

CHAPTER IV

RESULTS

During the study period (January 1992 to June 1998), there were 36 recurrent cases who met all eligible criteria (33 from Maharaj Nakorn Chiangmai Hospital and 3 from BMA Medical College and Vajira Hospital). Among these, pathological blocks of invasive cervical cancer were not available in 4 cases, all from Maharaj Nakorn Chiangmai Hospital [radical hysterectomy specimens had no residual tumor in 1 case (patient underwent conization at other hospital) and pathological blocks were not available in 3 cases].

Hence, we included 32 recurrent cases in this study. The diagnosis of recurrence were pathologically confirmed in 19 cases (59.4%), imaging confirmed in 6 cases (18.8%), while in 7 cases (21.9%), the diagnosis was based solely on physical examination. Locoregional recurrence only was found in 27 cases (84.4%). Four cases (12.5%) had distant recurrence, and 1 case (3.1%) had both locoregional and distant recurrence.

We were able to find 2 matching controls for 31 cases, while the other case (44-year old patient, stage Ib adenocarcinoma with tumor of 6 cm.) had only one matching control. Altogether, we recruited 63 controls in this study. (57 from Maharaj Nakorn Chiangmai Hospital and 6 from BMA Medical College and Vajira Hospital) More than 80% of controls were operated within 2 years compared to cases.

4.1 Patients' basic characteristics

The basic characteristics of cases and controls were demonstrated in Table 4.1. The mean age \pm SD of the cases and controls were 41.5 ± 7.4 and 41.3 ± 6.7 years respectively. About 94% were in stage Ib and 63% had squamous cell carcinoma. The mean tumor size \pm SD of cases and controls were 2.7 ± 1.6 cm. and 2.6 ± 1.4 cm. respectively.

Table 4.1 Basic characteristics of cases and controls.

	Cases (n=32)		Controls (n=63)	
	Number	%	Number	%
Age (years)				
21-30	-	-	2	3.2
31-40	19	59.4	31	49.2
41-50	8	25.0	24	38.1
51-60	5	15.6	4	6.3
61-70	-	-	2	3.2
Mean \pm SD	41.5 \pm 7.4		41.3 \pm 6.7	
Stage				
Ib	30	93.7	59	93.7
Ila	2	6.3	4	6.3
Tumor characteristics				
No definite lesion	3	9.4	7	11.1
Exophytic	17	53.1	34	54.0
Ulceroinfiltrative	12	37.5	22	34.9
Histology				
Squamous	20	62.5	40	63.5
Adenocarcinoma	9	28.1	19	30.2
Adenosquamous	3	9.4	4	6.3
Tumor size (cm.)				
≤ 1	5	15.6	10	15.9
>1-2	7	21.9	18	28.6
>2-3	12	37.5	16	25.4
>3-4	2	6.2	12	19.0
>4-5	4	12.5	7	11.1
>5-6	2	6.3	-	-
Mean \pm SD	2.7 \pm 1.6		2.6 \pm 1.4	

4.2 Immunoreactivity of bcl-2 expression

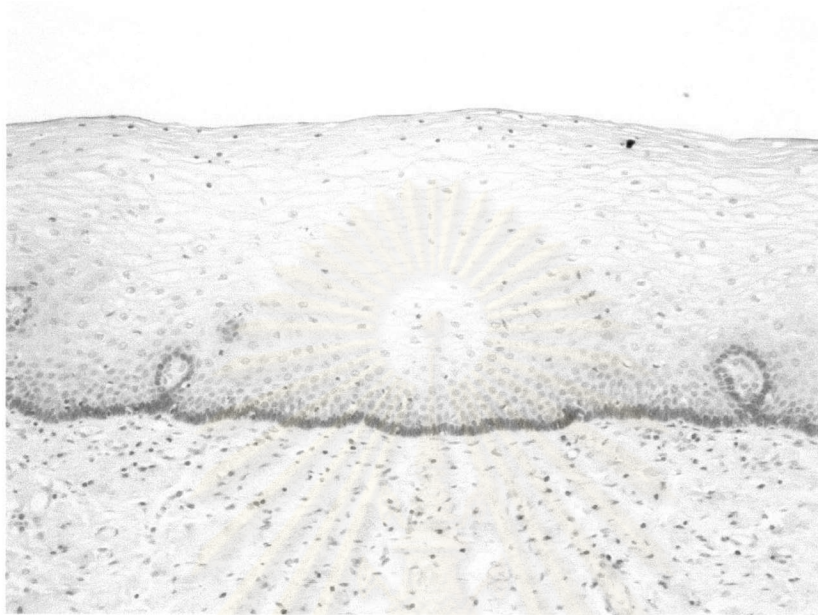


Figure 4.1 Bcl-2 immunohistochemical staining of normal cervical tissue.

Expression of bcl-2 was observed in the basal layer of normal cervical squamous epithelium.



Figure 4.2 Bcl-2 immunohistochemical staining of cervical cancer tissue.

Expression of bcl-2 was observed in the cytoplasm of the tumor cells.

In normal cervical tissue, there was consistent expression of bcl-2 protein in the basal layer of cervical squamous epithelium (Figure 4.1). In cervical cancer tissue, the immunoreactivity for positive bcl-2 was observed in the cytoplasm of the tumor cells (Figure 4.2). In almost all tumors, bcl-2 expression was heterogeneous and the expressions sometimes vary from strongly positive to negative within the same tumor.

4.3 Association between bcl-2 expression and tumor recurrence

Positive bcl-2 expression (> 5%) was found in 43.7% (95% CI: 26.5%-60.9%) of cases compared to 50.8% (95% CI: 38.5%-63.1%) of controls, which were not statistically significant difference (Table 4.2). Even we reset the cut off point for positive bcl-2 from 5% (+1) to 10% (+2) or 30%(+3), the positive bcl-2 were not significantly different between cases and controls (Table 4.3), which means that bcl-2 expression was not significantly associated with the recurrence of cervical cancer.

