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Expression of COX-2 and Bcl-2 in primary fallopian tube carcinoma: correlations with clinicopathologic features

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Abstract: The aim of this study was to evaluate the expression of COX-2 and Bcl-2 in primary fallopian tube carcinoma (PFTC), as well as their correlations with clinicopathologic features. We studied a cohort of 33 patients with a pathological diagnosis of PFTC. Thirty normal tubal tissues used for controls were obtained from patients diagnosed with uterine myomas. Expression analysis for COX-2 and Bcl-2 was performed using the immunohistochemical technique. The rate of preoperative diagnosis was 18.2%. With a median survival of 61.0 months (95% CI: 43.2 to 78.8 months), the estimated five-year overall survival rate in the 33 patients was 39.0%. Increased expression of COX-2 and Bcl-2 was observed in tumor specimens compared to normal controls ($p = 0.026$; $p = 0.003$). The expression rate of COX-2 in node-positive tumors was significantly higher than that of node-negative tumors ($p = 0.024$). Moreover, the expression rate of COX-2 was statistically significantly higher in patients with infiltration through the serosa ($p = 0.019$). Positive significant associations were observed between Bcl-2 staining index and FIGO stage ($p = 0.015$), and between Bcl-2 staining and lymph node metastasis ($p = 0.010$). There was a significant correlation between COX-2 expression and Bcl-2 staining index ($r = 0.517$, $p = 0.002$). We conclude that COX-2 and Bcl-2 may potentially be useful prognostic markers for PFTC. The exact molecular mechanism for correlations between COX-2 and Bcl-2 remains to be elucidated. *(Folia Histochemica et Cytobiologica 2011, Vol. 49, No. 3, 389–397)*

Key words: primary fallopian tube carcinoma, COX-2, Bcl-2, clinicopathologic features

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Introduction

Primary fallopian tube carcinoma (PFTC) is one of the rarest malignant tumors of the female genital tract, although the incidence of PFTC is gradually increas-

ing [1]. Tube malignancy mainly affects postmenopausal women in the fifth and sixth decades of life, with symptoms such as abdominal pain, vaginal bleeding, abnormal watery discharge and pelvic mass [2]. In addition to the surgical principle of optimal cytoreduction used for patients with ovarian carcinoma, the adoption of chemotherapeutic agents has also been advocated for patients with PFTC. A combination of paclitaxel and platinum-based therapy has been recommended as the standard chemotherapy regimen for PFTC patients [3]. PFTC has rarely been studied, and only a few reports with fairly small numbers have investigated the molecular events associated with this cancer [4, 5]. Analyses of potential molecular mediators would be helpful to guide treatment and to elucidate the mechanisms of disease pathogenesis.

The cyclooxygenase (COX) genes encode for the rate-limiting enzyme involved in the conversion of arachidonic acid to H2-prostaglandin [6]. Two COX genes, COX-1 and COX-2, have been identified. COX-1 is constitutively expressed. COX-2 gene expression has been demonstrated to increase in response to cytokines, mitogens, and growth factors [7]. High expression of COX-2 in a wide variety of solid epithelial tumors also favors the growth of malignant cells by inhibition of apoptosis and the promotion of angiogenesis [8]. Some studies have found a high correlation between the expression of this enzyme and cancer growth or prognosis [9, 10].

The Bcl-2 family is implicated in the regulation of apoptosis. The Bcl-2 family, with its antiapoptotic subgroups (Bcl-2, Bcl-xl, Mcl-1, and A1) and its death-promoting subgroups (Bax, Bcl-xs, Bak, Bad, and Bik) plays a central role in the regulation of cell death [11]. The Bcl-2 gene encodes a 26-kDa protein located in the mitochondrial inner membrane and cell cytosol. Bcl-2 protein acts as an inhibitor of apoptosis; it contributes to cancer pathogenesis and may be involved in resistance to cancer treatment [12]. The expression of Bcl-2 in a variety of tumor types has prompted intense research directed toward the role of this molecule in the pathogenesis of tumors.

So far as we know, no study has investigated the expression of COX-2 and Bcl-2 protein in PFTC. Thus, the aim of the current study was to investigate the expression of COX-2 and Bcl-2 and to correlate their expressions with the clinicopathologic features in PFTC. The relationship between Cox-2 and Bcl-2 protein expression was also evaluated.

