

# Low Preoperative Prolactin Levels Predict Non-Organ Confined Prostate Cancer in Clinically Localized Disease

Antonio Benito Porcaro<sup>a</sup> Alessandro Tafuri<sup>a</sup> Marco Sebben<sup>a</sup>  
Giovanni Cacciamani<sup>a</sup> Claudio Ghimenton<sup>b</sup> Matteo Brunelli<sup>b</sup>  
Aldo Petrozziello<sup>c</sup> Carmelo Monaco<sup>a</sup> Filippo Migliorini<sup>a</sup>  
Salvatore Siracusano<sup>a</sup> Walter Artibani<sup>a</sup>

<sup>a</sup>Department of Urology, University of Verona, Azienda Ospedaliera Universitaria Integrata Verona, Verona, Italy;

<sup>b</sup>Department of Pathology, University of Verona, Azienda Ospedaliera Universitaria Integrata Verona, Verona, Italy;

<sup>c</sup>Department of Internal Medicine, Endocrinology Section, Azienda Ospedaliera Universitaria Integrata Verona, Verona, Italy

## Keywords

Prostate cancer · Prolactin · Radical prostatectomy · Pituitary hormones · Hypothalamus-pituitary-testis-prostate axis · Preoperative serum prolactin levels · Preoperative serum testosterone levels · Prostate cancer induction · Prostate cancer progression · New prostate cancer therapy targets

## Abstract

**Introduction:** To evaluate the association between preoperative serum prolactin (PRL) levels and risk of non-organ confined prostate cancer (PCa) in clinically localized disease.

**Materials and Methods:** From December 2007 to December 2011, 124 patients with clinically localized PCa were retrospectively evaluated. Non-organ confined disease in the surgical specimen was defined according to extra-capsular extension, seminal vesicle invasion, positive surgical margins, and lymph node invasion. The association between clinical factors and serum levels of pituitary-testis hormones with the risk of non-organ confined disease was evaluated. **Results:** Perioperative factors associated with non-organ con-

fined disease include prostatic-specific antigen (OR 1.144;  $p = 0.025$ ), proportion of biopsy positive cores (BPC, OR 36.702;  $p = 0.007$ ), bioptical Gleason Score  $>6$  (OR 2.785;  $p = 0.034$ ), and PRL (OR 0.756,  $p < 0.0001$ ). The association was strong for BPC (area under the curve [AUC] 0.704;  $p < 0.0001$ ) and PRL (AUC 0.299;  $p < 0.0001$ ). When we dichotomized according to median value, PRL  $\leq 7.7$   $\mu\text{g/L}$  was an independent predictor of extraprostatic disease (OR 6.571;  $p < 0.0001$ ) with fair discrimination power (AUC 0.704;  $p < 0.0001$ ). **Conclusion:** Low preoperative PRL levels predict the risk of non-organ confined PCa in clinically localized disease.

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## Introduction

Prolactin (PRL) is a hormone secreted by pituitary, which in physiological conditions is also produced in many other tissues [1]. The PRL actions are completed on

A.B.P. and A.T. contributed equally to this manuscript.

