Prognostic value of estrogen receptors in patients who underwent prostatectomy for non-metastatic prostate cancer

YAVUZ MERT AYDIN[1], AHMET BILGEHAN ŞAHIN[2], RABIA DÖLEK[3], BERNA AYTAÇ VURUŞKAN[3], GÖKHAN OCAKOĞLU[4], HAKAN VURUŞKAN[1], İSMET YAVAŞCAOĞLU[1] and BURHAN COŞKUN[1]

Departments of [1]Urology, [2]Medical Oncology, [3]Pathology and [4]Biostatistics, Bursa Uludag University, 16059 Bursa, Turkey

Received September 7, 2022; Accepted November 29, 2022

DOI: 10.3892/ol.2023.13664

Abstract. Estrogen receptors in prostate cancer (PCa) are a subject of debate. The aim of the present study was to investigate whether estrogen receptor-α (ERα) and estrogen receptor-β (ERβ) impact the biochemical recurrence (BCR) of non-metastatic PCa after surgery. Following the application of the exclusion criteria, data from 108 patients who underwent laparoscopic radical prostatectomy between January 2011 and December 2019 were retrospectively evaluated. A total of 36 patients with BCR constituted the BCR group. The control group was formed using the Propensity Score Matching (PSM) method with a 1:2 ratio, including parameters with well-studied effects on BCR. The median follow-up time was 74.3 (range, 30-127.5) months in the BCR group and 66.6 (range, 31.5-130) months in the control group. Pathology specimens from the two groups were immunohistochemically stained with ERα and ERβ antibodies. Logistic regression analysis and survival analysis were performed. No differences in clinico-pathological characteristics were detected between the two groups. The patients with ERα(-)/ERβ(+) staining results had a significantly fewer BCRs than other patients (P=0.024). In the logistic regression analysis, patients with ERα(-)/ERβ(+) PCa also had a significantly lower risk of recurrence (P=0.048). In the survival analysis, the 5-year BCR-free survival rate of patients with ERα(-)/ERβ(+) PCa was higher than that of other patients (85.7 vs. 66.1%; P=0.031). Excluding the effects of well-studied risk factors for recurrence by the PSM method, the present study showed that ERα and ERβ have prognostic value for non-metastatic PCa. The 5-year BCR-free survival rate is significantly higher in patients whose PCa tissue has ERα(-)/ERβ(+) staining results.

Correspondence to: Dr Yavuz Mert Aydın, Department of Urology, Bursa Uludag University, 3 Izmir Street, Gorukle Campus, 16059 Bursa, Turkey
E-mail: yavuzmertaydin@gmail.com

Key words: prostate cancer, estrogen receptor-α, estrogen receptor-β, laparoscopic radical prostatectomy, biochemical recurrence-free survival

Introduction

Prostate cancer (PCa) is the most common type of cancer in men aged >50 years and the fifth most common cause of cancer-associated mortality (1). Androgens are essential for prostate development, growth and secretory functions, and the development of PCa is primarily androgen dependent (2). However, some studies have indicated that estrogens may also play a role in the development of PCa (3).

Estrogens exert their effects at the cellular level through two receptors: Estrogen receptor-α (ERα) and estrogen receptor-β (ERβ) (3). Various mechanisms for ERα and ERβ in PCa have been identified. These include the fusion of transmembrane protease serine 2 with v-ets avian erythroblastosis virus E26 oncogene homolog, the most common gene fusion in the pathogenesis of PCa, which is increased through ERα activity and decreased via ERβ activity (4). In addition, the ERβ receptor has been reported to inhibit cell growth, decrease epithelial-mesenchymal transition-related aggressive behavior and induce apoptosis in PCa cells (5,6). Although the results in the literature are conflicting, ERα is associated with malignant transformation from high-grade prostatic intraepithelial neoplasia (PIN) to PCa, while ERβ has antiproliferative, anti-invasive and pro-apoptotic effects (7-14).

To the best of our knowledge, while numerous preclinical studies have evaluated the effects of ERα and ERβ on PCa, only three studies have investigated their impact on patients with non-metastatic PCa (15-17). Moreover, these three studies have conflicting results and certain limitations. Therefore, the present study aimed to clarify whether estrogen receptors affect the development of biochemical recurrence (BCR) after prostatectomy by balancing well-known risk factors for BCR in patients with non-metastatic PCa.

Materials and methods

Study population. The electronic medical records of 514 patients who underwent laparoscopic radical prostatectomy (LRP) for PCa between January 2011 and December 2019 were reviewed in the Department of Urology, Bursa Uludag University (Bursa, Turkey). Patients who had incomplete medical records (n=87), pathological T4 stage disease after LRP (n=2), positive surgical margins (n=111) and adjuvant androgen deprivation therapy (ADT) or external beam radiotherapy (EBRT) due to high

risk (n=56) were excluded. Lymph node involvement, which was planned to be an exclusion criterion, was not observed in any of the remaining 258 patients. BCR was defined as a prostate-specific antigen (PSA) level >0.2 ng/ml in at least two serial measurements 3 weeks apart (18). Patients with BCR (n=36) were identified and constituted the BCR group. The control group (n=72) was established by the propensity score matching (PSM) method among the other 222 patients in a 1:2 ratio. The 150 patients that remained after PSM were excluded from the study (Fig. 1). The Institutional Review Board of Bursa Uludag University approved the study (approval no. 2021-2/15). The study protocol complied with the tenets of the Declaration of Helsinki.