Material and methods

Clinical data and tumor specimen acquisition. A total of 33 cases were diagnosed with PFTC. All were treated in one of the following hospitals: the First Affiliated Hospital of Anhui Medical University, Anhui Provincial Hospital, and the First People's Hospital of Hefei, Anhui between January 1995 and October 2009. They were all submitted to surgery as the primary treatment. None of these patients received preoperative chemotherapy, radiotherapy or immunotherapy. Clinical information and paraffin blocks were available for all the patients. Data on clinical parameters, including age, signs, symptoms, preoperative examination, preoperative diagnosis, treatment and follow-up information were gathered retrospectively from patient records. The variables studied in the pathology review were: tumor size, location, histological type, grading, depth of tubal wall infiltration, lymph node status and vascular space invasion. The histological specimens were all sent to the First Affiliated Hospital of Anhui Medical University, where the specimens were re-assessed by two independent pathologists. All cases were staged retrospectively according to the modified International Federation of Gynecology and Obstetrics (FIGO) staging system. The diagnostic criteria for PFTC proposed by Hu et al. and modified by Sedlis was applied [13]. Optimal cytoreduction was defined as residual tumor no greater than 1 cm in diameter at the conclusion of surgery. The 1 cm cut-off was used as the threshold in order to homogeneously assess patients diagnosed during different periods of time in this long retrospective study [3].

For patients with measurable disease after primary cytoreductive surgery, overall tumor response was defined using the modified Response Evaluation Criteria In Solid Tumors (RECIST), which also requires that complete response (CR) includes normalization of CA-125 levels. Non-measurable disease included cystic lesions and ascites. Pelvic and abdominal CT scan and/or MRI and chest X-ray and/or chest CT scan were repeated after the third and the sixth treatment courses.

Thirty normal tubal tissues used for controls were obtained from patients diagnosed with uterine myomas. They had hysterectomy and bilateral salpingo-oophorectomy performed in the First Affiliated Hospital of Anhui Medical University between January 2004 and January 2007. The specimens were also re-assessed by two independent pathologists, excluded for tumor or inflammation of the fallopian tubes.

Histology. The formalin-fixed, paraffin-embedded samples were sectioned at 4 μm and stained with hematoxylin and eosin. The histological diagnosis was re-examined by two independent pathologists. Moreover, the most representative blocks were selected to be cut into new 4 μm-thick sections for immunohistochemical study.

Immunohistochemical staining. Immunohistochemical staining was performed by a peroxidase-labelled streptavidin-biotin method. Sections were dewaxed and antigen retrieval was achieved by steaming the slides for 20 minutes in retrieval solution. Endogenous peroxidase activity was

blocked by incubation in 3% hydrogen peroxide solution. The antibodies used were a polyclonal rabbit antibody against COX-2 protein (clone sc-7951; Santa Cruz Biotechnology, Santa Cruz, CA, USA) in a 1:25 dilution at room temperature for one hour, and a monoclonal mouse antibody against Bcl-2 protein (clone sc-7382; Santa Cruz Biotechnology, Santa Cruz, CA, USA) in a 1:100 dilution at room temperature for one hour. After washing three times with phosphate buffered saline (PBS), sections were incubated with biotinylated goat anti-rabbit/anti-mouse immunoglobulin (Zymed Laboratories, USA) for ten minutes. They were then washed three times with PBS, treated with streptavidin–peroxidase reagent (Zymed Laboratories, San Francisco, CA, USA) for ten minutes, and then washed again with PBS three times. Finally, specimens were incubated in diaminobenzidine (DAB) for five minutes, followed by being counterstained with hematoxylin for one minute, rinsed twice in distilled water, and dehydrated with ethanol followed by xylene. Tissue from a colon cancer known to overexpress COX-2, and tissue from a tonsil specimen with a known expression of Bcl-2, served as positive controls. The primary antibody was replaced with PBS as a negative control.

Immunohistochemical evaluation. Semiquantitative evaluation of the immunohistochemical results was performed by two independent observers blinded to patient status. The IHC quantification for COX-2 and Bcl-2 followed previous published criteria based on intensity and percentage taken together. Firstly, the percentage of tumoral cells was scored as 0: < < 5%, 1: 5–25%, 2: 26–50%, 3: 51–75% or 4: > 75% of positive cells respectively. Secondly, the intensity was validated as 0: no reaction, 1: weak reactivity, 2: moderate or 3: intense. With these two molecules, a combined score was subsequently determined by multiplying the scores of intensity and percentage from different tumoral areas. The cytoplasmic positivity for COX-2 and Bcl-2 was graded as follows: negative (scored 0–1), +1 (scored 2–3), +2 (scored 4–6), +3 (scored > 6). The expression of proteins was classified into positive (Grade +1∼ ∼ +3) and negative (Grade 0) expressions [14, 15].