target tissues via the circulating route and mediated by an unique transmembrane PRL-receptor (PRL-R) that is expressed in most tissues [2]. The PRL-R signaling pathway involves the signal transducer and activator of transcription-5 (STAT5) [3], and it is essential for adaptive functions related to reproduction, mammary gland differentiation, mammapoiesis, and maternal behavior or beta-cell proliferation during pregnancy. In the prostate, the Jak2/STAT5 cascade seems to be the only pathway activated by PRL that promotes secretory functions of luminal cells. The involvement of PRL in hormone-dependent tumors such as breast and prostate cancers (PCa) has been suggested in the late 60s–70s based on experimental studies involving animal models [4]. Subsequently, immunohistochemical studies of PRL expression in human PCa specimens showed that PRL was present in more than 50% of the tumors (localized, recurrent/hormone-refractory) and was associated with a high Gleason score [5, 6]. In addition, the PRL-R gene (5p13.2) is located in a frequently amplified region in the PCa cells oncogene (5p13.1-p13.3 locus) and significantly associated with negative oncological outcomes [7]. However, the role of PRL in the pathogenesis of PCa remains unclear and controversial. In fact, some studies found that high expression of STAT5 in prostate tumors was associated with high-grade and worse prognosis disease [6–8]. On the contrary, Agarwal et al. [9] reported negative results from a phase I trial of LFA102, a humanized monoclonal antibody that binds to and inhibits PRL-R signaling, in patients with PRLR-positive metastatic breast cancer or metastatic castration-resistant PCa. Clinical studies investigating associations between preoperative PRL and PCa biology are missing. We have shown that low preoperative PRL levels are associated with aggressive PCA in terms of estimated percentage of biopsy positive cores (BPC), bioptical Gleason Score (bGS), and tumor volume [10, 11].

The aim of the present study was to evaluate associations between preoperative PRL levels and non-organ confined PCa in Caucasian patients with clinically localized disease.

## Patients and Methods

The present retrospective analysis is part of a study aimed to evaluate a potential link between PCa and hypothalamus-pituitary-testis-prostate axis. Collection and use of data for analysis had the approval of the Institutional Review Board. The study included 220 patients who underwent radical prostatectomy with or without limited lymph node dissection from December 2007 to December 2011. All procedures were performed by expert sur-

geons. Exclusion criteria were as follows: (i) treatment with 5 $\alpha$ -reductase inhibitors, luteinizing hormones (LH)-releasing hormone analogs, testosterone replacement or (ii) previous radiation of the pelvis.

The present analysis relates to 124 Caucasian patients with clinically localized PCa, simultaneously investigated for basal serum levels of PRL, follicle-stimulating hormones (FSH), LH, total testosterone (TT), and prostatic-specific antigen (PSA). After signed informed consent, preoperative simultaneous serum samples of PRL, FSH, TT, and PSA were obtained from a cubital vein, at least 1 month after trans-rectal ultrasound biopsy. The samples were performed between 08:00 and 08:30 and analyzed in our laboratory. PRL (normal range 3.07–20.05  $\mu$ g/L), FSH (normal range 1.0–14.0 IU/L), LH (normal range 2.1–10.0 UI/L), TT (normal range 9–29 nmol/L), and PSA (normal range 2–4  $\mu$ g/L) were measured by using immune-chemiluminescent tests carried out by the ADVIA Centaur XP Immunoassay System (Siemens Company). The 14 cores trans-rectal ultrasound scan guided prostate biopsy technique was routinely performed. bGS was evaluated after computing primary and secondary Gleason patterns. Furthermore, proportion of BPC was assessed.

Surgical specimens were fixed overnight (in 10% neutral buffered formaldehyde), coated with India ink and then weighted. Tissue sections of 4  $\mu$ m were prepared in the standard fashion and stained with hematoxylin and eosin [12]. Patients were classified according to the primary tumor stage, lymph nodes, and metastatic status, using the TNM categories recommended by the 2010 International Union Against Cancer TNM classification system [13]. Tumors were graded according to the pathological Gleason grading score system (pGS) after computing the primary and secondary Gleason patterns.

### Statistical Analysis

Patients were clustered in 2 groups, according to the presence or absence of organ confined PCa. In the surgical specimen, criteria defining non-organ confined disease included at least one of the following pathological features: extracapsular extension, seminal vesicle invasion, positive surgical margins, and lymph node invasion. The evaluated factors included age, PSA, FSH, LH, PRL, BPC, bGS, prostate weight (PW), and pGS.

Data on continuous variables are presented as medians with interquartile range. Differences between groups were analyzed with the Mann-Whitney U test. Data on categorical variables are presented as frequencies (percentages) and differences between groups analyzed with Pearson's chi-square or Fishers' exact test as appropriate. The association of significant factors with the risk of non-organ confined disease was assessed by the multivariate binomial logistic regression model. The receiver-operating characteristic (ROC) curve and area under the curve (AUC) for independent factors were also evaluated. The software used to run the analysis was IBM-SPSS version 20. All tests were 2-sided, with a significance level of  $p < 0.05$ .

