia contributes to this problem (National Cancer Registry, 2003).

For the past 20 years the presence of high-risk human papilloma virus (HPVs) genotypes has been associated with cervical dysplasia and its progression to cancer. This is now being used as an adjunct to detect cervical lesions in conjunction with the Pap smear (Solomon et al., 2002). Human papillomavirus (HPV) infection is now considered to be the main cause of CIN and cancer, and it is one of the most common sexually transmitted diseases in the world (Sedlacek, 1999; Kjellberg et al., 2000; van der Graaf et al., 2002). Human papillomavirus is one of the most common sexually transmitted infections in sexually active adolescent girls and young women of several economically developed countries (Richardson et al, 2003; Syrjänen et al, 2005; Kitchener et al, 2006; Dunne et al,

2007). The association between certain HPVs and cervical cancer is well documented and research over the past 20 years ago has revealed that the virus is etiologically related to the development of most cases of cervical cancer (Franco, 1991; Bosch et al., 1995). Strong evidence has been observed for the role of persistent high-risk HPV types 16 and 18 in the etiology of cervical cancer, as worldwide they are responsible for approximately 70% of all cases (Clifford et al, 2006; Markowitz et al, 2007).

The invasive phase of cervical cancer is preceded by an intraepithelial phase (cervical precursor lesion/intraepithelial neoplasia, CIN), and not all women who develop these precursor lesions will have invasive carcinoma in the future. The traditional concept regarding the natural history of cervical cancer considers CIN1, CINII and CIN III to be stages of a single progressive disease (Schiffman, 1995). A definitive diagnosis is obtained by cervical biopsy and examination of the stained tissue. The predictive value of a biopsy is higher than that of the Pap test because the anatomical arrangement is preserved allowing evaluation of pathological features in relation to histological architecture (Yeoh and Chan, 1997).

Therefore, the objective of this study was to determine the histological features and cancer cell morphology of various cervical lesions among Malaysian women.

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Materials and Methods

Retrospective study was conducted at the end of 2007, data was collected from histopathology laboratory of Ipoh hospital from 1st January 2005 to 31st December 2006 to evaluated 77 cervical biopsies were obtained from the patients. Histopathological findings were classified as normal or presenting of CIN I, CIN II, CIN III and HPV according to the cellular morphology criteria that described by Richart (1990). Briefly, flat lesions with strictly defined koilocytic atypia and no evidence of proliferation were considered to represent HPV infection. Mild nuclear atypia with minimal proliferation was graded as CIN1. Moderate atypia and proliferation (2/3 of the epithelium) was graded as CIN II and severe nuclear atypia with intense proliferation (full thickness of the epithelium) was graded as CIN III. Invasive squamous carcinomas and glandular lesions were excluded (Godoy et al., 2008). The biopsy specimens were processed by fixation, dehydration and staining, then examined under light microscopy. The data were analyzed using the software program Excel.

Results

Incidence and Demographic Analysis

Seventy seven cases were studied and classified based on the histopathology findings, and analysed based on cervical lesion categories. The majority of cases, 33 (42%), were cervical intraepithelial neoplasia (CIN): CIN III with 21 (27%), and CIN I, CIN II and CIN II-III with four cases (5%) each. Non-keratinizing squamous cell carcinoma and metastatic squamous cell carcinoma (MSCC) accounted for 12 (16%) and 10 (13%) respectively. Well, moderate and poorly differentiated adenocarcinomas were identified in 4 (5%), 3 (4%) and 6 (8%), respectively. Another nine different cases diagnosed as human papillomavirus positive, 6 (8%), while Condylomas acuminate and incomplete squamous metaplasia had two and one cases respectively (Table 1).