Statistical analysis. Overall survival (OS) curve was plotted, and median survival time was estimated using the Kaplan–Meier method [16]. OS was defined as the time from the day of operation to the time of death; data on survivors were censored at the last follow-up. Median overall survival was defined as the time-point where the Kaplan–Meier curve crossed the horizontal line at 50% survival probability. Distributions for immunostaining of molecules between PFTC and normal controls were compared and analyzed by the Chi-square test. Associations between immunohistochemical parameters and clinicopathologic parameters were determined using the Fisher's exact test. The Pearson Chi-square test was used to assess the relation between COX-2 and Bcl-2 expression; p values < 0.05 was regarded as statistically significant in two tailed tests, using SPSS for Windows 13.0 software.

Results

Patients' characteristics

A total of 33 patients with a pathological diagnosis of PFTC were collected in this retrospective analysis. The median age at diagnosis was 55 years (range 40–73). Clinicopathologic features of patients with PFTC are listed in Table 1.

One patient (3%) was nulliparous, eight patients (24%) had experienced one live birth, and 24 patients (73%) two or more live births. At the time of diagnosis, 22 patients (66.7%) were postmenopausal. The commonest signs and symptoms were: abnormal watery discharge (54.5%), abdominal pain (30.3%), abnormal vaginal bleeding (24.2%) and a palpable pelvic and/or abdominal mass (18.2%). The tumor was

Table 1. Patients' characteristics*

Variables	No. of patients (%)
Age, median, range	55 (40–73)
Menopausal	
Yes	22 (66.7)
No	11 (33.3)
FIGO stage	
I	6 (18.2)
II	10 (30.3)
III	15 (45.5)
IV	2 (6.0)
Histological type	
Serous adenocarcinoma	19 (57.6)
Papillary adenocarcinoma	4 (12.0)
Tubal adenocarcinoma	5 (15.2)
Adenocarcinoma NOS	5 (15.2)
Grade	
G1	5 (15.1)
G2	16 (48.5)
G3	12 (36.4)
Depth of tubal wall infiltration	
Intramucosal or submucosa/muscularis	24 (72.7)
Through serosa	9 (27.3)
Location	
Left	16 (48.5)
Right	15 (45.5)
Bilateral	2 (6.0)
Residual tumor	
≤ 1 cm	23 (69.7)
> 1 cm	10 (30.3)
Lymph node metastasis	
No	22 (66.7)
Yes	11 (33.3)
Chemotherapy agents	
TP (paclitaxel + cisplatin/carboplatin)	16 (48.5)
CAP (cyclophosphamide + epirubicin + + cisplatin/carboplatin)	17 (51.5)

*FIGO — International Federation of Gynecology and Obstetrics; NOS — not otherwise specified

an incidental finding at laparotomy in two patients (6.1%) who underwent surgery for nonmalignant indications. Despite this, a preoperative diagnosis of PFTC was made only in six patients (18.2%). Among these patients, it was demonstrated that transvaginal ultrasound examination with color Doppler can detect areas of neovascularization within the fallopian tube, and thus aid in the preoperative diagnosis of PFTC. The majority of patients were of serous histology, and in 12 patients (36.4%) the tumors were poorly differentiated. CA-125 was measured preoperatively in all patients and was found to be elevated (> 35 U/ml) in 30 of these patients (range, 8.84–1,000 U/ml; median, 162.2 U/ml).

For patients with early-stage disease, the surgical procedure included a total abdominal hysterectomy, bilateral salpingo-oophorectomy, omentectomy, and routine pelvic and para-aortic lymph node sampling of lymph nodes that were visible or palpable after opening of the visceral peritoneum. For patients with advanced disease, aggressive cytoreductive surgery with removal of as much tumor as possible is warranted. Positive lymph nodes were found in 11 of 25 patients with palpable lymph nodes removed at primary surgery. Suboptimal debulking surgery with residual disease > 1 cm was recorded in ten (30.3%) cases. Adjuvant chemotherapy with platinum-containing regimen was offered to all the patients involved in our study. Sixteen (48.5%) patients received a combination of paclitaxel and platinum, whereas the other 17 patients (51.5%) were treated with a regimen of platinum-epirubicin-cyclophosphamide combinations. Objective response was assessed in ten (30.3%) patients with measurable disease. One of these patients had a complete clinical response (CR), three had partial response (PR), two had stable disease (SD) and four had progressive disease (PD), inducing an overall response rate (CR + PR) of 40.0%. Clinical benefit (CR + PR + SD) was observed in 60% of patients with measurable disease.

