ORIGINAL PAPER

UROLOGICAL ONCOLOGY

Clinical, demographic and histopathological prognostic factors for urothelial carcinoma of the bladder

Ulku Kucuk[1], Emel Ebru Pala[1], Ebru Cakır[1], Ozlem Sezer[1], Umit Bayol[1], Rauf Taner Divrik[2], Ozgur Cakmak[3]

[1]*Izmir Tepecik Research and Training Hospital, Department of Pathology, Izmir, Turkey*
[2]*Sifa Universitiy Medical Faculty, Department of Urology, Izmir, Turkey*
[3]*Izmir Tepecik Research and Training Hospital, Department of Urology, Izmir, Turkey*

Citation: Kucuk U, Pala EE, Cakır E, Sezer Or, Bayol U, Divrik RT, Cakmak O. Clinical, demographic and histopathological prognostic factors for urothelial carcinoma of the bladder. Cent European J Urol. 2015; 68: 30-36.

Article history
Submitted: Sept. 5, 2014
Accepted: Nov. 23, 2014
Published on-line: March 13, 2015

Corresponding author
Ulku Kucuk
Izmir Tepecik Research and Training Hospital
Pathology Department, Yenisehir
35100 Izmir, Turkey
Gaziler Street
phone: +90 505 525 02 76
kucukulku@hotmail.com

Introduction Our aim is to evaluate the influence of clinical and histopathological parameters, including age, gender, tumor stage, grade, tumor differentiation, necrosis, lymphovascular/perineural invasion (LVI/PNI) and concomitant carcinoma in situ (CIS), on outcomes of patients with urothelial carcinoma of the bladder (UCB).

Material and methods A total of 84 patients who underwent radical cystectomy (RC) (n = 11) and radical cystoprostatectomy (n = 73) for muscle-invasive bladder cancer at our hospital between 2007-2013, were included in the study.

Results The mean age of patients at diagnosis was 66.1, of whom 75 were males and 9 were females. Of the 84 patients, 38 were ≤65 years and 46 were >65 years. Mean tumor diameter was 3.66 cm. There were 38 cases which showed divergent differentiations. Concomitant CIS was observed in 30 tumors, 41 cases showed tumor necrosis, 44 PNI and 61 LVI.

The rate of overall survival (OS) in patients aged ≤65 years was statistically significantly higher than in those aged >65 years. A negative statistical relationship was found between OS with lymph node metastasis (LNM) and tumor differentiation. On the other hand, necrosis did not remain significant on multivariate analysis. No statistically significant relationship was found between smoking, tumor stage, PNI, LVI and concomitant CIS and OS.

Conclusions In this study, advanced age, LNM, tumor differentiation were found to be independent prognostic risk factors associated with OS after RC. These additional factors, which may explain the different clinical course in patients with similar tumor stage and lymph node status, should be taken into consideration in treatment planning.

Key Words: urothelial carcinoma of the bladder ‹› overall survival ‹› tumor necrosis ‹› histology

INTRODUCTION

Urothelial carcinoma of the bladder (UCB) is the second most common genitourinary malignancy and is associated with a heterogeneous clinical outcome [1-4]. Radical cystectomy (RC) with bilateral pelvic lymph node dissection (PLND) is currently the gold standard treatment for muscle-invasive UCB [1, 5-9]. Unfortunately, 40% of patients with organ-confined disease at the time of cystectomy subsequently suffer recurrence. Several studies have evaluated the risk factors for recurrence and survival after cystectomy. Advanced pathologic stage, nodal involvement, grade and urinary obstruction have been reported as prognostic factors for survival and recurrence. However, some bladder cancer cases of similar stage and grade have demonstrated variable clinical outcomes after RC, so many attempts have been made to determine new and reliable prognostic factors [10-14].

The aim of the present study is to evaluate the influence of clinical and detailed histopathological pa-

doi: 10.5173/ceju.2015.01.465

rameters including age, gender, tumor stage, grade, tumor differentiation, necrosis, lymphovascular invasion (LVI), perineural invasion (PNI) and concomitant carcinoma in situ (CIS), on outcomes of patients with UCB treated with RC.

