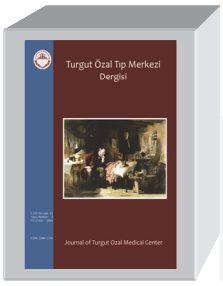

ORİJİNAL MAKALE/ORIGINAL ARTICLE
J Turgut Ozal Med Cent 2014:21(3):207-14

Journal Of Turgut Ozal Medical Center
www.jtomc.org

Relationship of COX-2, BAX, BCL-2, Ki67, p53 Expression to Clinicopathologic Parameters and Their Impact on Prognosis in Renal Cell Carcinoma

Fatma Seher Pehlivan[1], Ayşegül Sarı[2], Sacit Nuri Görgel[3], Yelda Morgül[2], Uğur Balcı[3], Murat Ermete[2], Ertuğrul Şefik[3]

[1]Private Tınaztepe Hospital, Pathology Clinic, İzmir, Turkey
[2]Katip Celebi University, Training and Research Hospital, Pathology Clinic, İzmir, Turkey
[3]Katip Celebi University, Training and Research Hospital, 1st Urology Clinic, İzmir, Turkey

Abstract

Objective: Renal cell carcinoma (RCC) is a malignancy originating from the renal cortex and is accounted for 85% of all malignant kidney tumors. Currently, tumor stage and nuclear grade are recognized as the most important determinants of prognosis in patients with RCC. Nonetheless, staging and grading do not suffice for predicting the prognosis in many patients. However, determining such patients bears importance in terms of oncology. Therefore, additional parameters are required to define the tumor behavior. In this study, we aimed to investigate the relation of Cox-2, Bax, Bcl-2, and p53 expression, as well as Ki-67 proliferation index to histopathologic parameters, and their impact on prognosis.

Material and Methods: Our study included 70 RCC patients who were referred to the Pathology Department of Izmir Ataturk Teaching and Research Hospital from the First Urology Clinic between May 1998 and May 2008. The data including tumor diameter, histologic tumor type, nuclear grade, tumor stage, metastatic status, life expectancy of patients as well as staining percentages of Bax, Cox-2, Bcl-2, p53, and Ki-67 were compared.

Results: In this study, no relationship was found between those markers and clinicopathologic parameters including the prognosis.

Conclusion: Tumor pathologic stage showed a statistically significant predictive value for metastasis development, however, it failed to predict survival. There is a need for studies including standardized methods, larger samples, and longer follow-up periods in order to reveal effects of Cox-2, Bax, Bcl-2, P53 markers, and Ki-67 proliferation index on prognosis.

Key Words: Renal Cell Carcinoma; Cox-2; Bax; Bcl-2; ki67; p53.

Renal Hücreli Karsinomlarda Cox-2, Bax, Bcl-2, Ki-67 ve p53 Ekspresyonunun Klinikopatolojik Parametrelerle İlişkisi ve Prognoza Etkisi

Özet

Giriş ve Amaç: Renal Hücreli Karsinom (RHK), böbrek korteksinden kaynaklanan bir malignite olup malign böbrek tümörlerinin %85' ini oluşturmaktadır. Günümüzde RHK' lu hastalarda prognozun en önemli belirleyicileri olarak evre ve nükleer derece kabul edilmektedir. Bununla birlikte pek çok hastada evreleme ve dereceleme hastalığın seyrinin belirlenmesinde yetersiz kalmaktadır. Prognozu ön görülemeyen bu hastaların belirlenmesi onkolojik açıdan önemlidir. Bu nedenle tümörün davranışının belirlenmesi amacıyla ek parametrelere ihtiyaç vardır. Bu çalışmada RHK' da Cox-2, Bax, Bcl-2, p53 ekspresyonu ve Ki-67 proliferasyon indeksinin histopatolojik parametreler ile ilişkisini ve prognoza etkisini araştırmayı amaçladık.

Gereç ve Yöntem: Çalışmaya İzmir Atatürk Eğitim ve Araştırma Hastanesi Patoloji bölümüne Mayıs 1998 - Mayıs 2008 tarihleri arasında 1. Üroloji kliniğinden gönderilen 70 adet RHK olgusu dahil edildi. Çalışmada, tümör çapı, tümörün histolojik tipi, nükleer derecesi, evresi, metastaz durumu ve hasta yaşam süresi ile immunohistokimyasal olarak uygulanan Bax, Cox-2, Bcl-2, p53 ve Ki 67 ekspresyonunun boyanma yüzdeleri istatiksel olarak karşılaştırıldı.

Bulgular: Çalışmamızda bu belirteçler ile prognoz dahil hiçbir klinikopatolojik parametre arasında anlamlı ilişki bulunmadı.

Sonuç: Tümörün patolojik evresi sadece metastaz gelişimini öngörmede istatiksel anlamalılık göstermiş olup sağ kalımı belirlemede etkisiz kalmıştır. RHK' da Cox-2 ekspresyonu, Bax, Bcl-2 ve p53 gibi apoptik belirteçler ve Ki67 proliferasyon indeksinin prognoz üzerine etkisinin olup olmadığını net olarak belirleyebilmek için standardize edilmiş yöntemlerle, daha fazla sayıda olgu ve takip süresi içeren çalışmalara ihtiyaç olduğunu düşünmekteyiz.

Anahtar Kelimeler: Renal Hücreli Karsinom; Cox-2; Bax; Bcl-2; ki67; p53.