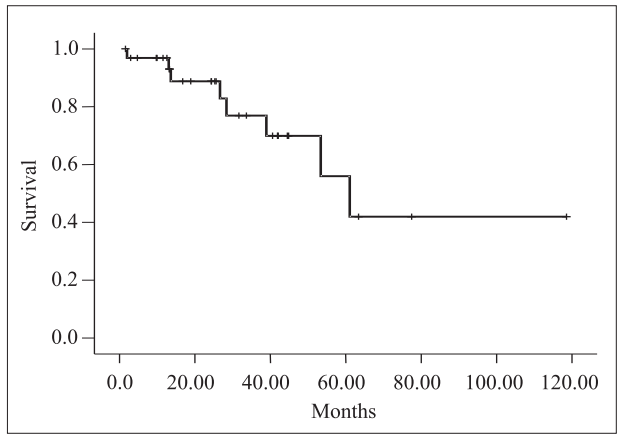

Figure 1. Overall survival curve for the entire population of the study

At the time of analysis, only one patient was lost to follow-up, 12 (36.4%) patients had relapsed and eight (24.2%) had died. With a median follow-up of 25 months (range: 4–119 months), the median overall survival was 61 months (95% CI: 43.2 to 78.8 months) for the entire population. The one, three, and five year overall survival rates were 88%, 68%, and 39%, respectively. Figure 1 demonstrates the overall survival curve by Kaplan–Meier method for the 33 patients with PFTC.

Immunohistochemical results

Expression of COX-2 and Bcl-2 in PFTC and normal fallopian tube tissues

Table 2 summarizes the results from the immunohistochemical analysis of 33 tumor specimens and 30 normal controls. The expression of COX-2 and Bcl-2 was detected as diffuse brown cytoplasmic or membranous reaction. The expression analysis of COX-2 revealed that in 14 of 33 (42.4%) tumor tissues, COX-2 staining was positive (Figure 2A) whereas in five of

Table 2. Expression of COX-2 and Bcl-2 in PFTC and normal fallopian tube*

Variables	COX-2 expression			Bcl-2 expression		
	Negative	Positive	Expression rate (%)	Negative	Positive	Expression rate (%)
PFTC (n = 33)	19	14	42.4	17	16	48.5
Normal tube (n = 30)	25	5	16.7	26	4	13.3
χ^2	4.95			8.96		
p	0.026			0.003		