MATERIAL AND METHODS

A total of 84 patients who underwent RC (n = 11) and radical cystoprostatectomy (n = 73) for muscle-invasive bladder cancer (MIBC) at our institute between 2007-2013, were included in the study. Of the total, 79 underwent standard PLND. Bladder cancer was diagnosed histopathologically by transurethral resection in all patients before cystectomy. RC and standard PLND were performed using the standard technique. Surgical specimens were re-examined by 2 genitourinary pathologists applying a standardized reporting protocol. Tumor staging and grading were standardized according to the American Joint Committee on Cancer and World Health Organization. Tumor differentiation, depth of tumor invasion, necrosis, LVI, PNI and concomitant CIS were assessed histopathologically.

Statistically analyses of prognostic effects of age (65 years), gender, smoking status, pathologic tumor stage, lymph node metastasis (LNM), tumor differentiation, LVI, PNI and necrosis on overall survival (OS) were performed. Univariate OS after RS were estimated using the Kaplan-Meier method and log-rank statistics. Multivariate Cox regression models addresses OS after RS. The chi-square test was used to determine correlations among the variables. Statistical significance was set at $p < 0.05$. Statistical analyses were performed with SPSS v.15.

RESULTS

The mean age at diagnosis was 66.1 (min. 42, max. 84) and there were 75 (89.3%) males and 9 (10.7%) females. Of the 84 patients, 38 (45.2%) were under 65 years, whereas 46 (54.8%) were over 65 years. Mean tumor diameter was 3.66 cm (min: 0.70 cm, max: 8 cm). The average overall follow-up time was 17.6 months (SD ±15.1). At the time of analysis, 33 (39.3%) patients were alive with disease, whereas 51 (60.7%) were dead. In 75 patients with available habitual data, 64 (85.3%) were recorded as heavy smokers.

The pathologic tumor stages were 4 (4.8%), 8 (9.5%), 20 (23.8%), 37 (44%) and 15 (17.9%) for Ta, T1, T2, T3 and T4 respectively. Of the patients with Ta and T1 tumors who had an extensive mass, which could not be totally excised by TUR, or intensive gross hematuria, underwent RC. Of the total 84 cases, 79 underwent standard PLND and LNM was detected in 25 patients (29.8%).

Of the 84 cases, 82 were high grade on histopathological examination. Both of the low grade tumors were stage Ta and exhibited no tumor necrosis, CIS, LVI and PNI. One of the patients was alive and the other one died of a non-tumoral cause.

The histologic type was pure urothelial carcinoma (UC) in 46 (54.8%) cases. Of the 38 (45.2%) cases which showed divergent differentiations or components, 26 (68.4%) had squamous differentiation, 7 (18.4%) sarcomatoid, 1 (2.6%) glandular differentiation, 1 (2.6%) clear cell, 1 (2.6%) neuroendocrine, 1 (2.6%) micropapillary and 1 (2.6%) squamous plus sarcomatoid components.

Concomitant CIS was observed in 30 (30.7%) tumors. 41 (48.8%) cases showed tumor necrosis, 44 (52.4%) PNI and 61 (72.3%) LVI. Demographic, clinical and pathological characteristics are summarized in Table 1.

The relationship of tumor necrosis with pathologic tumor stage and LNM was evaluated. Accordingly, tumor necrosis was found in 25% (1/4) of Ta tumors, 25% (2/8) of T1 tumors, 50% (10/20) of T2 tumors, 48.6% (18/37) of T3 tumors and 66.7% (10/15) of T4 tumors. No statistically significant relationship was found between tumor necrosis and pathologic tumor

Table 1. *Univariate analysis of demographic, clinical and pathological characteristics for overall survival*