## Results

The study included 124 Caucasian patients. Clinical and pathological features of the population and subpopulation of patients are reported in Table 1. As shown,

**Table 1.** Distribution and association of factors in the patient population stratified PCa extension in the surgical specimen in clinically localized disease

Factors	Population	PCa extension in the surgical specimen		<i>p</i> value
		organ confined	non-organ confined	
Cases, <i>n</i> (%)	124	54 (43.6)	70 (56.4)	
<i>Clinical</i>				
Age, years, median (IQR)	66.5 (60.7–70.5)	66.6 (59.8–70)	66.4 (61.3–71.2)	0.389
PSA, ng/mL, median (IQR)	5.7 (4.11–8.7)	5.3 (3.6–7.1)	6.4 (4.7–10.6)	0.003
TT, nmol/L, median (IQR)	15.1 (11.6–20.5)	15.6 (11.7–20)	15.1 (11.4–20.6)	0.811
FSH, IU/L, median (IQR)	6.3 (3.8–9.1)	6.5 (4–9)	6 (3.7–9.7)	0.739
LH, IU/L, median (IQR)	4.2 (3.1–6.1)	4.1 (3.1–5.9)	4.3 (2.9–6.6)	0.427
PRL, ng/L, median (IQR)	7.7 (5.6–9.2)	8.5 (7.3–10.8)	6.4 (5.2–8.06)	<0.0001
BPC, proportion, median (IQR)	0.31 (0.14–0.50)	0.21 (0.14–0.31)	0.42 (0.21–0.50)	<0.0001
cT, <i>n</i> (%)			33 (47.1)	0.017
1c	70 (56.5)	37 (68.5)		
2	54 (43.5)	17 (31.5)	37 (52.9)	
bGS, <i>n</i> (%)				0.003
6	72 (58.19)	40 (74.1)	32 (45.8)	
7	47 (37.9)	14 (25.9)	33 (47.19)	
8	5 (4)	0 (0.0)	5 (7.1)	
<i>Pathological</i>				
PW, g, median (IQR)	50 (42.5–71.4)	59.1 (45.5–80.6)	47 (39.2–57.7)	0.002
pGS, <i>n</i> (%)				0.003
6	38 (30.6)	25 (46.3)	13 (18.5)	
7	72 (58.1)	28 (51.9)	44 (63)	
>7	14 (11.3)	1 (1.9)	13 (18.5)	
pT, <i>n</i> (%)				<0.0001
2	65 (52.4)	54	11 (15.7)	
3a	50 (40.3)	0	50 (71.4)	
3b	9 (7.3)	0	9 (12.9)	
Surgical margin, <i>n</i> (%)				<0.0001
Negative	70 (56.5)	54	16 (22.9)	
Positive	54 (43.5)	0	54 (77.1)	
pN, <i>n</i> (%)				0.17
X	102 (82.3)	48 (88.9)	54 (77.1)	
0	20 (16.1)	6 (11.1)	14 (20.0)	
1	2 (1.6)	0 (0.0)	2 (2.9)	

PSA, prostatic specific antigen; bGS, bioptic Gleason Score; PW, prostate weight; pGS, pathological Gleason Score; IQR, interquartile range; PCa, prostate cancer.

the median age was 66.5 years with a median PSA of 5.7 ng/mL and median proportion of BPC of 0.31. Preoperative hormonal levels along the pituitary-testis-prostate axis are also described. All patients had clinically localized disease which was staged T1c in 70 (56.5%) and T2 in 54 (43.5%) subjects. A bGS >6 was present in 52 (38.3%) cases. Evaluation of the surgical specimen showed that the median PW was 50 g and 86 cases (69.4%) had a Gleason score >6; moreover, in 59 subjects the specimen evaluation showed extraprostatic disease including extracapsular extension in 50 cases