*PFTC — primary fallopian tube carcinoma

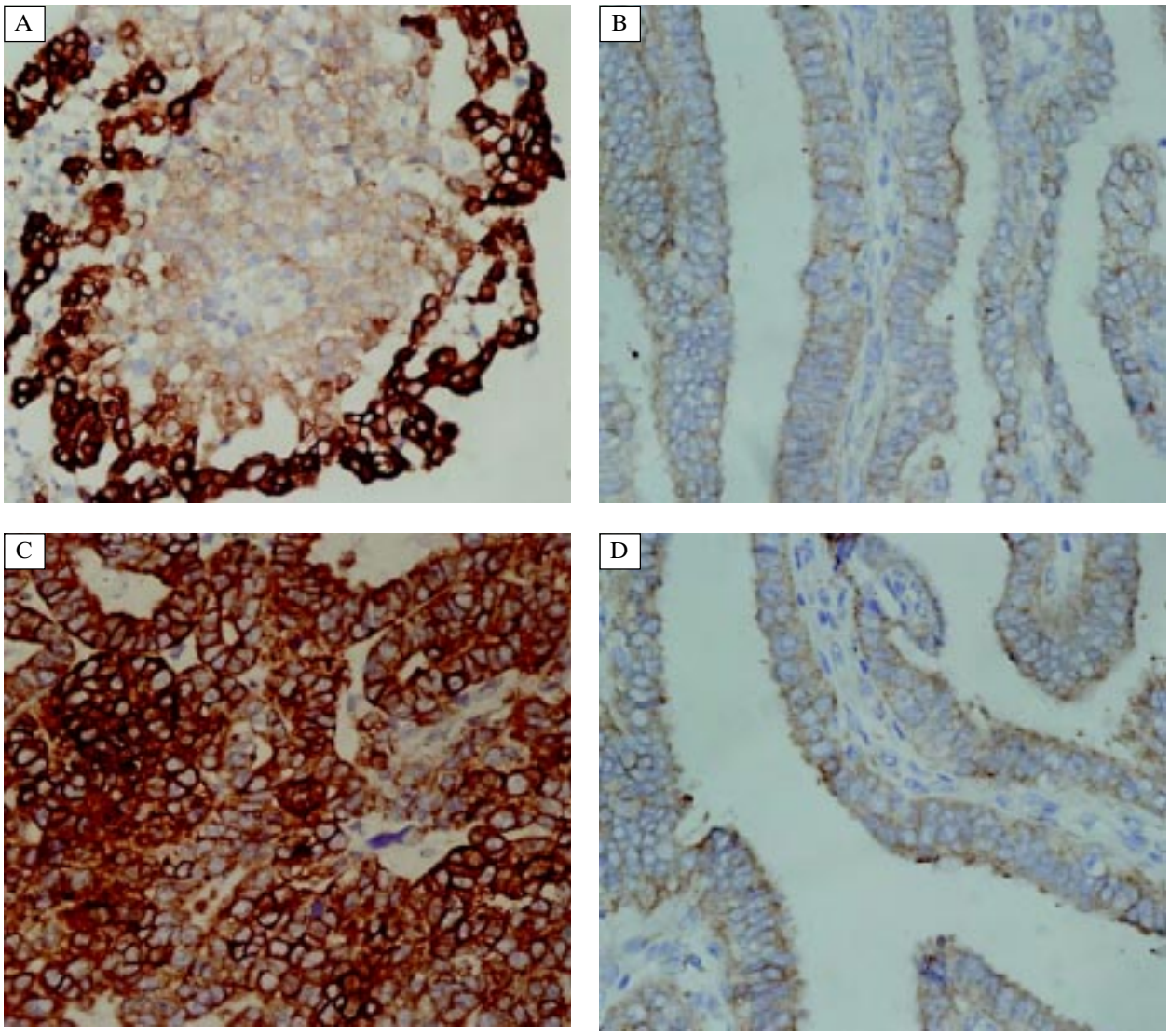

Figure 2. A. Diffuse and marked membranous and cytoplasmic positivity for COX-2 in primary fallopian tube carcinoma (Streptavidin-peroxidase × 400). **B.** Membranous positivity for COX-2 in normal fallopian tube tissue (Streptavidin--peroxidase × 400). **C.** Intense membranous positivity for Bcl-2 in primary fallopian tube carcinoma (Streptavidin--peroxidase × 400). **D.** Membranous positivity for Bcl-2 in normal fallopian tube tissue (Streptavidin-peroxidase × 400)

30 (16.7%) normal tissues, cytoplasmic expression was recorded (Figure 2B). There was a significant difference between the expression rate in tumor tissues and in normal tissues ($\chi^2 = 4.95$; $p = 0.026$). Positive expression of Bcl-2 was observed in 16 of 33 (48.5%) tumor tissues (Figure 2C) but in only four of 30 (13.3%) normal tissues (Figure 2D). The difference between Bcl-2 expression rate in tumor tissues and normal tissues was also statistically significant ($\chi^2 = 8.96$; $p = 0.003$).

COX-2 and Bcl-2 protein expression in relation to classification of clinicopathologic features

Clinicopathologic features including menopausal, FIGO stage, histological type, grade, depth of tubal wall infiltration and lymph node metastasis grouping of the cancers in relation to COX-2 and Bcl-2 expression are shown in Table 3. Expression of COX-2 was significantly correlated with depth of tubal wall infiltration in PFTC ($p = 0.019$). We also found the expression rate of COX-2 in tumors with lymph node metastasis was significantly higher than that of node--negative tumors (72.7% vs. 27.3%, $p = 0.024$). However, there were no significant associations between COX-2 expression and other parameters including menopausal, FIGO stage, histological type and grade. As far as Bcl-2 staining is concerned, a statistically significant higher expression rate of Bcl-2 was found in cases with lymph node metastasis than in cases without lymph node metastasis (81.8% vs. 31.8%,