Clinicopathologic factors	Category	n (%)	p values
Age	>65 ≤65	46 (54.8%) 38 (45.2%)	<0.001
Gender	Male Female	75 (89.3%) 9 (10.7%)	0.23
Tobacco consumption	Absence Presence	11 (14.7%) 64 (85.3%)	0.81
Pathologic stage	Ta T1 T2 T3 T4	4 (4.8%) 8 (9.5%) 20 (23.8%) 37 (44%) 15 (17.9%)	0.15
Lymph node status	N0 N1 Nx	54 (64.3%) 25 (29.8%) 5 (6%)	0.001
Histopathologic Differentiation	Absence Presence	46 (54.8%) 38 (45.2%)	0.011
LVI	Absence Presence	23 (27.7%) 61 (72.3%)	0.37
PNI	Absence Presence	40 (47.6%) 44 (52.4%)	0.06
UCIS	Absence Presence	54 (64.3%) 30 (35.7%)	0.24
Tumor necrosis	Absence Presence	43 (51.2%) 41 (48.8%)	0.025

stage (p = 0.32). Tumor necrosis was found in 60% (15/35) of N1 cases, 46.3% (25/54) of N0 cases and 20% (1/5) of Nx cases. No statistically significant relationship was found between tumor necrosis and LNM (p = 0.21).

The evaluation of OS data revealed that 55.6% (5/9) of female patients, and 37.3% (28/75) of male cases were alive. There was no statistically significant relationship between OS and gender (p = 0.23).

In this study, 57.9% (22/38) of the patients aged ≤65 years and 23.9% (11/46) of patients aged >65 were alive. The rate of OS in patients aged ≤65 years was statistically significantly higher than those aged >65 years (p <0.001) (Figure 1).

On the other hand, 36.4% (4/11) of non-smokers and 45.3% (29/64) of smokers were alive. No statistically significant relationship was noted between smoking and OS (p = 0.81).

With regard to pathological tumor stage, 75% (3/4) of Ta patients, 75% (6/8) of T1 patients, 40% (8/20) of T2 patients, 37.8% (14/37) of T3 patients, and 13.3% (2/15) of T4 patients were alive. The cause of death in the Ta and T1 patients was not related to the primary tumor. No statistically significant re-

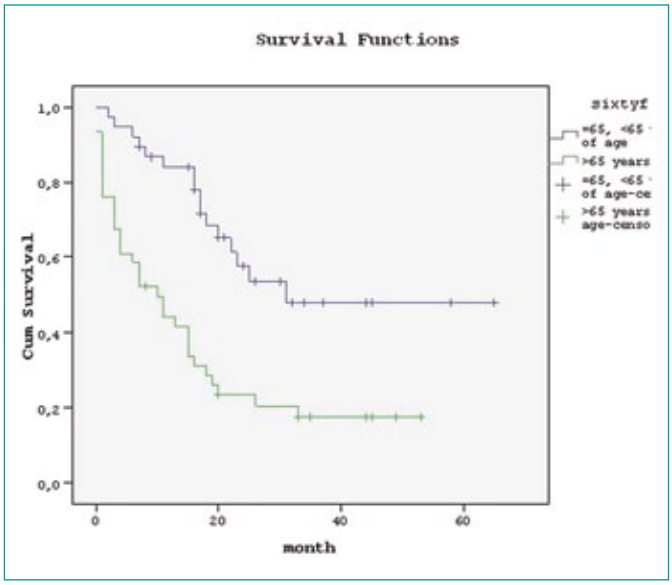

Figure 1. *Kaplan-Meier curves of overall survival stratified according to age (≤65, <65).*

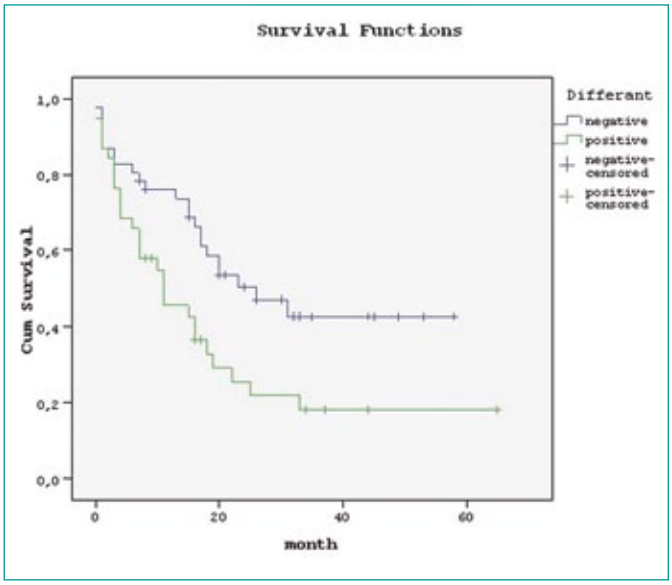

Figure 3. *Kaplan-Meier curves of overall survival stratified according to the histological differentiation of tumors.*