Regarding age, the patients' age was between 21 to 100 years old. The highest CIN rate was 14 (43%) out of 33 cases in the age group of 41-50 years old and the lowest rate was two (6%) in age groups of 51-60, 61-70 years old. The squamous cell carcinoma showed the second
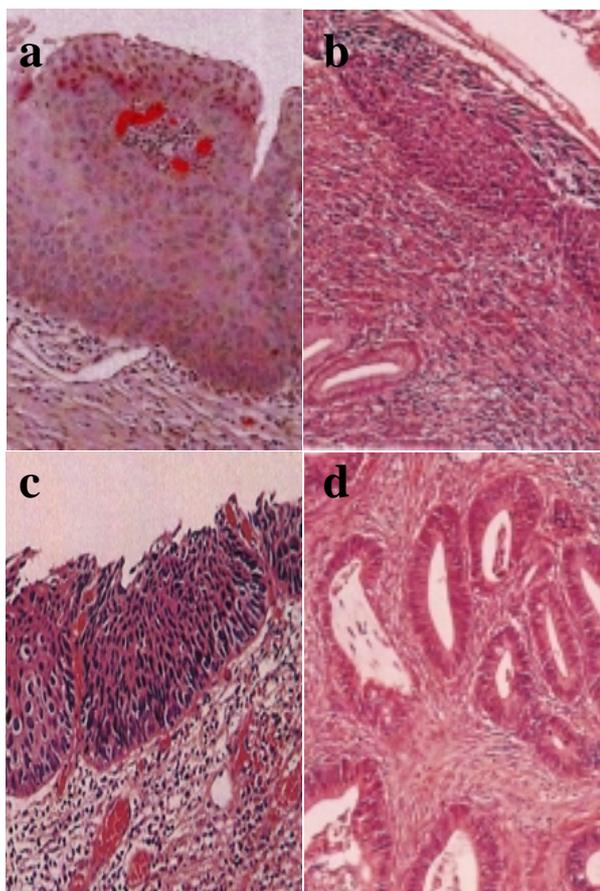
Figure 1. Histopathology Photographs of H&E Stained Cervical Lesions. a) CIN I showing mild dysplasia, pleomorphism and koilocytosis of the cervix epithelium x10; b) CIN II showing typical disorganization of the epithelium accompanied by nuclear hyperchromatism, increased nucleo-cytoplasmic ratio and mild pleomorphism x10; c) showing the epithelium replaced by enlarged pleomorphic nuclei, atypical cells with mitotic figures and koilocytic changes above the basal layers, anisokaryosis and hyperchromasia. x 20; d) fibrous stroma with features of invasive change in an adenocarcinoma consisting of crowded glands and papillary structure with a complex pattern of growth x 20.

majority on cervical cancer incidence of 22 cases (29%). The highest rate showed in the period of 41-50 years with 8 cases (36%), while the lowest rate was in age period of 31-40 and 91-100 years old. Adenocarcinoma was recorded the third cervical cancer, where the highest rate

Table 1. Numbers and Percentages of Cervical Lesion Patients according to Stages of Severity (n=77)

Type of Cervical Lesion	Case number	%
Non-keratinized SCC	12	15.6
Metastatic SCC	10	13.0
CIN I	4	5.2
CIN II	4	5.2
CINII-III	4	5.2
CIN III	21	27.3
Well Differentiated Adenocarcinoma	4	5.2
Moderately Differentiated Adenocarcinoma	3	3.9
Poorly Differentiated Adenocarcinoma	6	7.8
Human Papillomavirus Lesions (HVPs)	6	7.8
Condylomas Acuminate (CA)	2	2.6
Incomplete Squamous Metaplasia (ISM)	1	1.3

Table 2. Numbers and Percentages of Cervical Lesions according to Age Groups (n=77)

Age Group	CIN	SCC	AC	HVPs	CA	ISM
21-30	3 (9)	-	-	2 (33)	-	1
31-40	12 (6)	1 (5)	4 (31)	4 (67)	2	-
41-50	14 (43)	8 (36)	1 (8)			
51-60	2 (6)	4 (18)	6 (46)	-	-	-
61-70	2 (6)	3 (8)	2 (15)	-	-	-
71-80	-	5 (27)	-	-	-	
81-90	-	-	-	-	-	-
91-100	-	1 (5)	-	-	-	-
Total	33	22	13	6	2	1

AC, adenocarcinoma; CA, Condylomas acuminate; CIN, cervical intraepithelial neoplasia; ISM, Incomplete squamous metaplasia; SCC, squamous cell carcinoma

was 6 cases (46%) in aged of 51-60 years and lowest was one case (8%) in age group 41-50 years old. The HPVs, condylomas acuminate and incomplete squamous metaplasia showed lowest rate among cervical cancer and the majority of them was in age 3-40 years (Table 2).

Histopathology

The histopathology results showed CIN I with mild papillary projections of the epithelium with some degree of nuclear enlargement, pleomorphism and disorganized of basal layer, mild koilocytosis, binucleated cells and low of nucleo-cytoplasmic ratio are seen (Figure 1a).

The CIN II showed typical disorganization of the lower part of the epithelium accompanied by nuclear hyperchromatism, increased nucleo-cytoplasmic ratio and mild pleomorphism, scanty mitotic figures and binucleated cells. The superficial layers of the epithelium showed the spindling of the cells but there was no koilocyte and microinvasive carcinoma (Figure 1b). The main histopathological changes in CIN II-III were consisted in the replacement of epithelium which involving half to full thickness equivalent the CIN II changing to CIN III. The cells have enlarged nuclei with coarse chromatin pattern and prominent nucleoli. The nuclei-cytoplasmic ratio was moderate to high, few mitotic figures, also there was enlarged the nuclei, pleomorphism and hyperchromatism with disorganization of the basal layers.