INTRODUCTION

RCC is the most common cancer arising from the renal tubule. It constitutes 3% of all adult malignancies. In renal tumours, however, approximately 85% of the cases are RCC cases (1). The incidence of RCC is rapidly increasing. The reason for this is the frequency of routine examinations in recent years with advancing imaging methods (2). Today, pathological stage and nuclear grade are known as the two most important prognostic factors in kidney tumors (4). In spite of the fact that researches on other potential clinical, anatomical, histological, and bimolecular parameters still continue to

determine prognostic factors, the ideal prognostic markers of the disease has not yet been identified.

Cyclooxygenases are rate-limiting enzymes in prostaglandin biosynthesis. They have got two isoforms: Cox-1 and Cox-2. While Cox-1 is released from many of the cells that enable homeostasis, Cox-2, being pro-inflammatory, can be expressed by the effects of a variety of cytokines and growth factors (4). COX-2 expression has been found to have increased in gastrointestinal tract, breast, cervix, lung, prostate and bladder cancers (5-11) but, as far as Cox-2 expression is concerned, there are different data in RCC (4, 12-17).

The best known and most frequently mutated genes among apoptosis regulators, p53, Bcl-2, and Bax and the effects of these markers on prognosis are still a matter of discussion as it has been shown in the studies conducted on RCC (4, 18, 19). Ki-67 expression, evaluated to assess cell proliferation activity in a number of tumour tissues, has been recognized as an excellent marker for tumor proliferation. RCC studies with the proliferation marker Ki-67 have disclosed a relationship between histological grade and tumor stage and some researchers showed that Ki-67 may act as an independent predictor of survival (20-22).

The aim of our study is to assess the relationship between histologic and clinicopathologic parameters in RCC and apoptosis and proliferation markers such as Cox-2 and Bax, Bcl-2, p53, and Ki-67 and to determine their effects on prognosis.

MATERIAL AND METHODS

70 nephrectomy patients diagnosed with RCC at Izmir Ataturk Training and Research Hospital, Department of Pathology, are included in the study. All these patients were operated at the 1st Urinary clinic between May 1998 and May 2008. Determined by 10% formaldehyde solution, embedded in paraffin blocks, and hematoxylin-eosin (HE) stained preparations of these patients that had been archived were prepared. The data about tumor histological type, nuclear grade, pathological stage, and perirenal fat invasion, were revised and recorded accompanied by renal vein invasion pathologies. The information about the type of operation, recurrence, metastasis, follow-ups, and final state of these patients were obtained from the patients' files at the urology clinic. While grading the tumours, we made use of the Fuhrman's nuclear grading system. For staging, on the other hand, we used the 2002 TNM staging (23). Because sarcomatoid change is a marker of progression in RCC, it is regarded as a specific type in the statistical evaluation (17). Parallel to this, because it has got easier prognosis compared to that of clear cell RCC and it represents a separate subgroup, cystic RCC is evaluated as a specific type (24).

In all cases, we transferred 4-5 micron-thick sections from the paraffin blocks that represented the tumour to poly-L-lysine slides. All sections were applied the necessary immunohistological processes with the antibodies of the following substances: Cox-2 (Mouse monoclonal antibody, Novacastra), Bax (epitope A specific rabbit polyclonal antibody, Lab Vision), Bcl-2 (Liquid Mouse monoclonal antibody, Novacastra), Ki-67 (Rabbit monoclonal antibody, Thermo Scientific), p53 (Ab-5, Thermo Scientific).

Immunohistochemical Evaluation:

All immunohistochemically stained sections were examined under a light microscope. For Cox-2, Bax, and Bcl 2, cytoplasmic and membranous staining was considered positive. The prevalence of staining was scored by proportionalizing the total area with carcinoma with positively stained area: (%0)=0, (1-25%)=1, (26-50%)=2, (51-75%)=3, (76-100%)=4. The intensity of staining was classified as follows: 0 (negative), 1 (weak), 2 (moderate), and 3 (strong). During the statistical evaluation, we first evaluated staining intensities and percentages separately, and then re-evaluated them after determining the final staining score. The final staining score (0-7) was obtained by combining staining intensity and prevalence. Scores greater than 2 were considered positive (17). This method was applied to Cox-2, Bcl-2, and Bax separately.

Nuclear staining was considered positive for Ki-67 and p53. In each case and for each antibody, we scanned and studied the areas with the most intense positive staining and the thinnest areas of the section at x100 magnification of the light microscope. With x400 magnification, however, we calculated the percentage of cells with positive staining in about 1000 tumor cells of the selected areas. In our statistical analysis, 10% and over was considered positive for p53 (5). Calculating the median value for Ki-67 proliferation index, we considered the values below the median (≤ 1%) as low index and the values above the median (> 1%) as high index.

Statistical analyses were performed using SPSS (Statistical Package for Social Sciences) programme (version 13.0, SPSS, Inc, Chicago, IL). Continuous data are given as median values (min-max). In comparing the continuous data within two groups, we used the Mann Whitney U test, while multiple group comparisons (more than two) were carried out by using the Kruskal-Walls test. Differences between categorical variables were analysed by chi-square test. We conducted Kaplan-Meier method for the determination of survival analysis. The comparisons of survival time, however, were performed by using the Log-Rank test. Two-way p-value ≤ 0.05 was considered to be statistically significant.