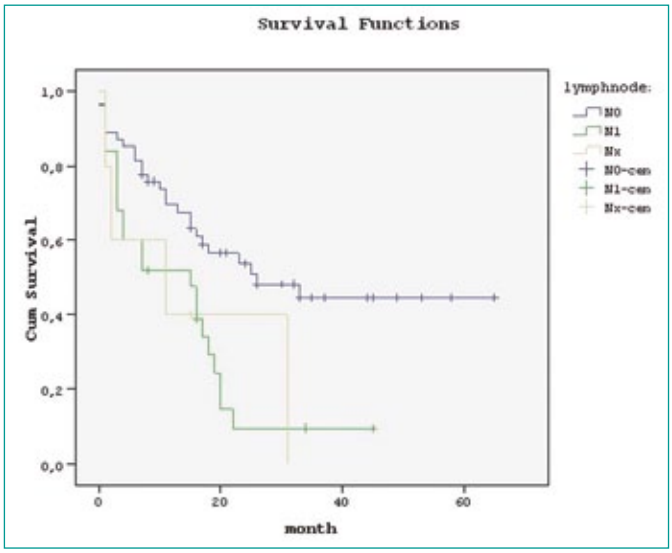

Figure 2. *Kaplan-Meier curves of overall survival stratified according to lymph node status.*

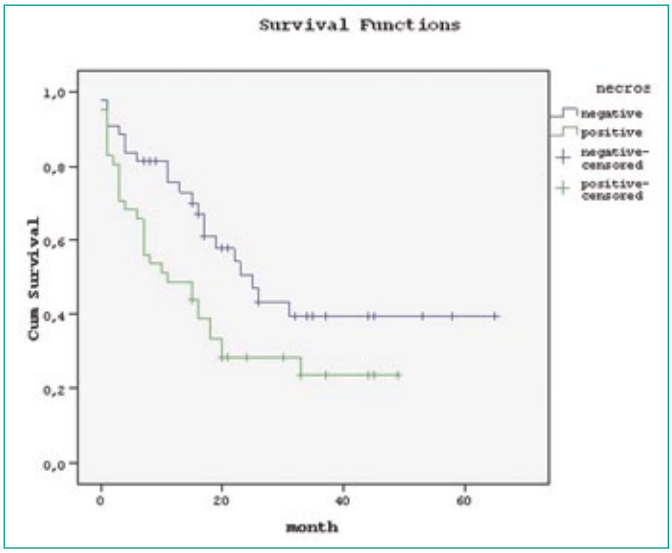

Figure 4. *Kaplan-Meier curves of overall survival stratified according to tumor necrosis.*

lationship was found between OS and tumor stage (p = 0.15).

In this study, 51.9% (28/54) of the patients with no LNM, 16% (4/25) of the patients with metastasis and 20% (1/5) of the patients who did not undergo lymph node dissection were alive. A negative statistical relationship was found between LNM and OS in patients undergoing lymph node dissection (p = 0.001) (Figure 2).

The evaluation of the relationship between tumor differentiation and OS revealed that 26.9% (7/26) of patients with squamous differentiation and 14.3% (1/7) of patients with sarcomatoid differentiation were alive. One patient with micropapillary differentiation and another with clear cell differentiation were alive, whereas those with tumors exhibiting squamous and sarcomatoid differentiation, glandular and neuroendocrine differentiation were not alive. A negative statistically significant relationship was found between tumor differentiation and OS (p = 0.01) (Figure 3).