Immunohistochemical (IHC) staining and pathological assessment. IHC staining and pathological reevaluation were performed by two independent pathologists, one of whom had 26 years of experience in uropathology. The tissue sections of the patients stained with hematoxylin and eosin (H&E) were examined, and for each case, the slides best representing the morphology of the lesion and Gleason score (GS) were selected for evaluation. Paraffin blocks of these specimens were obtained from the pathology archive. From these blocks, 4-μm sections were prepared for the IHC staining of ERα and ERβ. Paraffin sections were baked overnight at 50˚C. The sections to be used for the IHC method were prepared on positively charged slides. The deparaffinization step was performed using the Ventana Discovery XT platform with EZ prep solution (catalogue number 950-100; Roche Diagnostics) at 75˚C for 8 min. The standard antigen retrieval method was heat-induced epitope retrieval in Tris-Ethylene diamine tetra acetic acid buffer (pH 7.8) at 95˚C for 64 min [standard cell conditioning solution (CC1)], performed using the Ventana Discovery XT (catalogue number 950-124; Roche Diagnostics). For blocking endogenous peroxides and protein, the Ventana Discovery XT platform was used with Inhibitor ChromoMap at 37˚C for 4 min. The primary antibodies used were ERα (1:250 dilution; F-10; catalogue number sc-8002) and Erβ (1:250 dilution; B-1; catalogue number sc-390243) (both Santa Cruz Biotechnology, Inc.). ERα primary antibody was incubated at 37˚C for 20 min, while Erβ primary antibody was incubated at 37˚C for 32 min.

The ultra views Universal diaminobenzidine (DAB) Detection Kit solution (Roche Diagnstics) was used. The kit contains 5 pre-diluted and ready-to-use dispensers comprising DAB inhibitor, horseradish peroxidase (HRP) multimer, DAB chromogen, DAB H_2O_2 and copper. Slides were lightly counterstained with hematoxylin (catalogue number 760-2021; Roche Diagnostics) and incubated at 37˚C for 12 min. Post-counterstain slides were incubated for 8 min with Bluing Reagent (catalogue number 760-2037; Roche Diagnostics). Slides were washed in warm tap water with detergent, and after dehydration in graded ethanol and xylene, they were coverslipped. Endocervical tissue was used as a positive control for the ERα antibody, and testicular tissue served as a positive control for the ERβ antibody. Finally, tumor specimens were observed under a light microscope (model BX51TF; Olympus Corporation).

The IHC stained slides were analyzed simultaneously with H&E stained slides from the archive for each case to avoid the false positivity that may occur due to intense background staining during evaluation. The ERα antibody was evaluated with regard to the nuclear staining of tumor epithelial cells, and ERβ antibody was evaluated with regard to the cytoplasmic staining of tumor epithelial cells. The percentage and intensity of the tumor epithelial cells staining with ERα and ERβ antibodies were evaluated. The staining percentage was calculated by counting the number of cells positively stained with antibodies in 100 cells at x10 optical magnification. For IHC staining, ER expression levels ≥1% were considered positive (+). The strength of staining was scored as follows: -, no staining, +, weak staining, ++, moderate staining and +++, strong staining (Fig. 2).

Statistical analysis. The 1:2 matching by PSM was performed considering the preoperative PSA level, International Society of Urological Pathology (ISUP) grade and pathological T stage (pT) as matching parameters. During PSM, ISUP grade groups were divided into three categories: ISUP grade group 1 was the first group, ISUP grade groups 2 and 3 were the second group and ISUP grade groups 4 and 5 were the third group. Categorical variables were analyzed using χ^2 and Fisher's exact tests. The Shapiro-Wilk test was used to test whether the quantitative data were normally distributed. The Mann-Whitney U test was used to compare the non-normally distributed quantitative data by group. Quantitative data are expressed as the median (range). Four groups were defined according to estrogen staining status: ERα(+)/ERβ(+), ERα(+)/ERβ(-), ERα(-)/ERβ(+) and ERα(-)/ERβ(-). Each group was compared with the other three groups. Univariate and multivariate logistic regression analyses were performed to determine the independent risk factors for BCR. Multivariate logistic regression analysis was performed with variables with P<0.25 in the univariate analysis. Survival analysis was performed using Kaplan-Meier and the log-rank test. P<0.05 was considered to indicate a statistically significant result. SPSS software (IBM SPSS Statistics for Windows, version 25.0; IBM Corp) was used for the analyses.

Results

Characteristics of the cohort. The clinicopathological characteristics of the entire study population are presented in Table I. The median age at diagnosis was 62.1 years in the BCR group and 63.9 years in the control group. ISUP Grade 2/3 patients comprised more than three-quarters of each group. Patients with pT3 cancer predominated in the two groups, and most patients had perineural invasion. The median follow-up time was 74.3 months (range, 30-127.5 months) in the BCR group and 66.6 months (range, 31.5-130 months) in the control group. No significant difference was found in patient characteristics between the two groups (P>0.05).