RESULTS

64.3% (n=45) of the patients included in our study were males while 35.7% (n = 25) were females. Patients' ages ranged between 20 and 83; the median age was 57. 50% (n=35) of the tumours were located in the right kidney and the remaining 50% (n=35) were in the left kidney. 91.4% of the materials were case of radical nephrectomy (n=64); 8.6% were partial nephrectomy materials (n=6). Other parameters of the tumour are given in Table 1.

Table 1. Clinicopathological findings of the 70 patients with RCC.

		n	%
Tumour type			
	Clear Cell	45	62,9
	Papillary	9	12,9
	Chromophobe	11	15,7
	Sacromatoid	3	4,3
	Cystic	2	2,9
Tumour diameter (cm)			
	<4	10	14,3
	4-7	22	31,4
	>7	38	54,3
Tumour grade (Furhman)			
	G1	7	34,2
	G2	34	28,5
	G3	17	34,2
	G4	12	3,1
Tumour stage (pT)			
	pT1	24	34,2
	pT2	20	28,5
	pT3	24	34,2
	pT4	2	3,1
Reccurence			
	Present	8	12,9
	Absent	54	87,1
Metastasis			
	Present	7	11,3
	Absent	55	88,7
Present state			
	Alive	50	80,6
	Dead	12	19,4

During the follow-up, the control computed tomography showed recurrence in 11.4% of the patients who underwent operation (n=8). In our research, we were able to reach 64 of the 70 patients. Of these patients, 78.1% have survived (n=50) and 21.9% were dead (n=14). Of these 14 patients who passed away, 2 had died immediately after the operation; these two patients were not included in the statistics in assessing the prognosis. Of the 64 patients whose files were accessible, the maximum follow-up period was 96 months, the minimum follow-up period was 2 months, and the median follow-up time was 17 months. The mean survival time was 70.75 months (95% confidence interval [CI] = 57.85 to 83.66).

Immunohistochemical Findings

With Cox-2, Bax, and Bcl-2, we observed cytoplasmic, membranous, and granular staining patterns in tumor cells (Figure 1-2). In all cases, non-tumoral renal tissues, and distal and proximal tubules displayed staining. No staining was observed in the glomeruli. We also observed Cox-2 immunoreactivity in histiocytes and mononuclear inflammatory cells. The expression ratios of COX-2, Bax, and Bcl-2 in the tumor tissue are indicated in Table 2.

We did not find any correlation between Cox-2 staining intensity and percentages, and any of the prognostic parameters. Analysing final staining score of the Cox-2 expression (0-7), (>2 positive, <2 negative) and all the parameters, we could obtain a statistically significant result only in relation to nuclear grading (p=0.034) (Table 3).

The percentage, intensity, and the end score of Bax and Bcl-2 staining expression did not prove to have any statistically significant relationship with any of the parameters (p>0.05). Of the 70 patients, 94.3% showed p53 expression less than 10% (n=60) while the remaining 5.7% had p53 expression more than 10% (n=4). Again, this made us think that there was no significant correlation between p53 expression and any of the clinical and pathological parameters.

Table 2. COX-2, Bax , and Bcl-2 staining percentages, intensity, and scores.

		Cox-2 (n/%)	Bax (n/%)	Bcl-2(n/%)
%				
	None	28/ 40,0	18/25,7	29/41,4
	%1-25	20/28,6	25/35,7	23/32,9
	%26-50	12/17,1	7/10,0	2/2,9
		4/5,7	10/14,3	7/10,0
	%76-100	6/8,6	10/14,3	9/12,9
Intensity				
	None	28/40,0	18/25,7	29/41,4
	Weak	21/30,0	32/45,7	27/38,6
	Moderate	18/25,7	14/20	9/12,9
	Strong	3/4,3	6/8,6	5/7,1
Score				
	Score ≤2 (negative)	44/62,9	38/54,3	49/70
	Score >2 (positive)	26/37,1	32/45,7	21/30

Table 3. COX-2 and nuclear grading of the tumours.

		grade 1-2 n/%	grade 3-4 n/%	p
Cox-2 (n/%)				**0,034***
	negative	30/68,2	14/31,8	
	positive	11/42,3	15/57,7	

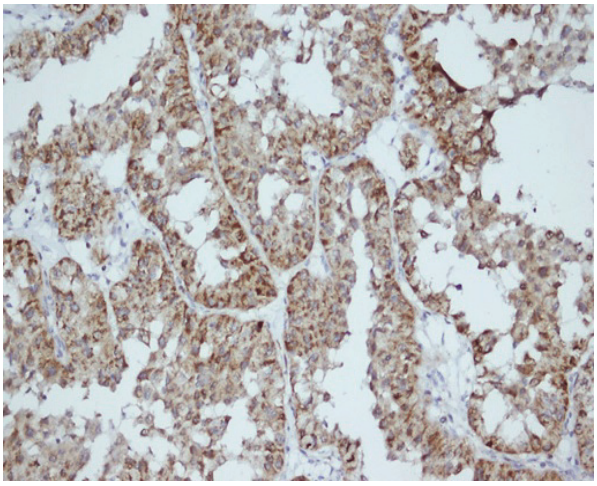

Figure 1a. Cycstoplasmic Cox-2 expression in conventional RCC.