Microinvasive carcinoma had a maximum depth of about two millimeters deep but there was no lymphatic channel invasion (Figure 1c). The histopathology of squamous cell carcinoma (SCC) showed tumor tissues composed of lobules of papillary structures together with the configuration of condyloma which lined with stratified squamous epithelium with large, pale vesicular nuclei and relatively low mitotic figures. The superficial part of the tumor was no conclusive of invasion changes except small infiltrative islands of tumors cells were seen. In the no-keratinizing squamous cell carcinoma, the tumor tissue showed well squamous differentiation which consisted of irregular trabeculae and aggregates of malignant epithelial cells infiltrating the stroma. The neoplastic cells showed with hyperchromatic and pleomorhpic nuclei with some mitotic figures.

Histopathological of adenocarcinoma showed the fragments of tumor tissues which appeared to be polypoidal and exophytic, these consisted of packed and some confluent glands. In some other part, fibrous stromas with features of invasive changes, also the tumor tissue consisted of crowded glands and papillary structure with a complex pattern of growth. The cytological atypia showed pleomorphic and hyperchromatic with obvious mitotic figures (Figure 1d).

Rare cases were found such as incomplete squamous metaplasia which showed with no significant pathological changes of hyperchromatic, nuclear pleomorphisim and mitotic figures. Wart-like papillomatous lesions that resembled condylomata were characterized by hyperplasia of the squamous epithelium with marked cytoplasmic vaculation (koilocytosis), nuclear chromatin condensation and nuclear atypia.

Discussion

Cervical cancer is one of the most common cancers in women, which involved reversible changes in the cervical tissue leading to various cellular abnormalities and ultimately to cervical cancer (Kisseljov et al., 2008). The results of this study showed that the CIN (42%) was the highest incidence among cervical cancer. The severity of cervical cancer considers CIN1, CINII and CINIII to be staged of a single progressive disease, studies have shown that in most women with a CIN II or CIN III, CIN1 being the morphological manifestation of HPV infection, while CIN II/CIN III being the true pre-malignant of cervical cancer pathological changes addition to the morphological manifestation of HPV infection (Schiffman, 1995), each stage was characterized by specific morphological changes (Kisseljov et al., 2008). The cervical screening program includes Pap smear, colposcopy and tissue biopsy in women aged 25–55 years (Domingo et al., 2008).

The present study results showed that the squamous cell carcinoma (29%) was second majority. The adenocarcimoma was showed (17%) the third majority on cervical cancer in different cervical neoplasia stages. Cervical carcinogenesis associated with infection of high-risk human papillomaviruses contains several early genes that are necessary for viral replication, those genes (E6 and E7) play a key role in the induction of cervical carcinogenesis. By multifunctional and participate in many cellular functions associated with cell proliferation (Kisseljov et al., 2008). Epithelial cells receive important stimuli from the environment through soluble growth factors and insoluble extracellular matrix proteins. Extracellular matrices in concert with growth factors can profoundly influence cell phenotypes and behaviors (Kuphal et al., 2005).

Human papillomavirus types 16 and 18 have been categorized as human carcinogens based on their strong associations with cervical cancer in previous studies. Another studies showed strong associations between invasive cervical cancer and less common HPV types. Overall HPV prevalence was 97% in the cervical cancer cases and 20% in the control subjects. Strong associations were found between invasive cervical cancer and specific HPV types (Pedro et al., 2000). But the result of this study was no agreed with previous study where the human papillomavirus showed only 8% among the cervical cancer.

The cervical cancer is the first cancer that can be effectively prevented by vaccination (Kisseljov et al., 2008). The Malaysia Drug Authority approved the use of the quadrivalent HPV vaccine (Gardasil®, Merck & Co., Inc., Whitehouse Station, NJ, USA) in October 2006, but its use is exclusively in private health centers. A National Immunization Technical Committee under the Disease Control Division of the Malaysian Ministry of Health has been given the responsibility to study and make recommendations on the role of the HPV vaccine in Malaysia by 2009. If HPV vaccination is integrated into this program, the target population should be extended to

include girls and women aged 11–24 years, and those who have not been vaccinated or have not completed the full course (Domingo et al., 2008).

In conclusion, the CIN was the highest rate of CC, SCC showed the second majority and third majority was adenocarcimoma on cervical cancer. The cervical cancer showed association diversity of tumor progression correlation with histopathological cancer cells morphology.

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