IHC staining. ERα staining was positive in 24 patients (66.7%) in the BCR group and 39 patients (54.1%) in the control group. ERβ staining was positive in 15 patients (41.7%) in the BCR group and 41 patients (56.9%) in the control group. Fig. 3 displays the percentages of patients with positive staining of the ER receptors. No difference was detected between the two groups in ERα staining status, ERα

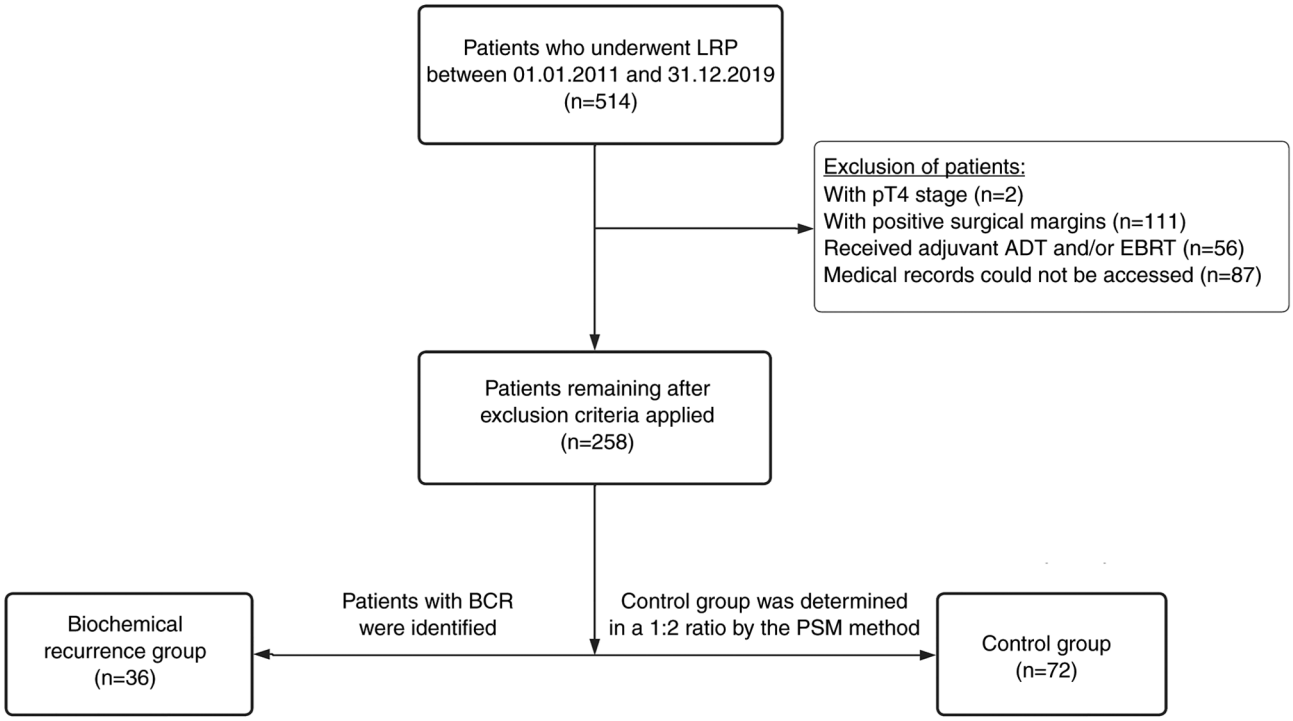

Figure 1. Study design chart. LRP, laparoscopic radical prostatectomy; PT4, pathological T stage 4; ADT, androgen deprivation therapy; EBRT, external beam radiotherapy; BCR, biochemical recurrence; PSM, Propensity Score Matching.

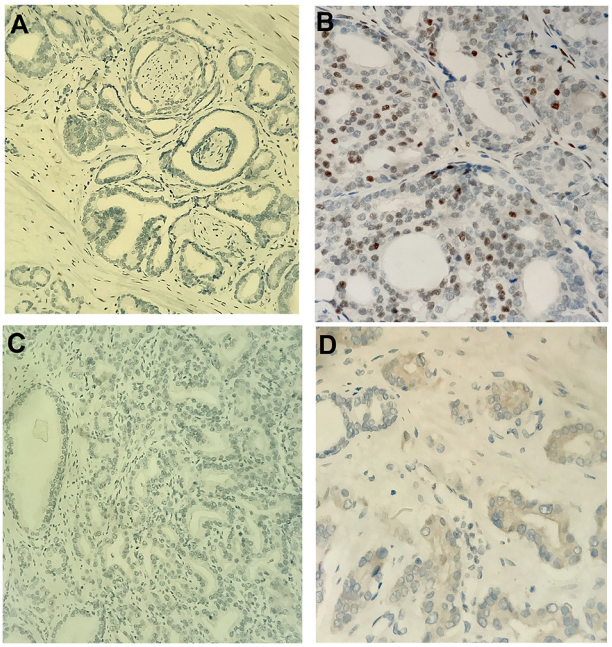

Figure 2. Microscopic view of immunohistochemical staining results. (A) ERα negative (magnification, x10), (B) ERα +++ (magnification, x40), (C) Erβ negative (magnification, x10) and (D) ERβ +++ (magnification, x40) staining. ERα, estrogen receptor α; ERβ, estrogen receptor β.

strength of staining, ERα staining percentage, ERβ staining status, ERβ strength of staining and ERβ staining percentage (P>0.05). Only patients in the ERα(-)/ERβ(+) group had significantly fewer BCRs among the four ERα/ERβ groups (P=0.024; Table II).