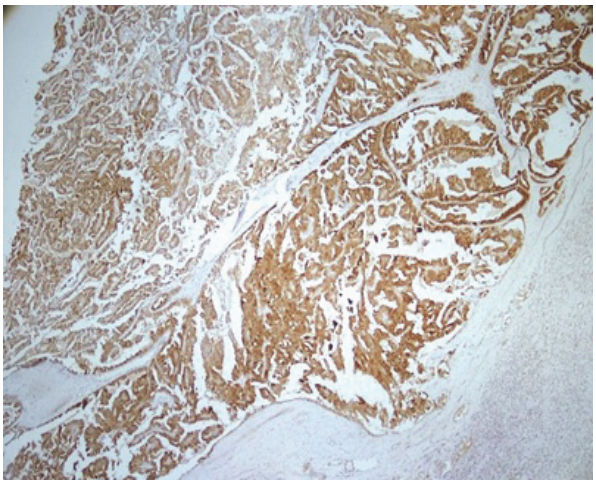

Figure 1b. Strong (+3) and 100% Cox-2 expression in papillary RCC.

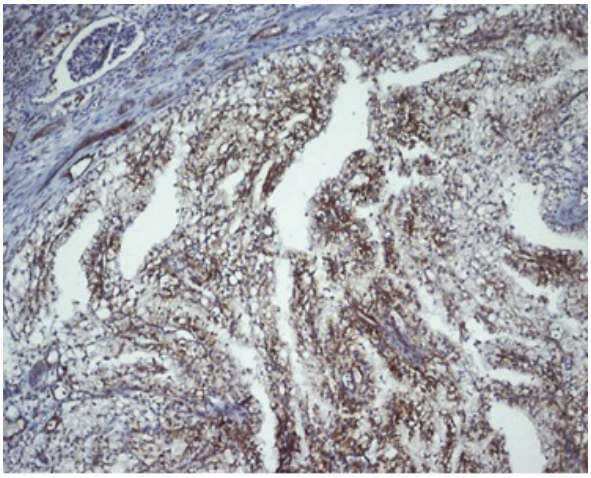

Figure 2a. Bax positivity in conventional RCC.

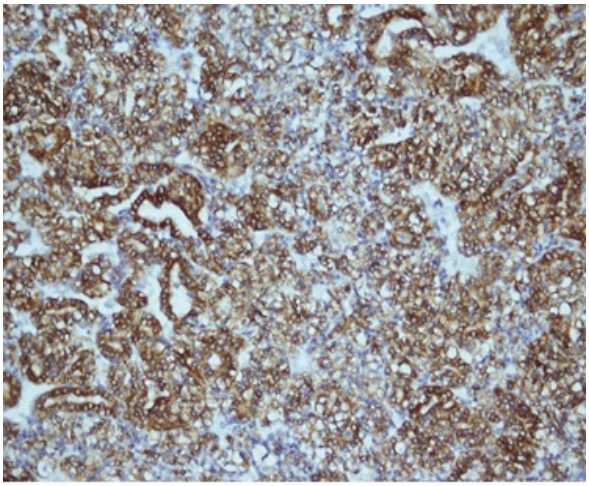

Figure 2b. Bcl-2 positivity in conventional RCC.

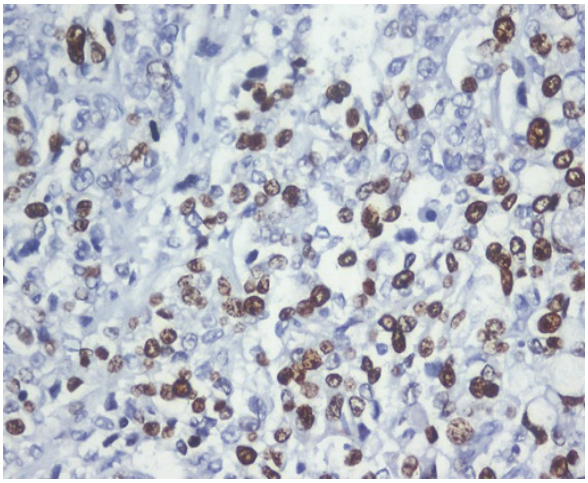

Figure 3a. Ki-67 proliferation index in RCC.

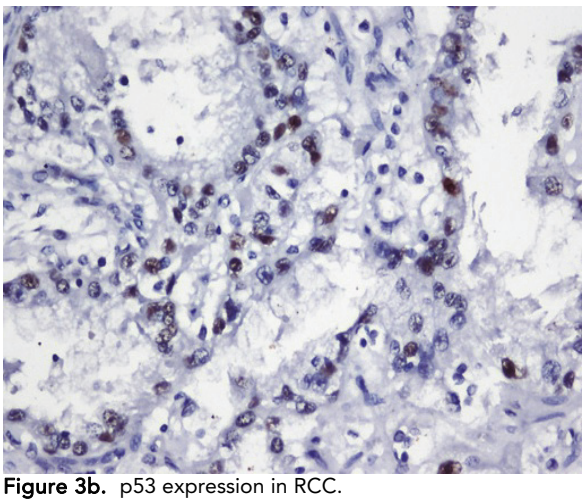

Figure 3b. p53 expression in RCC.

Ki-67 proliferation index was between 1% and 20% in all of the cases. The proliferation was ≤1 in % 77.1% cases (n=54), 1>-≤5% in 18.6% (n=13), and >5% in 4.3% (n=3),

respectively. The 2 of 3 patients with >5% Ki67 proliferation index had a proliferation index of 10%, while the other case had a Ki67 proliferation index of 20%.