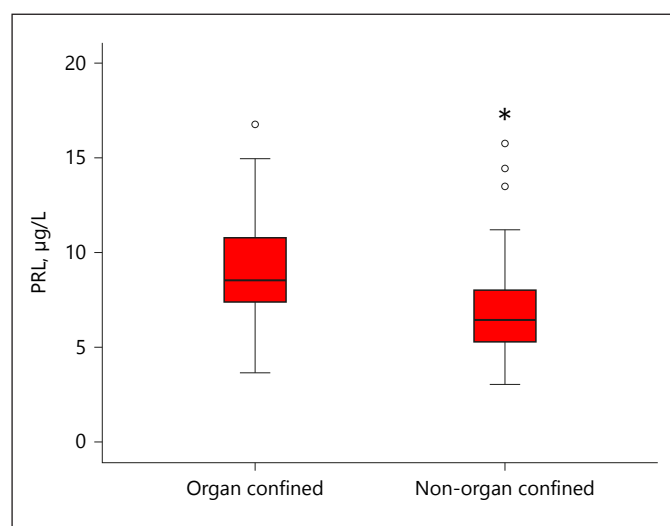
(40.3%) and seminal vesicle invasion in 9 patients (7.3%). Positive surgical margins were detected in 54 (43.5%) subjects, and the nodal status was unknown, negative, and positive in 102 (82.3%), 14 (20.0%), and 2 (2.9%) patients respectively.

Overall, extraprostatic cancer was detected in 70 (56.4%) cases. Patients with non-organ confined PCa, when compared to cases with intraprostatic disease, had higher median values of PSA (6.4 vs. 5.3 ng/mL;  $p = 0.03$ ), BPC (0.42 vs. 0.21;  $p < 0.0001$ ), lower PRL levels (6.4 vs. 8.5 ng/L;  $p < 0.0001$ ; Fig. 1), and higher rates of

**Table 2.** Clinical factors associated with the risk of non-organ confined PCa in clinically localized disease

	Multivariate model I			Multivariate model II (adjusted OR)		
	OR	95% CI	<i>p</i> value	OR	95% CI	<i>p</i> value
PSA	1.138	1.014–1.276	0.028	1.144	1.020–1.283	0.022
PRL	0.525	0.649–0.882	<0.0001	0.756	0.648–0.881	<0.0001
BPC	28.304	1.936–413.712	0.015	36.702	2.680–502.594	0.007
cT1c	Ref					
cT2	1.629	0.666–1.619	ns			
bGS = 6	Ref			Ref		
bGS >6	2.765	1.067–7.170	0.036	2.785	1.082–7.167	0.034

PSA, prostatic specific antigen; PRL, prolactin; BPC, bioptic positive cores; bGS, bioptic Gleason Score; PCa, prostate cancers.



**Fig. 1.** PCa extension in the surgical specimen in clinically localized disease. PRL, prolactin. \* Patients with non-organ confined PCa, when compared to cases with intraprostatic disease, had lower PRL levels (6.4 vs. 8.5 ng/L;  $p < 0.0001$ ).

cT2 (52.9 vs. 31.5%;  $p = 0.017$ ) as well as of bGS >6 (54.2 vs. 25.9%;  $p = 0.003$ ). Moreover, differences between 2 groups were also detected for median values of PW (47 vs. 58.1 g;  $p = 0.002$ ) and rates of high-grade PCa (18.5 vs. 1.9%;  $p = 0.003$ ) in the surgical specimen. We did not find significant differences in univariate analysis in TT medians evaluation between organ confined and non-organ confined PCa groups (15.6 vs. 15.1 nmol/L,  $p = 0.811$ ).