Outcomes. In the univariate logistic regression analysis to determine the risk factors for BCR, the P-values for ERα(+), ERβ(-) and the ERα(-)/ERβ(+) group were 0.216, 0.136 and 0.018, respectively (Table III). Multivariate logistic regression analysis revealed that only ERα(-)/ERβ(+) staining was an independent risk factor for BCR (P=0.048). The risk of BCR was 5.8-fold lower in this group (OR=5.840). The log-rank test revealed that the 5-year BCR-free survival (BFS) rate was significantly higher in the ERα(-)/ERβ(+) staining group than in other patients (85.7 vs. 66.1%; P=0.031; Fig. 4).

Discussion

The role of estrogen receptors in PCa has been studied extensively, but remains a matter of debate (19,20). Considering that PCa tumor cells may use pathways other than those associated with androgen receptors as resistance mechanisms in advanced disease (21), it may be reasonable to evaluate the role of estrogen receptors in early-stage patients. A limited number of studies have investigated the role of estrogen receptors in PCa in non-metastatic castration-naive disease, and their results are conflicting (15-17). The present study evaluated ERα and ERβ receptors using PSM analysis, in order to reduce the effect of well-known risk factors on BCR, and the results revealed that patients with ERα(-)/ERβ(+) PCa had a 5.8-fold lower risk of BCR than other patients and the 5-year BFS rate was significantly higher in patients with ERα(-)/ERβ(+) staining than in other patients (85.7 vs. 66.1%).

Megas *et al* (15) reported that ERα upregulation increased the risk of BCR 4.04-fold and low ERβ expression increased the risk of disease progression 6.59-fold. The authors also

Table I. Clinicopathological characteristics of all patients.

Characteristics	BCR group (n=36)	Control group (n=72)	P-value
Age, median (range)	62.1 (53.5-73.5)	63.9 (49.6-77.5)	0.391
BMI, median (range), kg/m^2	26.9 (22.1-35.6)	26.5 (20.2-40.1)	0.632
Preop PSA level, median (range), ng/ml	8.2 (3.1-40.0)	9.2 (4.1-23.0)	0.736
ISUP grade, n (%)			0.863[a]
1	5 (13.9)	11 (15.3)	
2,3	28 (77.8)	57 (79.1)	
4,5	3 (8.3)	4 (5.6)	
pT, n (%)			1.000[b]
pT2	6 (16.7)	13 (18.1)	
pT3	30 (83.3)	59 (81.9)	
PNI, n (%)			0.378[a]
Negative	3 (8.3)	11 (15.3)	
Positive	33 (91.7)	61 (84.7)	
LVI, n (%)			0.257[a]
Negative	34 (94.4)	71 (98.6)	
Positive	2 (5.6)	1 (1.4)	
Follow-up time, median (range)	74.3 (30-127.5)	66.6 (31.5-130)	0.160

[a]Fisher's exact test; [b]χ^2 test. BCR, biochemical recurrence; BMI, body mass index; kg, kilogram; m^2, square meter; preop, preoperative; PSA, prostate-specific antigen; ng, nanogram; ml, milliliter; ISUP, International Society of Urological Pathology; pT, pathological T stage; PNI, perineural invasion; LVI, lymphovascular invasion.

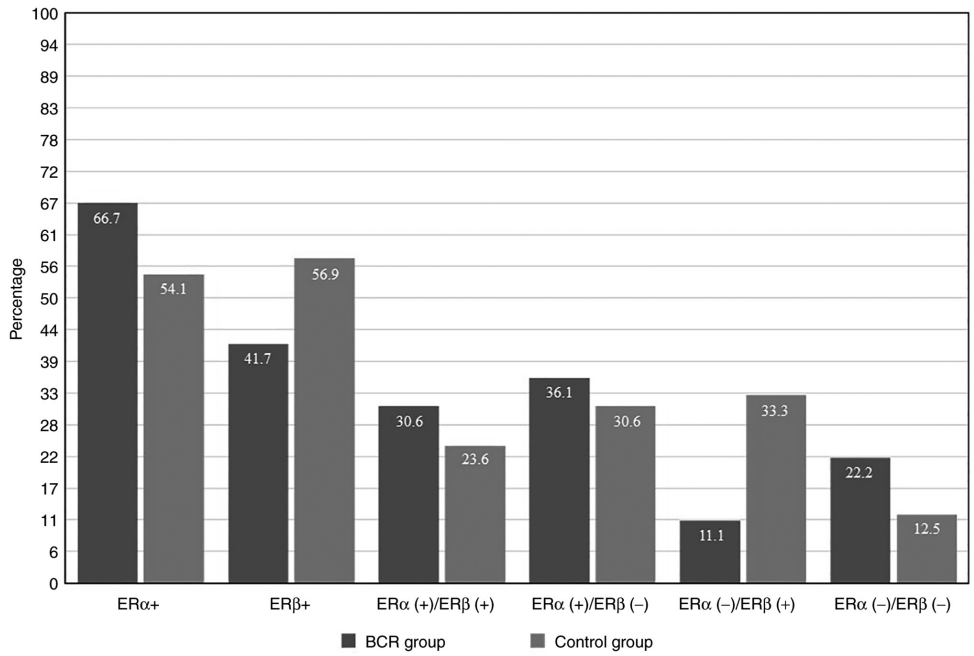

Figure 3. Percentages of patients with various ERα and ERβ staining results. ERα, estrogen receptor α; ERβ, estrogen receptor β; BCR, biochemical recurrence.

found that patients with concurrent ERα negative and ERβ positive staining had longer BFS than other patients, which is consistent with the present study. However, the study group in the previous study was heterogeneous, since 29% of patients received adjuvant ADT + EBRT and 33% received adjuvant ADT. Moreover, although patients with non-metastatic locally advanced PCa were included, the distribution of parameters such as preoperative PSA level and ISUP grades was not explicitly presented. Since PSM was performed in the present study, this is likely to have reduced the effect of these parameters on BCR and revealed the impact of ERα and ERβ more clearly. Although no significant differences regarding ERα and ERβ

Table II. Immunohistochemical staining results for ERα and ERβ.