Regarding 1% Ki-67 proliferation index as the median value and considering this value as the threshold between the low and high indexes, we found out that 77.1% of the patients had low Ki67 proliferation indexes (n=54) and 22.9% of them had high indexes (n=16). However, there was no significant association between Ki-67 proliferation index and any of the histopathological and clinical parameters.

Comparing the stage (pt) and grade (D1-2 and D3-4), one of the histopathological data, with other parameters, we found statistically significant differences between tumour nuclear grade and stage (p=0.006), as well as between tumour size (p=0.006) and perirenal fat tissue invasion (p=0.025) (Table 4). Tumour grade was significantly associated with tumour stage (p=0.004), tumor size (p=0.000), and metastasis (p=0.03) (Table 5). None of Cox2, Bcl2, Bax, and p53 expressions or Ki-67 proliferation index seemed to have any notable relationship with any of the parameters, including tumour stage, in predicting survival (p>0.05).

Table 4. The relationship between tumour's nuclear grade and other parametres.

		Grade 1-2 n/%	Grade 3-4 n/%	p
Stage				**0,006***
	1	20/83,3	4/16,7	
	2	12/60,0	8/40,0	
	3	8/33,3	16/66,7	
	4	1/50,0	1/50,0	
Tumour diameter				**0,006***
	<4	9/90,0	1/10,0	
	4-7	16/72,7	6/27,3	
	>7	16/42,1	22/57,9	
Fat tissue invasion				**0,025**
	Absent	36/65,5	19/34,5	
	Present	5/33,3	10/66,7	

*statistically significant at p<0,05

Table 5. The relationship between the tumour stage and other parametres.

		pT 1 (n/%)	pT 2(n/%)	pT 3(n/%)	pT 4(n/%)	p
Grade						**0,004***
	1-2	20/48,8	12/29,3	8/19,5	1/2,4	
	3-4	4/13,8	8/27,6	16/55,2	1/3,4	
Tumour diameter						**0,000***
	<4	10/100,0	0/0,0	0/0,0	0/0,0	
	4-7	14/63,6	1/4,5	7/31,8	0/0,0	
	>7	0/0,0	19/50,0	17/44,7	2/5,3	
Recurrence						0,848
	Absent	21/38,9	12/22,2	18/33,3	3/35,6	
	Prsent	2/25,0	3/37,5	3/37,5	0/0,0	
Metastasis						
	Absent	22/40	14/25,5	18/32,7	1/1,8	
	Present	1/14,3	1/14,3	3/42,9	2/28,6	
Present state						
	Alive	21/42,0	12/24,0	16/32,0	1/2,0	
	Dead	2/16,7	3/25,0	5/41,7	2/16,7	

*statistically significant at p<0,05

DISCUSSION

The most important determinants of prognosis in patients with RCC are regarded to be stage and nuclear grade (4). However, in many patients, stage and grading fall short in determining the course of the disease. Patients with the same grades and stages may have different survival rates (3, 16). Determining these patients with unforeseen prognosis is important in terms of oncology. Therefore additional parameters are needed to determine the behavior of the tumour (25).

The relationship between Cox-2 and tumour growth can be observed in the tumour (5-11). Thus the suitability of COX-2 as a desired treatment method in the treatment of human malignancies is still being evaluated clinically (26). Cox-2 expression may be initiated by inflammation. In many cancer cells, a number of signalling pathways such as mitogen-activated kinase (MAPK) or protein kinase C (PKC) may result in Cox-2 up-regulation and eventually these play a role in the growth and invasion of tumour cell through prostaglandin synthesis (12).

While Cox-2 is influential through target ras genes, it may also share synergistic interaction with other genes

such as p53 and Bcl-2. This point plays a critical role in the development and progression of RCC (27).

Although there are numerous studies on the relationship between Cox-2's role in the development of RCC and its prognosis, different findings in these publication hampers access to a final conclusion. For example, in two studies, Cox-2 is reported to have poor prognosis in positive RCC cases (13, 14, 30) while other studies associate Cox-2 with positive prognosis (14) or at least prove that it cannot be related to prognosis (4, 15-17). There are studies that report that Cox-2 expression can be associated with tumour grade, tumour stage (17, 29), tumour size (4). However some reports claim that Cox-2 expression is not related to any of the parameters (16).

In 70 RCC patients we went through, Cox -2 immunoreactivity is separately evaluated in terms of three different scores: staining percentage, intensity, and final staining score. Associating the intensity and percentage of Cox-2 staining with the histological type of tumour, it is observed that papillary RCC displays more cytoplasmic staining in percentages. These findings are consistent with Dirim et al.'s studies (16). As far as histopathological and clinical parameters are concerned, we could identify that a significant correlation between grade and the final score of Cox2. There was no other relationship between Cox2 and other parameters.