Table 3. Relations between COX-2 expression and clinicopathologic features in PFTC*

Variables	n	COX-2 expression				Bcl-2 expression			
		Negative	Positive	Expression rate (%)	p	Negative	Positive	Expression rate (%)	p
Menopausal									
Yes	22	13	9	40.9	1.000	10	12	54.5	0.465
No	11	6	5	45.5		7	4	36.4	
FIGO stage									
I–II	16	11	5	31.3	0.296	12	4	25.0	0.015
III–IV	17	8	9	52.9		5	12	70.6	
Histological type									
Serous	19	9	10	52.6	0.286	8	11	57.9	0.296
Other types	14	10	4	28.6		9	5	35.7	
Grade									
G1, G2	21	13	8	38.1	0.716	12	9	42.9	0.200
G3	12	6	6	50.0		5	7	58.3	
Depth of tubal wall infiltration									
Intramucosal or submucosa/muscularis	24	17	7	29.2	0.019	14	10	41.7	0.259
Through serosa	9	2	7	77.8		3	6	66.7	
Lymph node metastasis									
No	22	16	6	27.3	0.024	15	7	31.8	0.010
Yes	11	3	8	72.7		2	9	81.8	

*FIGO — International Federation of Gynecology and Obstetrics; PFTC — primary fallopian tube carcinoma

Table 4. Correlation between COX-2 and Bcl-2 expression in primary fallopian tube carcinoma

COX-2	Bcl-2		r	p
	Negative	Positive		
Negative	14	5	0.517	0.002
Positive	3	11		

p = 0.010). In addition, the expression rate of Bcl-2 was higher in cases staged III and IV with respect to cases staged I and II (70.6% vs. 25.0%, p = 0.015). There was no significant difference in expression rates of Bcl-2 in patients among menopausal, histologic type, grade and depth of tubal wall infiltration.

Correlation between COX-2 expression and Bcl-2 expression

Table 4 summarizes the correlation between the immunohistochemical results of COX-2 and Bcl-2 in PFTC. Using the Pearson Chi-square test, a positive significant correlation was found between COX-2 expression and Bcl-2 expression (r = 0.517, p = 0.002).

Discussion

In the current study, we have performed a retrospective analysis of 33 PFTC patients in three different hospitals. Furthermore, we have investigated the expression of COX-2 and Bcl-2 and correlated their expressions with the clinicopathologic features in PFTC.

PFTC most frequently occurs in the fourth, fifth and sixth decades of life, with a median age of occurrence of 55 years (range, 17–88 years) [1]. High parity and a history of pregnancy have been reported to decrease the PFTC risk significantly [2]. Unlike these reports, our study showed that only one patient had a history of infertility. It may be that the protective and risk factors of PFTC vary by race. The rate of preoperative diagnosis is in the range of 0–10% in the literature [17]. However, preoperative diagnosis of PFTC was made in six patients (18.2%) in our cohort. Most of the patient characteristics described in this material were quite comparable to those reported in the literature. However, the five-year survival rate of 39% was somewhat lower in the present series than in most reports. In a large population-based registry study of 151 PFTC patients, the reported five-year survival rate was 44%, and the median survival time was 52 months

[13]. Gadducci et al. [18] reported that for the whole series the five-year survival rate was 57%. It is likely that the shorter follow-up time may partly account for the relatively lower survival rate in our study. Additionally, 51.6% of the patients were in an advanced stage of their disease. It is known that stage is an important prognostic factor for outcome [1, 13].

COX-2 is normally undetectable in most tissues and can be induced by a number of stimuli, including cytokines such as IL-1, interferon-γ and tumor necrosis factor [19]. In the current study, we showed for the first time that increased expression of COX-2 *in vivo* was accompanied by increased expression of Bcl-2 protein in PFTC compared to normal fallopian tube tissues. This result suggests that COX-2 and Bcl-2 may both play important roles in promoting tumorigenesis. Moreover, we found significant correlations between COX-2 expression and depth of tubal wall infiltration and lymph node metastasis in our study, a finding that was compatible with previous reports on other organs [12, 20]. Therefore, COX-2 might have played a role in activating the metastatic potential of tumor cells in our patients.