In multivariate analysis (Table 2), clinical factors associated with the risk of non-organ confined disease were PSA (OR 1.144;  $p = 0.025$ ), BPC (OR 36.702;  $p = 0.007$ ),

bGS >6 (OR 2.785;  $p = 0.034$ ), and PRL (OR 0.756,  $p < 0.0001$ ). As shown in Table 3, independent pathological factors associated with the risk of non-organ confined PCa included pGS = 7 (OR 2.530;  $p = 0.033$ ) and pGS >7 (OR 19.672;  $p = 0.007$ ); moreover, considering a combined model including clinical and pathological factors, PSA, BPC, and PRL remained independent predictors together with pGS = 7 and pGS >7 (see multivariate model II with adjusted ORs in Table 3).

Receiver-operating characteristic curves and AUC of clinical predictors of the risk of non-organ confined PCa are depicted in Figure 1 and Table 4. The association of clinical factors with the risk of non-organ confined PCa was positive for PSA, BPC, and bGS >6 but inverse for PRL; moreover, the discrimination power was poor for PSA (AUC 0.655;  $p < 0.003$ ) and bGS >6 (AUC 0.642;  $p = 0.050$ ) but fair for both BPC (AUC 0.704;  $p < 0.0001$ ) and PRL (AUC 0.299;  $p < 0.0001$ ; Figure 2)

Because of the fair discrimination power of PRL in predicting specimen non-organ confined PCa, we dichotomized PRL according to its median values (PRL  $\leq 7.7$  µg/L vs. PRL  $\geq 7.7$  µg/L). On multivariate analysis, PRL  $\leq 7.7$  µg/L was an independent predictor of PCa extraprostatic extension (OR 6.571;  $p < 0.0001$ ) together with PSA (OR 1.123;  $p = 0.035$ ) and BPC (OR 63.381;  $p < 0.001$ ) but bGS >6 lost prediction power (Table 5). In this model, PRL  $\leq 7.7$  µg/L acquired fair discrimination power in predicting non-organ confined disease (AUC 0.704; 95% CI of AUC 0.611–0.798;  $p < 0.0001$ ), as shown in Figure 3. Moreover, interaction of PRL  $\leq 7.7$  µg/L with PSA (AUC 0.715; 95% CI 0.625–0.806;  $p < 0.0001$ ) and BPC (AUC 0.754; 95% CI 0.670–0.839;  $p < 0.0001$ ) increased the discrimination power of both PSA and BPC alone (Table 4).

**Table 3.** Clinical and pathological factors associated with the risk of non-organ confined PCa in clinically localized disease

	Multivariate model I*			Multivariate model II (adjusted OR)**		
	OR	95% CI	<i>p</i> value	OR	95% CI	<i>p</i> value
PSA				1.145	1.017–1.289	0.025
PRL				0.733	0.621–0.866	<0.0001
BPC				28.916	2.076–402.832	0.012
PW	0.99	0.597–1.004	0.152			
pGS						
6	Ref			Ref		
7	2.53	1.077–5.944	0.033	2.9	1.065–7.898	0.037
>7	19.672	2.261–171.181	0.007	30.251	2.533–361.345	0.007

\* Model including only significant pathological factors.

\*\* Model including both clinical and pathological factors.

PSA, prostatic specific antigen; PRL, plasmatic prolactin; PW, prostate weight; pGS, pathological Gleason Score; PCa, prostate cancer.

**Table 4.** Discrimination power of clinical factors associated with the risk of non-organ confined PCa in clinically localized disease

	AUC	95% CI	<i>p</i> value	Discrimination	Discrimination power
PSA	0.655	0.559–0.751	0.003	Positive	Poor
PRL	0.299	0.204–0.395	<0.0001	Negative	Fair
BPC	0.704	0.611–0.798	<0.0001	Positive	Fair
bGS >6	0.642	0.544–0.740	0.05	Positive	Poor
PRL × PSA*	0.715	0.625–0.806	<0.0001	Positive	Fair
PRL × BPC*	0.754	0.670–0.839	<0.0001	Positive	Fair

\* PRL ≤7.7, AUC, ROC curve (see also Figure 2 and Figure 3).