Apoptosis is an important defence mechanism that enables the identification and elimination of programmed cell death and cells with damaged DNA. Abnormalities in any of apoptotic mechanisms leads to the formation of cancer by leading to the deterioration of the control of cell proliferation. As regulators of apoptosis, p53, Bcl-2, and Bax are the most frequently mutated genes in cancers (19). p53 is one of the most well-known apoptotic genes and, undergoing genetic mutation, it causes tumorigenesis. Conversely, Bcl-2 is anti-apoptoticly involved.Bcl-2 is a member of Bcl-2 homologues family that regulate apoptosis in different ways. While some members of this multigene family (Bcl, Bcl-XL, Bag-1) block cell death, others (Bax, Bad, Bcl-XS) stimulate apoptosis. p53 is induced in response to DNA heavy damage. Once induced, p53 starts apoptosis by giving way for Bax induction, a member of the pro-apoptotic Bcl-2 family (19,30).

Mutant p53 expression is seen rarely in RCC. Vasvada et al.'s study reports that none of their RCC patients had p53 expression while Sejima et al. report to have observed p53 expression in only one of their RCC 53 patients (19, 31). Moch et al. have found p53 expression incidence rate to be 16% in RCC but they have also failed to identify a relationship between p53 expression, stage, and prognosis (32).

In some studies, high levels of p53 expression were observed in tumours of higher grades and stages (31,34); parallel to these findings, there are also studies that present a significant relationship between p53 over expression and shorter life span (32, 35).

Throughout the study, 94.3% of our 70 patients showed less than 10% staining (n=66) whereas only 5.7% showed staining more than 10% (n=4). Besides there was no significant relationship between p53 over expression and clinicopathological data.

Although mutant p53 is determined in low levels in RCC, the role of p53 in RCC pathogenesis cannot be denied. Other regulators of apoptosis can affect the function of wild-type p53. In vivo and in vitro studies have shown that Bax and Bcl2 bind p53 and affect its function. Bcl-2 prevents cell death though Bax accelerates it (19).There are publications that claim that there is no relationship between Bcl-2 expression and other important parameters like grade, stage, and life expectancy in RCC (31, 36-39) but Vasavada et al.'s study is one those works which report a close connection between high grade tumours and Bcl-2 and Bax expressions though they cannot prove that these expressions have any effect on prognosis either. Kallio et al.'s study of 138 patients illustrate that Bax positivity and Bcl-2 negativity can be associated with poor prognosis (18). In contrast, some other researches have found a relationship between increased Bcl-2 expression and small tumour size, low nuclear grades, and low stages (36, 40). Another study reports that Bax protein was expressed in only two of 57 RCC patients (41).

In our study, we also investigated the effects of Bax and Bcl-2 positivity on clinicopathological parameters and patient survival. 58.6% of our patients showed positive Bcl-2 staining while positive Bax staining was observed in 74.3% of our patients. There was no statistically significant connection between Bcl-2 and Bax expressions and any of the parameters.

Another prognostic indicator in RCC is the ratio of Ki-67 in the tumour. Cell proliferation is the simplest and most commonly used variable that determine tumour progression and prognosis. A nuclear antigen, Ki-67 is found in G1, S, G2, and M phases of the nuclei in all the cleaved cells. In other words, Ki-67 is a proliferation marker and to be found in proliferated active tumour cells. In RCC, Ki-67 proliferation index has already been associated with tumour grade and stage and it is said to be an independent risk factor in terms of prognosis (20, 21,39, 42). Papadopoulos et al. have found relationship between histological grade and proliferation rate that was determined by Ki-67 in RCC though they could not see the same relationship in clinical outcomes (43). In contrast, another study shows no connection between Ki-67 index, grade, and stage (18). Aaltomaa et al. report that Ki-67 positive cells concentration differ both in different tumours and within the same tumour. To eliminate the drawbacks, they have worked on the most proliferative and atypical areas (44). We, too, concentrated on the areas with high proliferative activity while in the assessment of Ki-67. Having identified Ki-67 proliferation index in tumour cells to be ≤1% in 54 cases and >1% in 16 cases, we did not observe any significant correlation between the proliferation index and the clinicopathological data.

As a result, we can conclude that ideal prognostic factors have not yet been defined for RCC although researches on potential prognostic factors are still carried out. Today, pathologic stage and nuclear grade are still the most important and strongest parametres in determining the prognosis of the disease. However, in some cases, even these parameters are insufficient to determine the prognosis. In this study, we evaluated the histological and clinicopathological parameters in renal cell carcinoma and the relationship between apoptosis and proliferation predictors such as Cox-2, Bax, Bcl-2, p53, and Ki-67, and assessed their effect on prognosis. Throughout our study, we did not determine any significant correlation between clinicopathological parameters including the aforementioned markers and prognosis. Pathological stage of the tumour only showed statistical significance in predicting the development of metastases yet it was ineffective in determining survival. This condition may be a result of the fact that we were unable to cover long follow-up periods. Considering the discordant results of the studies on Cox-2, Bax, Bcl-2, p53, and Ki-67 staining in RCC in the literature, we believe that more cases with longer follow-up periods and with standardised methods should be instigated in order to have a better understanding of these markers on prognosis in RCC.