In addition, 26.8% (11/41) of patients with tumor necrosis and 51.2% (22/43) of those without tumor necrosis were alive. A negative statistical relationship was found between tumor necrosis and OS (p = 0.025) (Figure 4).

No statistically significant relationship was found between PNI, LVI and concomitant CIS and OS (p = 0.06, p = 0.37, p = 0.24, respectively).

On univariate analysis, variables such as age (>65), LNM, tumor necrosis, differentiation status were all demonstrated to be significant prognostic factors affecting the OS. On the other hand, necrosis did not remain significant on multivariate analysis (Table 2).

DISCUSSION

UCB is the second most common tumor of the genitourinary tract [1-4]. Globally, UCB is the seventh most common cancer in males and seventeenth in females [1, 5]. Recently, the pathologic tumor stage and the presence of LNM have been reported to be the most important prognostic factors [1, 2, 12]. However, reports of different clinical outcomes, in patients with similar stages of disease following RC, have prompted the investigation of other factors that may affect prognosis.

A number of studies have reported that the prognosis of UC in females is much worse than that in males [5, 15-18]. A large European epidemiological study of 1.2 million patients reported that the 5-year cancer-specific mortality was 30% lower in females, which, however, was not the case in bladder carcinomas. The study also demonstrated that UC followed a more aggressive clinical course in females than that in males [5]. Horstmann et al. reported that, in a MICB series of 455 patients, 129 of whom were females, the 10-year survival was lower in females compared to that in males [19]. Aggressive tumor biology in females is considered to be responsible for shorter survival [5]. In our series, where most patients were males (89.3%), there were only 9 female patients and there was no statistically significant difference between OS and gender (p = 0.23). The absence of a statistically significant relationship between gender and OS can be attributed to the small number of female patients in this study.

In a study by Mitra et al., where 259 tumors with tumor differentiation were compared with pure UCB, the OS was lower in patients with differentiation and aged >65 years [13]. In our series, the rate of OS was higher in patients aged ≤65 years (57.9%) compared to those aged >65 years (23.9%) and the difference was statistically significant (p <0.001).

Previous studies have demonstrated that tobacco consumption and the number of cigarettes smoked per day are associated with advanced tumor stage and grade in newly-diagnosed UCB [14, 20]. A study of 1506 patients with UCB by Rink et al., reported the association between smoking and cancer-specific mortality, which, however, lost its significance on multivariate analysis [14]. In the mentioned study, cumula-

Table 2. *Multivariate analysis of parameters predicting overall survival*

Variable	Levels	Hazard Ratio	95% CI Lower Bound	95% CI Upper Bound	p value
Age (>65)	≤65 years >65 years	2.969	1.550	5.684	0.001
Lymph node metastasis	N0 N1	2.204	1.223	3.970	0.009
Differentation	Negative Positive	2.116	1.173	3.818	0.013
Tumor necrosis	Negative Positive	1.601	0.878	2.917	1.124

tive cigarette exposure was associated with aggressive tumor biology in patients treated with RC. Lifetime cumulative exposure to cigarette carcinogens leads to cumulative molecular changes in UCB, thus affecting the biologic and clinical behavior of the tumor [21]. In our study, of the 75 patients with available data on tobacco consumption, 64 were current smokers. Even though no statistically significant relationship was noted between tobacco consumption and OS, we believe that our series is not adequately powered to make such a comparison, due to the imbalance between the number of patients ($p = 0.81$).

Tumor stage and LNM have been reported to be the most important prognostic factors in UC [1, 8, 22]. In our series, even though the rates of OS decreased significantly as the tumor stage increased, no statistically significant relationship was found between tumor stage and OS, probably due to the small number of patients in our series ($p = 0.15$).

It has been reported in the literature that 25% of the patients undergoing cystectomy had LNM [23]. In our series, the rate of LNM was 29.8%, which is consistent with the literature. There was also a negative statistical relationship between LNM and OS ($p = 0.001$).