PSA, prostatic specific antigen; PRL, plasmatic prolactin; BPC, biopsy positive cores; bGS, bioptic Gleason Score; AUC, area under curve; ROC, receiver operating characteristic; PCa, prostate cancer.

## Discussion

The endocrine influence in PCa pathogenesis is well known, but the involvement of PRL, PRL-R, and STAT 5 is debated and unclear [4]. Although epidemiological study showed that PCa risk did not correlate with circulating PRL levels [14], it is known that PRL and its receptor are both expressed in the healthy human prostate [15]. Besides, it has been shown by immunohistochemical analysis that PRL is expressed in 54% of the general population of PCa patients, and the highest expression of PRL is observed in Gleason scores 7, 8, and 9 [6]. Also, Dagvadorj et al. [5] showed that PRL protein is expressed in 54% of patients with hormonal refractory PCa and in 62% of patients with metastatic PCa, thus suggesting PRL stimulation by auto- or paracrine mechanisms in prostate microenvironment. This hypothesis has been confirmed in the animal model, where the overexpression of

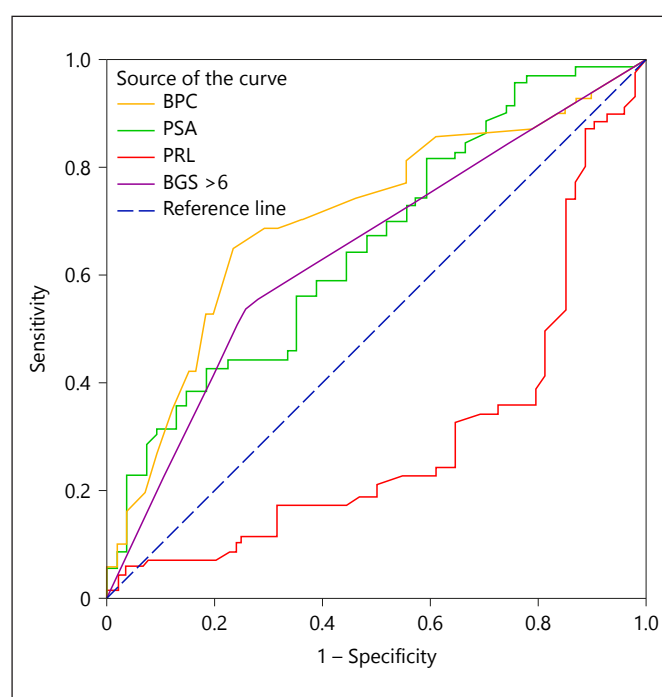
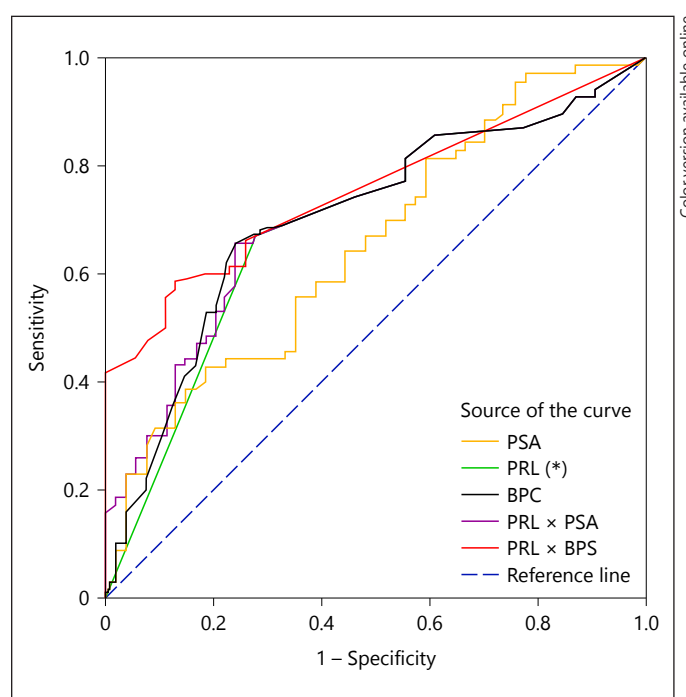
PRL in prostate epithelium caused hypertrophy and pre-malignant epithelial lesions, prostate intraepithelial neoplasia or PIN [5, 16]. In addition, other immunohistochemical studies demonstrated that PRL-R is overexpressed in more aggressive human PCa [17] and its mediator, STAT 5 (that is also one of androgen receptor [AR] mediators) is more expressed in AR-independent PCa. Furthermore, STAT5 stimulates the metastatic potency of AR-independent PCa and conferred stem-like properties to PCa cells [4]. Nevertheless, clinical studies investigating associations of PRL with PCa biology and carcinogenesis are missing and the topic is considered controversial because it is unexplored. In a dated study, Chen et al. [18] investigating the hormonal levels of the endocrine axis in metastatic PCa patients undergoing androgen deprivation, found that PRL levels were significantly lower in high-grade cancers that well responded to the hormonal treatment. Subsequently, we have