Concerning other prognostic factors for cervical cancer (Table 4.2), tumor grade and presence of lymph vascular space invasion were significantly associated with tumor recurrence. Patients with grade 2-3 had 6.9 times risk of recurrence compared to those with grade 1. Those with lymph-vascular space invasion had 5.4 times risk of recurrence compared to those without lymph-vascular space invasion. Patients with depth of invasion more than half of cervical thickness also had a trend to have higher risk of recurrence but did not reach statistical significance. In multivariable analysis, tumor grade and presence of lymph vascular space invasion were also significantly associated with tumor recurrence (Table 4.2).

Table 4.2 Bcl-2 expression and pathological findings of cases and controls.

	Cases (n=32)		Controls (n=63)		Crude			Adjusted ^a		
	No.	%	No.	%	OR	95% CI	p value	OR	95% CI	p value
Bcl-2										
Negative	18	56.3	31	49.2	1 ^b					
Positive	14	43.7	32	50.8	0.7	0.3 - 1.6	0.378	0.6	0.2 - 1.7	0.304
+1	5	15.6	14	22.2						
+2	6	18.7	8	12.7						
+3	3	9.4	10	15.9						
Tumor grade										
1	7	21.9	30	47.6	1 ^b					
2-3	25	78.1	33	52.4	8.1	1.8 - 36.9	0.007	6.9	1.4 - 35.0	0.019
Lymph-vascular space invasion (LVSI)										
Absence	9	28.1	35	55.6	1 ^b					
Presence	23	71.9	28	44.4	3.8	1.3 - 10.6	0.013	5.4	1.4 - 20.1	0.012
Depth of invasion										
≤1/2	8	25.0	26	41.3	1 ^b					
>1/2	24	75.0	37	58.7	2.1	0.8 - 5.4	0.129	1.7	0.6 - 5.1	0.320

^a adjusted for the remaining variables in the table.

^b reference group.

Table 4.3 Association of bcl-2 expression and tumor recurrence with different cut off point of bcl-2 expression.

	Crude			Adjusted ^a		
	OR	95%CI	p value	OR	95%CI	p value
Bcl-2						
Positive (+1,+2,+3 or >5%)	0.7	0.3-1.6	0.378	0.6	0.2-1.7	0.304
Positive (+2 ,+3 or >10%)	0.9	0.4-2.4	0.874	0.8	0.3-2.8	0.768
Positive (+3 or >30%)	0.5	0.1-2.1	0.352	0.2	0.02-2.5	0.230

^a adjusted for tumor grade, depth of invasion and lymph-vascular space invasion.

reference group

: positive (+1,+2,+3 or >5%) the reference group is negative (≤5%)

: positive (+2,+3 or >10%) the reference group is negative and 1+ (or ≤10%)

: positive (+3 or >30%) the reference group is negative, 1+ and 2+ (or ≤30%)

4.4 Association between bcl-2 expression and other prognostic factors for recurrence

Table 4.4 Association between bcl-2 expression and other prognostic factors.

	No. (n=95)	Bcl-2 positive Number (%)	Crude			Adjusted ^a		
			OR	95%CI	p value	OR	95%CI	p value
Tumor grade								
1	37	23(62.2)	1 ^b					
2-3	58	23(39.7)	0.4	0.2-0.9	0.034	0.4	0.1-0.9	0.023
Lymph-vascular space invasion (LVSI)								
Absence	44	17(38.6)	1 ^b					
Presence	51	29(56.9)	2.1	0.9-4.8	0.078	2.2	0.9-5.1	0.078
Depth of invasion								
≤1/2	34	13(38.2)	1 ^b					
>1/2	61	33(54.1)	1.9	0.8-4.5	0.140	1.9	0.8-4.7	0.151

^a adjusted for the remaining variables in the table.

^b reference group.

To assess the association between bcl-2 expression and other prognostic factors, bcl-2 expression was considered as a dependent variable (Table 4.4). From univariable and multivariable analysis, tumor grade was significantly associated with bcl-2 expression, while lymph-vascular space invasion had borderline significant association. Positive bcl-2 expression was found in 62.2% of grade 1 tumor compared to 39.7% in grade 2-3. Regarding lymph-vascular space invasion, positive bcl-2 expression was found in 56.9% of patients with lymph-vascular space invasion compared to 38.6% in those without lymph-vascular space invasion. Positive bcl-2 also tended to be found more often in patients with deep invasion (>1/2), but not reach statistical significance.

4.5 Association between bcl-2 expression and time to recurrence, site of recurrence

Table 4.5 Association between bcl-2 expression and time to recurrence, site of recurrence.

	Bcl-2 expression			p value
	Negative (n=18)	Positive (n=14)	Total (n=32)	
Time to recurrence (months)				0.111 ^a
Median	11.5	20.3	14.7	
Range	1.3-60.0	4.7-59.0	1.3-60.0	
Site of recurrence [No (%)]				
Without distant recurrence	16 (88.9%)	11 (78.6%)	27 (84.4%)	0.631 ^b
With distant recurrence	2 (11.1%)	3 (21.4%)	5 (15.6%)	

^a Mann - Whitney U test.

^b Fisher's exact test.

Among 32 cases with recurrence, association between bcl-2 expression (negative, positive) and time to recurrence, site of recurrence were evaluated (Table 4.5). Patients with positive bcl-2 tended to have longer median time to recurrence but did not reach statistical significance. Regarding site of metastasis, patients with positive and negative bcl-2 did not have statistically significant difference of distant recurrence.