REFERENCES

1. Eble JN SG, Epstein JI, Sesterhenn IA, , editor. World Health Organization Classification of Tumours. Lyon: IARC Press 2004.
2. Rodriguez R, Fishman EK, Marshall FF. Differential diagnosis and evaluation of the incidentally discovered renal mass. Semin Urol Oncol 1995;13:246-53.
3. Delahunt B. Histopathologic prognostic indicators for renal cell carcinoma. Semin Diagn Pathol 1998;15:68-76.
4. Cho DS, Joo HJ, Oh DK, Kang JH, Kim YS, Lee KB, et al. Cyclooxygenase-2 and p53 ekspression as prognostic indicators in conventional renal cell carcinoma. Yonsei Med J 2005;28;46:133-40.
5. Masunaga R, Kohno H, Dhar DK, Ohno S, Shibakita M, Kinugasa S, et al. Cyclooxygenase-2 expression correlates with tumor neovascularization and prognosis in human colorectal carcinoma patients. Clin Cancer Res 2000;6:4064-8.
6. Joo YE, Rew JS, Seo YH, Choi SK, Kim YJ, Park CS, et al. Cyclooxygenase-2 overexpression correlates with vascular endothelial growth factor expression and tumor angiogenesis in gastric cancer. J Clin Gastroenterol 2003;37:28-33.
7. Denkert C, Winzer KJ, Muller BM, Weichert W, Pest S, Kobel M, et al. Elevated expression of cyclooxygenase-2 is a negative prognostic factor for disease free survival and overall survival in patients with breast carcinoma. Cancer 2003;15;97:2978-87.
8. Chen YJ, Wang LS, Wang PH, Lai CR, Yen MS, Ng HT, et al. High cyclooxygenase-2 expression in cervical adenocarcinomas. Gynecol Oncol 2003;88:379-85.
9. Khuri FR, Wu H, Lee JJ, Kemp BL, Lotan R, Lippman SM, et al. Cyclooxygenase-2 overexpression is a marker of poor prognosis in stage I non-small cell lung cancer. Clin Cancer Res 2001;7:861-7.
10. Yoshimura R, Sano H, Masuda C, Kawamura M, Tsubouchi Y, Chargui J, et al. Expression of cyclooxygenase-2 in prostate carcinoma. Cancer 2000;1;89:589-96.
11. Shirahama T. Cyclooxygenase-2 expression is up-regulated in transitional cell carcinoma and its preneoplastic lesions in the human urinary bladder. Clin Cancer Res 2000;6:2424-30.
12. Miyata Y, Koga S, Kanda S, Nishikido M, Hayashi T, Kanetake H. Expression of cyclooxygenase-2 in renal cell carcinoma: correlation with tumor cell proliferation, apoptosis, angiogenesis, expression of matrix metalloproteinase-2, and survival. Clin Cancer Res 2003;9:1741-9.
13. Ronkainen H, Vaarala MH, Hirvikoski P, Ristimaki A. HuR expression is a marker of poor prognosis in renal cell carcinoma. Tumour Biol 2011;32:481-7.
14. Kankuri-Tammilehto MK, Soderstrom KO, Pelliniemi TT, Vahlberg T, Pyrhonen SO, Salminen EK. Prognostic evaluation of COX-2 expression in renal cell carcinoma. Anticancer Res 2010;30:3023-30.
15. Yoshimura R, Matsuyama M, Kawahito Y, Tsuchida K, Kuratsukuri K, Takemoto Y, et al. Study of cyclooxygenase-2 in renal cell carcinoma. Int J Mol Med 2004;13:229-33.
16. Dirim A, Haberal AN, Goren MR, Tekin MI, Peskircioglu L, Demirhan B, et al. VEGF, COX-2, and PCNA expression in renal cell carcinoma subtypes and their prognostic value. Int Urol Nephrol 2008;40:861-8.
17. Tuna B, Yorukoglu K, Gurel D, Mungan U, Kirkali Z. Significance of COX-2 expression in human renal cell carcinoma. Urology 2004;64:1116-20.
18. Kallio JP, Hirvikoski P, Helin H, Luukkaala T, Tammela TL, Kellokumpu-Lehtinen P, et al. Renal cell carcinoma MIB-1, Bax and Bcl-2 expression and prognosis. J Urol 2004;172:2158-61.
19. Vasavada SP, Novick AC, Williams BR. P53, bcl-2, and Bax expression in renal cell carcinoma. Urology 1998;51:1057-61.
20. Rioux-Leclercq N, Turlin B, Bansard J, Patard J, Manunta A, Moulinoux JP, et al. Value of immunohistochemical Ki-67 and p53 determinations as predictive factors of outcome in renal cell carcinoma. Urology 2000;55:501-5.
21. de Riese WT, Crabtree WN, Allhoff EP, Werner M, Liedke S, Lenis G, et al. Prognostic significance of Ki-67 immunostaining in nonmetastatic renal cell carcinoma. J Clin Oncol 1993;11:1804-8.
22. Yildiz E, Gokce G, Kilicarslan H, Ayan S, Goze OF, Gultekin EY. Prognostic value of the expression of Ki-67, CD44 and vascular endothelial growth factor, and microvessel invasion, in renal cell carcinoma. BJU Int 2004;93:1087-93.
23. Greene FL PD, Fleming ID., editor. American Cancer Joint Committee on Cancer in AJCC, Cancer Staging Manuel. Philedelphia: Lipincott-Raven; 2002.
24. Koga S, Nishikido M, Hayashi T, Matsuya F, Saito Y, Kanetake H. Outcome of surgery in cystic renal cell carcinoma. Urology 2000;56:67-70.
25. Kirkali Z, Yorukoglu K, Ozkara E, Kazimoglu H, Mungan U. Proliferative activity, angiogenesis and nuclear morphometry in renal cell carcinoma. Int J Urol 2001;8:697-703.
26. Khan Z, Khan N, Tiwari RP, Sah NK, Prasad GB, Bisen PS. Biology of Cox-2: an application in cancer therapeutics. Curr Drug Targets 2011;12:1082-93.
27. Chen Q, Shinohara N, Abe T, Watanabe T, Nonomura K, Koyanagi T. Significance of COX-2 expression in human renal cell carcinoma cell lines. Int J Cancer 2004;1;108:825-32.
28. Lee JW, Park JH, Suh JH, Nam KH, Choe JY, Jung HY, et al. Cyclooxygenase-2 expression and its prognostic significance in clear cell renal cell carcinoma. Korean J Pathol 2012;46:237-45.
29. Hashimoto Y, Kondo Y, Kimura G, Matsuzawa I, Sato S, Ishizaki M, et al. Cyclooxygenase-2 expression and relationship to tumour progression in human renal cell carcinoma. Histopathology 2004;44:353-9.
30. Ye D, Li H, Qian S, Sun Y, Zheng J, Ma Y. bcl-2/bax expression and p53 gene status in human bladder cancer:

relationship to early recurrence with intravesical chemotherapy after resection. J Urol 1998;160:2025-8; discussion 9.
31. Sejima T, Miyagawa I. Expression of bcl-2, p53 oncoprotein, and proliferating cell nuclear antigen in renal cell carcinoma. Eur Urol 1999;35:242-8.
32. Moch H, Sauter G, Gasser TC, Buchholz N, Bubendorf L, Richter J, et al. p53 protein expression but not mdm-2 protein expression is associated with rapid tumor cell proliferation and prognosis in renal cell carcinoma. Urol Res 1997;25 Suppl 1:S25-30.
33. Oda H, Nakatsuru Y, Ishikawa T. Mutations of the p53 gene and p53 protein overexpression are associated with sarcomatoid transformation in renal cell carcinomas. Cancer Res 1995;1;55:658-62.
34. Reiter RE, Anglard P, Liu S, Gnarra JR, Linehan WM. Chromosome 17p deletions and p53 mutations in renal cell carcinoma. Cancer Res 1993; 1;53:3092-7.
35. Uhlman DL, Nguyen PL, Manivel JC, Aeppli D, Resnick JM, Fraley EE, et al. Association of immunohistochemical staining for p53 with metastatic progression and poor survival in patients with renal cell carcinoma. J Natl Cancer Inst 1994; 5;86:1470-5.
36. Oudard S, Levalois C, Andrieu JM, Bougaran J, Validire P, Thiounn N, et al. Expression of genes involved in chemoresistance, proliferation and apoptosis in clinical samples of renal cell carcinoma and correlation with clinical outcome. Anticancer Res 2002;22:121-8.
37. Huang A, Fone PD, Gandour-Edwards R, White RW, Low RK. Immunohistochemical analysis of BCL-2 protein expression in renal cell carcinoma. J Urol 1999;162:610-3.
38. Uzunlar AK, Sahin H, Yilmaz F, Ozekinci S. Expression of p53 oncoprotein and bcl-2 in renal cell carcinoma. Saudi Med J 2005;26:37-41.
39. Hofmockel G, Wittmann A, Dammrich J, Bassukas ID. Expression of p53 and bcl-2 in primary locally confined renal cell carcinomas: no evidence for prognostic significance. Anticancer Res 1996;16:3807-11.
40. Lipponen P, Eskelinen M, Syrjanen K. Expression of tumour-suppressor gene Rb, apoptosis-suppressing protein Bcl-2 and c-Myc have no independent prognostic value in renal adenocarcinoma. Br J Cancer 1995;71:863-7.
41. Pammer J, Exner M, Regele H, Haitel A, Weninger W, Horvat R, et al. Expression of bcl-2, bcl-x, bax and bak in renal parenchyma, oncocytomas and renal cell carcinomas. Pathol Res Pract 1998;194:837-45.
42. Dudderidge TJ, Stoeber K, Loddo M, Atkinson G, Fanshawe T, Griffiths DF, et al. Mcm2, Geminin, and KI67 define proliferative state and are prognostic markers in renal cell carcinoma. Clin Cancer Res 2005;1;11:2510-7.
43. Papadopoulos I, Weichert-Jacobsen K, Wacker HH, Sprenger E. Correlation between DNA ploidy, proliferation marker Ki-67 and early tumor progression in renal cell carcinoma. A prospective study. Eur Urol 1997;31:49-53.
44. Aaltomaa S, Lipponen P, Ala-Opas M, Eskelinen M, Syrjanen K. Prognostic value of Ki-67 expression in renal cell carcinomas. Eur Urol 1997;31:350-5.

Received/Başvuru: 15.06.2013, Accepted/Kabul: 18.9.2013

Correspondence/İletişim

Fatma Seher PEHLİVAN
Özel Tınaztepe Hastanesi, Patoloji Kliniği, İZMİR, TURKEY
E-mail: seherfatma@hotmail.com

For citing/Atıf için

Pehlivan FS, Sari A, Gorgel SN, Morgul Y, Balci U, Ermete M, Sefik E. Relationship of COX-2, BAX, BCL-2, Ki67, p53 expression to clinicopathologic parameters and their impact on prognosis in renal cell carcinoma J Turgut Ozal Med Cent 2014;21:207-14 DOI: 10.7247/jtomc.2013.870