Chen et al. [21] reported a significantly higher expression of COX-2 in endometrial carcinoma than in normal endometrium tissue and COX-2 expression was related to the histological grade. A study from Thailand provided evidence that COX-2 was overexpressed in the colorectal tumor tissues and their presence was significantly correlated with poor differentiation [22]. However, in the present study, we were unable to identify any correlation between COX-2 staining and histological grade. To the best of our knowledge, there is no generally accepted grading system for PFTC, mainly because the same criteria are not applicable to all histologic types. Consistent with our notion, Ohno et al. [20] reported that COX-2 mRNA expression in gastric carcinoma tissue is correlated closely with depth of invasion. Another similar finding suggested that COX-2 was a good predictor for lymph node metastasis in mucoepidermoid carcinoma [12]. Compelling evidence suggests that COX-2 might enhance the metastatic potential as well as tumorigenicity and might be involved in the progression of some tumors.

COX-2 contributes to tumorigenesis through several mechanisms. These include an increase in proliferation, reduction in apoptosis, promotion of angiogenesis, modulation in inflammation and immune function, decrease in E-cadherin expression, and stimulation in the invasive/metastatic potential [23]. Studies have demonstrated the effects of non-steroidal anti-inflammatory drugs (NSAIDs) in the prevention of human cancers. Recent progress in the treatment and prevention of cancers of the lung, breast, esophagus, colon, bladder and prostate with NSAIDs, especially COX-2 inhibitors, has increased our understanding of COX-2 inhibition in both cancer treatment and prevention [24, 25]. Previous studies from our laboratory have suggested the effects of paeonol in the prevention of human cancers *in vitro* and *in vivo* by inducing apoptosis. Thus, the inhibition of COX-2 enzymatic activity in controlling neoplastic cell proliferation is the possible mechanism [26–28]. Therefore, these studies suggest the potential use of COX-2 inhibitors combined with other chemotherapy drugs or irradiation in the treatment of PFTC.

This study, the first to assess the clinical usefulness of Bcl-2 expression in PFTC, showed that Bcl-2 expression rate differed according to FIGO stage and was significantly higher in node-positive tumors compared to node-negative tumors. These findings indicate that Bcl-2 may be a critical factor affecting the tumorigenesis and progression of PFTC, as well as an indicator of biologic behavior. The role of Bcl-2 in the current results parallels those of other studies. Shimizu et al. [10] found that Bcl-2 gene expression increased during progression from normal tissue to Barrett's associated adenocarcinoma. Another study revealed that the expression of Bcl-2 was significantly higher in malignant than in benign tumors, and was higher in serous than in mucinous borderline ovarian tumors [29]. Bcl-2 has functions in tissue differentiation and development, apart from prolonged cell lifespan by blocking apoptosis. Reducing the capacity of apoptotic cell turnover could be an important step in the development of neoplasia [30].

Our findings indicated that COX-2 overexpression in tumor tissues was associated with Bcl-2 overexpression. To date, COX-2 has been determined to contribute to tumorigenesis and the malignant phenotype of tumor cells via the inhibition of apoptosis, increased angiogenesis and invasiveness. Growing evidence indicates that induction of COX-2 correlates with increased Bcl-2 expression. Tjiu et al. [31] found that Mcl-1 and Bcl-2, both members of the Bcl-2 family, were significantly upregulated in COX-2-overexpressing human basal cell carcinoma cells, whereas other Bcl-2 members were not. This implicated Mcl-1 and Bcl-2 in COX-2-mediated antiapoptotic effects. Sakamoto et al. [32] reported that COX-2 regulated the degree of apoptosis by modulating Bcl-2 protein in pleomorphic adenoma and mucoepidermoid carcinoma of the parotid gland. Exposure of a variety of cancer cells to a selective COX-2 inhibitor induces apoptosis [33]. A previous study has indicated that COX-2-derived eicosanoid products can induce Bcl-2 expression and inhibit programmed cell death [34]. The exact molecular mechanisms remain to be elucidated by a further genome-wide analysis.

Conclusions

Our results suggest that COX-2 and Bcl-2 may play important synergistic roles in the oncogenesis, development and metastasis of PFTC. We conclude that COX-2 and Bcl-2 may potentially be useful prognostic markers for PFTC. Future analysis of the role of these markers in the prognosis of PFTC is warranted, and should be based on larger cohorts of the tumors. The inhibition of COX-2 activity may have an important therapeutic benefit in the management of PFTC.

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Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this paper.

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