Type of staining result	BCR group (n=36)	Control group (n=72)	P-value[a]
ERa staining, n (%)			0.301
Positive	24 (66.7)	39 (54.1)	
Negative	12 (33.3)	33 (45.9)	
ERa strength of staining, n (%)			0.098[b]
Negative	12 (33.3)	33 (45.9)	
Weak	20 (55.6)	23 (31.8)	
Moderate	4 (11.1)	15 (20.9)	
Strong	0 (0)	1 (1.4)	
ERa staining percentage, median (range),	1 (0-40)	1 (0-30)	0.107
ERb staining, n (%)			0.196
Positive	15 (41.7)	41 (56.9)	
Negative	21 (58.3)	31 (43.1)	
ERb strength of staining, n (%)			0.392[b]
Negative	21 (58.3)	31 (43.1)	
Weak	9 (25.0)	26 (36.1)	
Moderate	5 (13.9)	14 (19.4)	
Strong	1 (2.8)	1 (1.4)	
ERb staining percentage, median (range),	0 (0-20)	1 (0-60)	0.085
ERa/ERb staining groups, n (%)			
ERα(+)/ERβ(+)	11 (30.6)	17 (23.6)	0.587
Non-ERα(+)/ERβ(+)	25 (69.4)	55 (76.4)	
ERα(+)/ERβ(-)	13 (36.1)	22 (30.6)	0.716
Non-ERα(+)/ERβ(-)	23 (63.9)	50 (69.4)	
ERα(-)/ERβ(+)	4 (11.1)	24 (33.3)	0.024
Non-ERα(-)/ERβ(+)	32 (88.9)	48 (66.7)	
ERα(-)/ERβ(-)	8 (22.2)	9 (12.5)	0.304
Non-ERα(-)/ERβ(-)	28 (77.8)	63 (87.5)	

[a]Analyzed by χ^2 test unless otherwise indicated; [b]Fisher's exact test. BCR, biochemical recurrence; ERα, estrogen receptor α; ERβ, estrogen receptor β.

staining, the strength of staining and staining percentage were detected between the groups in the present study, significant differences were observed when combinations of ERα and ERβ results were considered; this may be due to the limited number of patients.

Horvath *et al* (17) reported that patients with ERβ-positive tumors had shorter survival times than those with ERβ-negative tumors, but ERα staining was not evaluated. In addition, the authors performed a multivariate analysis, which showed that ERβ expression, pT and GS were independent predictors of PCa prognosis while the preoperative PSA level and surgical margin positivity were not. Grindstad *et al* (16) analyzed ERα and ERβ staining in the normal stroma, tumoral stroma, normal epithelial cells and tumoral epithelial cells in 535 patients undergoing radical prostatectomy in a multicenter study. The authors reported that upregulation of ERα in the tumor stroma increased clinical progression-free survival (CPFS) and cancer-specific survival, and that the upregulation of ERβ in the tumor stroma reduced BFS (16). Unlike the present study, ER receptor staining was performed on the tumor stroma and epithelium, and no analysis of ERα-ERβ groups was performed. In addition, patients who had received adjuvant EBRT or ADT and had pelvic lymph node involvement and positive surgical margins were included in the study, although patients who received EBRT and ADT before surgery were excluded (16). Furthermore, patients in these two studies were not homogeneous in terms of preoperative PSA level, ISUP grade, pT, pelvic lymph node involvement and surgical margins. Pelvic lymph node involvement and surgical margin positivity are the most important independent risk factors for disease progression (22,23). Several studies have demonstrated that adjuvant ADT plus EBRT and adjuvant ADT monotherapy increase BFS, CPFS and overall survival (OS) (24-27). The inclusion of patients with these poor prognostic features and those who have received adjuvant therapy may have confounded the effect of ERα and ERβ on oncologic outcomes and could explain the differences in BFS, CPFS and OS. In the current study, this effect has been minimized by the exclusion of patients with positive surgical margins and lymph nodes after LRP and those who received adjuvant therapy.

Table III. Logistic regression analysis for predictors of biochemical recurrence.

	Univariate analysis				Multivariate analysis			
Factor	OR	P-value	95% CI lower	95% CI higher	OR	P-value	95% CI lower	95% CI higher
Preop PSA level, ng/ml	1.026	0.511	0.956	1.109	-	-	-	-
Age, years	0.972	0.401	0.908	1.039	-	-	-	-
BMI, kg/m^2	0.994	0.920	0.890	1.111	-	-	-	-
ISUP grade								
1	-	0.853	-	-	-	-	-	-
2,3	1.081	0.895	0.342	3.412	-	-	-	-
4,5	1.650	0.592	0.264	10.313	-	-	-	-
pT, T2 (R) vs. pT3	1.102	0.858	0.381	3.188	-	-	-	-
PNI, negative (R) vs. positive	1.984	0.318	0.517	7.614	-	-	-	-
LVI, negative (R) vs. positive	2.059	0.480	0.278	15.248	-	-	-	-
ERα, negative (R) vs. positive	1.692	0.216	0.735	3.895	0.665	0.495	0.206	2.149
ERβ, positive (R) vs. negative	1.852	0.136	0.823	4.164	0.913	0.862	0.329	2.539
ERα(-)/ER(+)β (R) vs. other groups	4.0	0.018	1.268	12.622	5.840	0.048	1.012	33.706
ERα(+)/ERβ (-) (R) vs. other groups	1.285	0.516	0.552	2.990	-	-	-	-

OR, odds ratio; preop, preoperative; PSA, prostate-specific antigen; ng, nanogram; ml, milliliter; BMI, body mass index; kg, kilogram; m^2, square meter; ISUP, International Society of Urological Pathology; pT, pathological T stage; R, reference category; PNI, perineural invasion; LVI, lymphovascular invasion; ERα, estrogen receptor α; ERβ, estrogen receptor β.