UCB is a tumor manifesting clinical and morphological differences and has a distinct capacity for histological differentiation [13]. Squamous and glandular differentiation are the most common variants of UCB [13]. The rate of the variants of UC, which have been reported to be poor prognostic factors, varies over a wide range (from 7% to 81%) [24, 25]. This situation has been considered to be caused by differences in disease stage, sampling techniques and pathologic evaluation [12]. Furthermore, there is no standard technique for quantitative measurement of the extent of tumor differentiation [12]. In a study of 1984 patients with UCB by Xylinas et al., 488 patients had UC variants, with squamous differentiation being the most common (227 patients) and glandular differentiation, the second most common (75 patients) [12].

In patients with differentiation, the disease followed a more aggressive course biologically [12]. These patients were at significantly higher risk for disease recurrence and cancer-specific mortality compared to those with pure UCB patients. In addition, non-squamous variants were found to be associated with worse prognosis compared to pure UCB and UCB with squamous differentiation. In our series, squamous differentiation was the most common (26 patients, 68.4%), followed by sarcomatoid differentiation (7 patients, 18.4%). A statistically significant difference was found in OS between pure UCB and UCB with variant histology ($p = 0.01$). There was no statistically significant difference in OS between the subtypes of squamous and non-squamous differentiation ($p = 0.34$).

Previous studies have reported that the rate of LVI ranges from 35% to 55% in MIBC [8, 22, 26, 27]. It has been reported that routine HE staining may be enough to assess vascular invasion [26]. In our study, LVI was assessed on HE sections and the rate of LVI was found to be 72% (61 cases). The high rate of LVI is probably due to the difficulty in distinguishing LVI from tissue retraction artifacts. Thus, we believe that, the assessment of LVI carried out by HE staining, should be supported by the use of other immunohistochemical markers.

The prognostic role of LVI in UCB remains controversial. Some studies have reported that LVI is a poor prognostic factor, whereas other studies have suggested that LVI is not a prognostic predictor of LNM, survival or recurrence [8, 22, 26]. Some of the studies, which identified LVI as a poor prognostic factor, reported that LVI did not remain significant on multivariate analysis [8, 22, 27]. No statistically significant relationship was noted between LVI and OS in our study ($p = 0.37$).

Controversial results have been reported in studies investigating the effects of PNI on prognosis. Some studies have reported that PNI is associated with LNM and distant metastases, whereas other studies have demonstrated that tumor stage, LNM, urethral obstruction, LVI and PNI are significant prognostic factors on univariate analysis; however, only tumor stage and LNM are independent prognostic factors on multivariate analysis [10]. A retrospective review of 125 patients by Hong et al., reported that LVI and PNI were prognostic factors on univariate analysis, whereas only the tumor stage and vascular invasion were statistically significant on multivariate analysis [10]. In this study, there was a small difference in OS between patients with and without PNI, and no statistically significant relationship was noted ($p = 0.06$). A large series of 1425 patients with UC of the upper urinary tract by Zigeuner et al., reported that extensive tumor necrosis (>10% of the tumor) was associated with aggressive biology. However, this association was not observed in the presence of focal necrosis [11]. In a series of 98 patients with UCB by Ord et al., tumor necrosis was associated with advanced tumor stage and nodal metastasis, and tumor necrosis was found to be the independent prognostic factor on univariate and multivariate analyses [28]. In the present study, 73.2% of the patients with tumor necrosis were not alive at the time of follow-up. There was a negative statistical relationship between tumor necrosis and OS ($p = 0.025$) and also the tumor necrosis in N1 patients was higher than that in N0 patients, which however did not reach

the level of statistical significance (p = 0.21). Similarly, advanced tumor stage was associated with an increased rate of tumor necrosis, but no statistically significant relationship was found due to the small number of patients in our study (p = 0.32). Numerous cytogenetic, molecular, genetic and immunohistologic studies revealed similar molecular changes in CIS and invasive UC [4]. The presence of isolated or concomitant CIS carries a higher risk of the disease progressing to MIBC [29]. There are numerous studies reporting that the presence of CIS and concomitant non-invasive UC following RC is associated with a poor clinical course [4]. On the other hand, a study by Nuhn et al. of 3973 patients treated with RC, reported no association between concomitant CIS and clinical outcome and the prognostic value of concomitant CIS in UCB could not be confirmed [4]. Similarly in our study no statistically significant difference was noted in OS between the patients with and without concomitant CIS (p = 0.24).