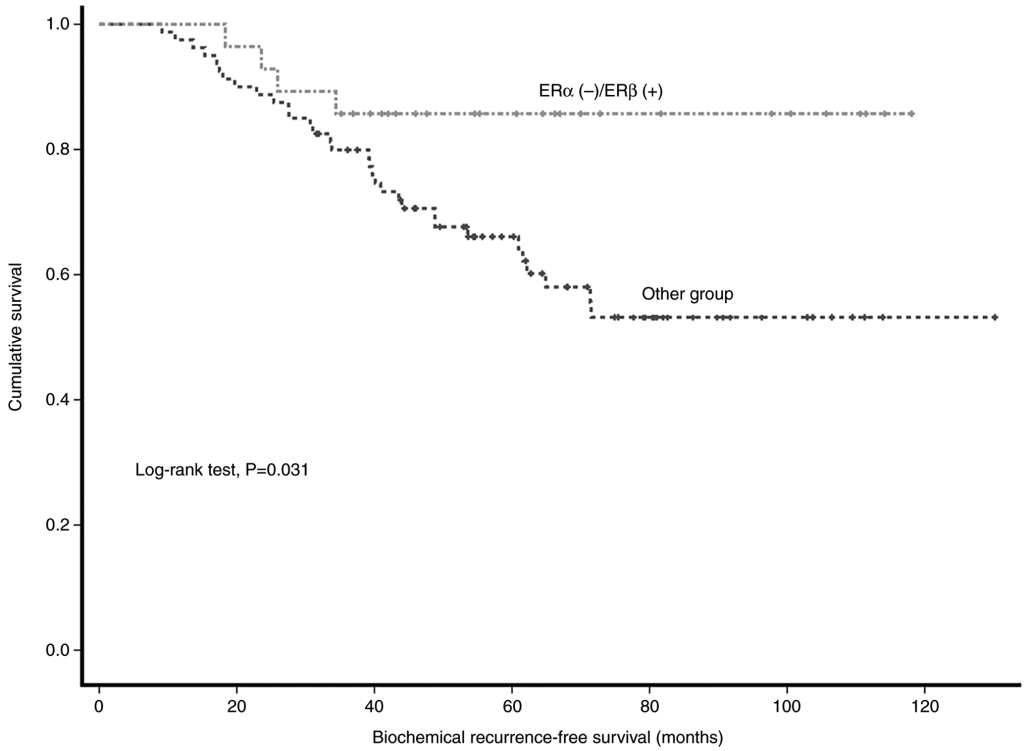

Figure 4. Kaplan-Meier survival curves according to ERα and ERβ status. ERα, estrogen receptor α; ERβ, estrogen receptor β.

ERα is most commonly localized in the prostatic stroma but is also found in the prostatic utricle and periurethral epithelium of the male reproductive system (28). Risbridger *et al* (7) and Prins *et al* (8) showed that ERα plays a role in the development of prostatic stromal hyperplasia, inflammatory cell infiltration, squamous metaplasia and PIN in studies with ERα and ERβ knockout mice, and Bonkhoff *et al* (9) indicated that ERα expression increases in luminal cells during the malignant transformation from high-grade PIN to PCa. ERβ is expressed in human prostate tissue, specifically in stromal and epithelial cells (29). Cheng *et al* (12) induced ERβ-negative PCa cells to express ERβ using an ERβ-encoding adenoviral vector and showed that ERβ inhibited the growth and invasion of the cells and increased their apoptosis. It has been suggested that the anti-proliferative effect of ERβ is achieved via the prevention of androgenic stimulation (30). Based on the findings of these studies, it can be concluded that ERα plays a role in the pathophysiological processes of chronic prostatitis, BPH and cancer development, and ERβ has anti-proliferative, anti-invasive, anti-inflammatory and pro-apoptotic effects.

The application of adjuvant treatment modalities or early salvage strategies has been a subject of debate in patients with a high risk of recurrence, such as those with a positive surgical margin and ≥pT3 cancer after radical prostatectomy (31). In this context, the use of estrogen receptor status in combination with other well-studied clinicopathological prognostic markers may help clinicians to select patients with a poor prognosis for adjuvant therapy.

The strengths of the present study are the pertinent exclusion-inclusion criteria that were applied and the homogeneous control group determined by the PSM method considering well-known risk factors for BCR. The retrospective design of the study, the lack of CPFS and radiological recurrence-free survival data and the limited number of patients due to the strict inclusion-exclusion criteria are limitations of the study. In addition, an OS analysis could not be performed because there were insufficient PCa-associated deaths.

In conclusion, estrogen receptors may have prognostic value for non-metastatic PCa. Moreover, the 5-year BFS rate of patients with ERα(-)/ERβ(+) stained PCa is significantly improved compared with that of other patients.

Acknowledgements

The authors would like to thank Ms. Züleyha Sarıkaya, Ms. Nihan Genç, Ms. Fatma Aydın Yazıcı, Ms. Zeliha Altun, Ms. Elif Bayazit, Ms. Pelin Bilir and Ms. Şerife Kirez (Department of Pathology, Bursa Uludag University) for their technical support in this study.

Funding

The Institution for Scientific Research Projects of Bursa Uludag University financially supported this study (project no. TTU-2021-420).

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

YMA was responsible for investigation, data curation, writing the original draft of the manuscript and visualization (designing tables and figures). ABS was responsible for conceptualization, methodology and for writing, reviewing and editing the manuscript. BAV and RD performed the histopathological examination and revised the manuscript critically for important intellectual content. GO was responsible for formal analysis and methodology. HV and IY provided substantial contributions to the design of the study, the interpretation of data, drafting the work and providing final approval of the version to be published. BC was responsible for substantial contributions to the design of the study, reviewing and editing the manuscript, as well as drafting the work and final approval of the version to be published. All authors read and approved the final version of the manuscript. YMA and BC confirm the authenticity of all the raw data.

Ethics approval and consent to participate

The Institutional Review Board of Bursa Uludag University approved the study (approval number: 2021-2/15). The study protocol complied with the tenets of the Declaration of Helsinki. The Clinical Research Ethics Committee of The Bursa Uludag University Faculty of Medicine waived informed consent due to the retrospective nature of the study.

Patient consent for publication

Not applicable.

Authors' information

The ORCID iD of Dr Yavuz Mert Aydın is 0000-0002-6287-6767. The ORCID iD of Ahmet Bilgehan Sahin is 0000-0002-7846-0870. The ORCID iD of Rabia Dolek is 0000-0002-1751-7693. The ORCID iD of Berna Aytac Vuruskan is 0000-0001-9549-8435. The ORCID iD of Gokhan Ocakoglu is 0000-0002-1114-6051. The ORCID iD of Hakan Vuruskan is 0000-0002-3917-4847. The ORCID iD of İsmet Yavascaoglu is 0000-0002-1788-1997. The ORCID iD of Burhan Coskun is 0000-0001-7206-6648.

Competing interests

The authors declare that they have no competing interests.

References

1. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A and Bray F: Global cancer statistics 2020: GLOBOCAN estimates of ıncidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 71: 209-249, 2021.
2. Bostwick DG, Burke HB, Djakiew D, Euling S, Ho SM, Landolph J, Morrison H, Sonawane B, Shifflett T, Waters DJ and Timms B: Human prostate cancer risk factors. Cancer 101 (10 Suppl): S2371-S2490, 2004.
3. Dobbs RW, Malhotra NR, Greenwald DT, Wang AY, Prins GS and Abern MR: Estrogens and prostate cancer. Prostate Cancer Prostatic Dis 22: 185-194, 2019.
4. Setlur SR, Mertz KD, Hoshida Y, Demichelis F, Lupien M, Perner S, Sboner A, Pawitan Y, Andrén O, Johnson LA, *et al*: Estrogen-dependent signaling in a molecularly distinct subclass of aggressive prostate cancer. J Natl Cancer Inst 100: 815-825, 2008.