CONCLUSIONS

Advanced age (>65), LNM, tumor differentiation and tumor necrosis were found to be independent prognostic risk factors associated with OS after RC. Tumor necrosis did not remain significant on multivariate analysis. The presence of concomitant CIS had no effect on prognosis. These additional factors, which may explain the different clinical course in patients with similar tumor stage and lymph node status, should be taken into consideration in treatment planning.

References

1. Bruins HM, Arends TJH, Pelkman M, Hulsbergen-van de Kaa H, van der Heijden AG, Witjes JA. Radical cystectomy in a Dutch University Hospital: Long term outcomes and prognostic Factors in a homogenous surgery-only series. Clin Genitourinaty Cancer. 2013; 12: 190-195.
2. Stein JP, Lieskovsky G, Cote R, Groshen S, Feng AC, Boyd S, et al. Radical cystectomy in the treatment of invasive bladder cancer: Long-term results in 1.054 patients. J Clin Oncol. 2001; 19: 666-675.
3. May M, Bastian PJ, Brookman-May S, Fritsche HM, Tilki D, Otto W, et al. Gender-specific differences in cancer-specific survival after radical cystectomy for patients with urothelial carcinoma of the urinary bladder in pathologic tumor stage T4a. Urol Oncol. 2013; 31: 1141-1147.
4. Nuhn P, Bastian PJ, Novara G, Svatek RS, Karakiewicz P, Skinner E, et al. Concomitant carcinoma in situ in cystectomy specimens is not associated with clinical outcomes after surgery. Urol Int. 2011; 87: 42-48.
5. Otto W. May M, Fritsche HM, Dragun D, Aziz A, Gierth M, et al. Analysis of sex differences in cancer-specific survival and perioperative mortality following radical cystectomy: Results of a large German multicenter stdy of nearly 2500 patients with urothelial carcinoma of the bladder. Gend Medi. 2012; 9: 481-489.
6. Honma I, Masumori N, Sato E, Takayanagi A, Takahashi A, Itoh N, et al. Local recurrence after radical cystectomy for invasive bladder cancer: An analysis of predictive factors. Urology. 2004; 64: 744.
7. Wright JL, Lin DW, Porter MP. The association between extent of lymphadenectomy and survival among patients with lymph node metastases undergoing radical cystectomy. Cancer. 2008; 112: 2401-2408.
8. Lotan Y, Gupta A, Shariat SF, Palapattu GS, Vazina A, Karakiewicz PI, et al. Lymphovascular invasion is independently associated with overall survival, cause-specific survival and local and distatnt recurrence in patients with negative lymph nodes at radical cystectomy. J Clin Oncol. 2005; 23: 6533-6539.
9. May M, Stief C, Brookman-May S, Otto W, Gilfrich C, Roigas J, et al. Gender-dependent cancer-specific survival following radical cystectomy. World J Urol. 2012; 30: 707-713.
10. Hong SK, Kwak C, Jeon HG, Lee E, Lee SE. Do vascular, lymphatic and perineural invasion have prognostic implications for bladder cancer after radical cystectomy? Urology. 2005; 65: 696-702.
11. Zigeuner R, Shariat SF, Margulis V, Karakiewicz PI, Roscino M, Weizer A, et al. Tumour necrosis is an indicator of aggressive biology in patients with urothelial carcinoma of the upper urinary tract. Eur Urol. 2010; 57: 575-581.
12. Xylinas E, Rink M, Robinson BD, Lotan Y, Babjuk M, Brisuda A, et al. Impact of histological variants on oncological outcomes of patients with urothelial carcinoma of the bladder treated with radical cystectomy. Eur J Cancer. 2013; 49: 1889-1897.
13. Mitra AP, Bartsch CC, Bartsch G, Miranda G, Skinner EC, Daneshmand S. Does presence of squamous and glandular differenriation in urothelial carcinoma of the bladder at cystectomy portend poor prognosis? An intensive case-control analysis. Urol Oncol. 2014; 32: 117-127.
14. Rink M, Zabor EC, Furberg H, Xylinas E, Ehdaie B, Novara G, et al. Impact of smoking and smoking cessation on outcomes in bladder cancer patients treated with radical cystectomy. Eur Urol. 2013; 64: 456-464.
15. Fajkovic H, Halpern JA, Cha EK, Bahadori A, Chromecki TF, Karakiewicz PI, et al. Impact of gender on bladder cancer incidence, staging and prognosis. World J Urol. 2011; 29: 457-463.
16. Cao D, Vollmer RT, Luly J, Jain S, Roytman TM, Ferris CW, et al. Comparison of 2004 and 1973 World Health Organization grading systems and their relationship to pathologic staging for predicting long-term prognosis in patients with urothelial carcinoma. Urology. 2010; 76: 593-599.
17. Stenzl A, Cowan NC, De Santis M, Kuczyk MA, Merseburger AS, Ribal MJ, et al. Treatment of muscle-invasive and metastatic bladder cancer: update of the EAU guidelines. Eur Urol. 2011; 59: 1009-1018.