5. McPherson SJ, Hussain S, Balanathan P, Hedwards SL, Niranjan B, Grant M, Chandrasiri UP, Toivanen R, Wang Y, Taylor RA and Risbridger GP: Estrogen receptor-beta activated apoptosis in benign hyperplasia and cancer of the prostate is androgen independent and TNFalpha mediated. Proc Natl Acad Sci USA 107: 3123-3128, 2010.
6. Mak P, Leav I, Pursell B, Bae D, Yang X, Taglienti CA, Gouvin LM, Sharma VM and Mercurio AM: ERβ ımpedes prostate cancer EMT by destabilizing HIF-1α and inhibiting VEGF-mediated snail nuclear localization: Implications for gleason grading. Cancer Cell 17: 319-332, 2010.
7. Risbridger G, Wang H, Young P, Kurita T, Wang YZ, Lubahn D, Gustafsson JA and Cunha G: Evidence that epithelial and mesenchymal estrogen receptor-α mediates effects of estrogen on prostatic epithelium. Dev Biol 229: 432-442, 2001.
8. Prins GS, Birch L, Couse JF, Choi I, Katzenellenbogen B and Korach KS: Estrogen imprinting of the developing prostate gland is mediated through stromal estrogen receptor alpha: Studies with alphaERKO and betaERKO mice. Cancer Res 61: 6089-6097, 2001.
9. Bonkhoff H, Fixemer T, Hunsicker I and Remberger K: Estrogen receptor expression in prostate cancer and premalignant prostatic lesions. Am J Pathol 155: 641-647, 1999.
10. Zhu X, Leav I, Leung YK, Wu M, Liu Q, Gao Y, McNeal JE and Ho SM: Dynamic regulation of estrogen receptor-beta expression by DNA methylation during prostate cancer development and metastasis. Am J Pathol 164: 2003-2012, 2004.
11. Zhang X, Leung YK and Ho SM: AP-2 regulates the transcription of estrogen receptor (ER)-beta by acting through a methylation hotspot of the 0N promoter in prostate cancer cells. Oncogene 26: 7346-7354, 2007.
12. Cheng J, Lee EJ, Madison LD and Lazennec G: Expression of estrogen receptor beta in prostate carcinoma cells inhibits invasion and proliferation and triggers apoptosis. FEBS Lett 566: 169-172, 2004.
13. Weihua Z, Warner M and Gustafsson JA: Estrogen receptor beta in the prostate. Mol Cell Endocrinol 193: 1-5, 2002.
14. McPherson SJ, Ellem SJ, Simpson ER, Patchev V, Fritzemeier KH and Risbridger GP: Essential role for estrogen receptor beta in stromal-epithelial regulation of prostatic hyperplasia. Endocrinology 148: 566-574, 2007.
15. Megas G, Chrisofos M, Anastasiou I, Tsitlidou A, Choreftaki T and Deliveliotis C: Estrogen receptor (α and β) but not androgen receptor expression is correlated with recurrence, progression and survival in post prostatectomy T3N0M0 locally advanced prostate cancer in an urban Greek population. Asian J Androl 17: 98-105, 2015.
16. Grindstad T, Skjefstad K, Andersen S, Ness N, Nordby Y, Al-Saad S, Fismen S, Donnem T, Khanehkenari MR, Busund LT, *et al*: Estrogen receptors α and β and aromatase as independent predictors for prostate cancer outcome. Sci Rep 6: 33114, 2016.
17. Horvath LG, Henshall SM, Lee CS, Head DR, Quinn DI, Makela S, Delprado W, Golovsky D, Brenner PC, O'Neill G, *et al*: Frequent loss of estrogen receptor-β expression in prostate cancer. Cancer Res 61: 5331-5335, 2001.
18. Nelson JB and Lepor H: Prostate cancer: Radical prostatectomy. Urol Clin North Am 30: 703-723, 2003.
19. Bonkhoff H: Estrogen receptor signaling in prostate cancer: Implications for carcinogenesis and tumor progression. Prostate 78: 2-10, 2018.
20. Qu LG, Wardan H, Davis ID, Pezaro C and Sluka P: Effects of estrogen receptor signaling on prostate cancer carcinogenesis. Transl Res 222: 56-66, 2020.
21. He Y, Xu W, Xiao YT, Huang H, Gu D and Ren S: Targeting signaling pathways in prostate cancer: Mechanisms and clinical trials. Signal Transduct Target Ther 7: 198, 2022.
22. Mottet N, Vice-Chair PC, Bergh RCN, Van Den, Mottet N, *et al*: Guidelines on Prostate Cancer. Update 53: 31-45, 2021.
23. Briganti A, Karnes JR, Da Pozzo LF, Cozzarini C, Gallina A, Suardi N, Bianchi M, Freschi M, Doglioni C, Fazio F, *et al*: Two positive nodes represent a significant Cut-off value for cancer specific survival in patients with node positive prostate cancer. A new proposal based on a Two-ınstitution experience on 703 consecutive N+ patients treated with radical prostatectomy, extended pelvic lymph node dissection and adjuvant therapy. Eur Urol 55: 261-270, 2009.
24. Thompson IM, Tangen CM, Paradelo J, Lucia MS, Miller G, Troyer D, Messing E, Forman J, Chin J, Swanson G, *et al*: Adjuvant radiotherapy for pathological T3N0M0 prostate cancer significantly reduces risk of metastases and ımproves survival: Long-term followup of a randomized clinical trial. J Urol 181: 956-962, 2009.
25. Bolla M, Van Poppel H, Tombal B, Vekemans K, Da Pozzo L, de Reijke TM, Verbaeys A, Bosset JF, van Velthoven R, Colombel M, *et al*: Postoperative radiotherapy after radical prostatectomy for high-risk prostate cancer: Long-term results of a randomised controlled trial (EORTC trial 22911). Lancet 380: 2018-2027, 2012.
26. Wiegel T, Bartkowiak D, Bottke D, Bronner C, Steiner U, Siegmann A, Golz R, Störkel S, Willich N, Semjonow A, *et al*: Adjuvant radiotherapy versus wait-and-see after radical prostatectomy: 10-year follow-up of the ARO 96-02/AUO AP 09/95 trial. Eur Urol 66: 243-250, 2014.
27. Hackman G, Taari K, Tammela TL, Matikainen M, Kouri M, Joensuu T, Luukkaala T, Salonen A, Isotalo T, Pétas A, *et al*: Randomised trial of adjuvant radiotherapy following radical prostatectomy versus radical prostatectomy alone in prostate cancer patients with positive margins or extracapsular extension. Eur Urol 76: 586-595, 2019.
28. Shapiro E, Huang H, Masch RJ, McFadden DE, Wilson EL and Wu XR: Immunolocalization of estrogen receptor α and β in human fetal prostate. J Urol 174: 2051-2053, 2005.
29. Prins GS and Korach KS: The role of estrogens and estrogen receptors in normal prostate growth and disease. Steroids 73: 233-244, 2008.
30. Weihua Z, Makela S, Andersson LC, Salmi S, Saji S, Marketon JW, Jensen EV, Nilsson S, Warner M and Gustafsson JA: A role for estrogen receptor? in the regulation of the ventral prostate. Proce National Acad Sci 98: 6330-6335, 2001.
31. Terlizzi M, Limkin EJ, Moukasse Y and Blanchard P: Adjuvant or salvage radiation therapy for prostate cancer after prostatectomy: Current status, controversies and perspectives. Cancers (Basel) 14: 1688, 2022.