18. Babjuk M, Oosterlinck W, Sylvester R, Kaasinen E, Böhle A, Palou-Redorta J, et al. EAU guidelines on non-muscle-invasive urothelial carcinoma of the bladder, the 2011 update. Eur Urol. 2011; 59: 997-1008.

19. Horstmann M, Witthuhn R, Falk M, Stenzl A. Gender-specific differences in bladder cancer: a aretrospective analysis. Gend Med. 2008; 5: 385-394.

20. Freedman ND, Silverman DT, Hollenbeck AR, Schatzkin A, Abnet CC. Association between smoking and risk of bladder cancer among men and women. JAMA. 2011; 306: 737-745.

21. Reznikoff CA, Sarkar S, Julicher KP, Burger MS, Puthenveettil JA, Jarrard DF, et al. Genetic alterations and biological pathways in human bladder cancer pathogenesis. Urol Oncol. 2000; 5: 191-203.

22. Hara S, Miyake H, Fujisawa M, Okada H, Arakawa S, Kamidono S, et al. Prognostic variables in patients who have undergone radical cystectomy for transitional cell carcinoma of bladder. Jpn J Clin Oncol. 2001; 31: 399-402.

23. Stein JP, Quek ML, Skinner DG. Lymphadenectomy for invasive bladder cancer. I.Historical perspective and contemporary rationale. BJU Int. 2006; 97: 227-231.

24. Chalasani V, Chin JL, Izawa JI. Histologic variants of urothelial bladder cancer and nonurothelial histology in bladder cancer. Can Urol Assoc J. 2009; 3: 193-198.

25. Black PC, Brown GA, Dinney CP. The impact of variant histology on the outcome of bladder cancer treated with curative intent. Urol Oncol. 2009; 27: 3-7.

26. Leissner J, Koeppen C, Wolf HK. Prognostic significance of vascular and perineural invasion in urothelial bladder cancer treated with radical cystectomy. J Urol. 2003; 169: 955-960.

27. Ennis RD, Petrylak DP, Singh P, Bagiella E, O'Toole KM, Benson MC, et al. The effect of cystectomy and perioperative methotrexate, vinblastine, doxorubicin and cisplatin chemotheraphy on the risk and pattern of relapse in patients with muscle invasive bladder cancer. J Urol. 2000; 163: 1413-1418.

28. Ord JJ, Agrawal S, Thamboo TP, Roberts I, Campo L, Turley H, et al. An investigation into the prognostic significance of necrosis and hypoxia in high grade and invasize bladder cancer. J Urol. 2007; 178: 677-682.

29. Shariat SF, Palapattu GS, Karakiewicz PI, Rogers CG, Vazina A, Bastian PJ, et al. Concomitant carcinoma in situ is a feature of aggressive disease in patients with organ-confined TCC at radical cystectomy. Eur Urol. 2007; 51: 152-160.