**Table 5.** Clinical factors associated with the risk of non-organ confined PCa in clinically localized disease

	Multivariate model I			Multivariate model II (adjusted OR)		
	OR	95% CI	<i>p</i> value	OR	95% CI	<i>p</i> value
PSA	1.135	1.014–1.269	0.027	1.123	1.008–1.251	0.035
BPC	27.763	2.230–345.655	<0.0001	63.381	5.789–643.879	0.001
PRL						
> Median	Ref			Ref		
≤ Median	6.542	2.669–16.036	<0.0001	6.571	2.727–15.831	<0.0001
bGS = 6	Ref					
bGS >6	2.4	0.927–6.214	0.071			

PSA, prostatic specific antigen; BPC, biopsy positive cores; PRL, prolactin; bGS, biopsy Gleason Score; PCa, prostate cancer.

**Fig. 2.** ROC curves of clinical factors associated with the risk of extraprostatic extension in patients with clinically localized PCa without clinical factors interactions. ROC, receiver-operating characteristic; BPC, biopsy positive cores; PSA, prostatic specific antigen; PRL, prolactin; bGS, biopsy Gleason Score.**Fig. 3.** ROC curves of clinical factors associated with the risk of extraprostatic extension in patients with clinically localized PCa with dichotomized PRL levels with and without interaction with PSA and BPC. ROC, receiver-operating characteristic; PSA, prostatic specific antigen; PRL, prolactin; BPC, biopsy positive cores.

shown that low preoperative PRL levels were associated with aggressive PCa in terms of estimated percentage of BPC, bGS, and tumor volume as well as percentage cancer involving the surgical specimen in advanced stage disease (*p* T3b/4) [10, 11].

The influence of hypothalamus-pituitary-testis axis in human PCa physiopathology is a topic that is increas-

ingly being investigated and, recently, an interesting theory has been proposed by Wang et al. [19]. The authors proposed that pivotal changes occur in prostate microenvironment because of variations in serum testosterone levels in aging males [20]. In particular, they supposed that patients with an annual testosterone reduction of more than 30 ng/dL had approximately 5-fold increase in

PCa risk and when a dramatic age-related decrease in serum testosterone is present, the paracrine mechanism of AR action will be replaced by an emergent autocrine mechanism, leading to the selection of pre-neoplastic cells that are less dependent on signals from stromal and basal cells. As a result, this could initiate uncontrolled AR-driven proliferation of luminal cells [19].

According to this theory and the results of the present investigation, we suggest that the decrease of serum PRL physiological levels, which might be caused by endogenous or external factors, can trigger pivotal changes of epithelial cells in the prostate microenvironment. This hypothesis is supported by studies showing that PRL levels increase slightly in healthy aging males while testosterone serum levels decrease [21]. As a theory, when serum PRL levels decrease, the prostatic cell microenvironment activates the paracrine mechanism to compensate the lack of local PRL with self-PRL production and PRL-R overexpression. This should cause cellular hyperstimulation that would result in DNA damage and uncontrolled luminal cells hyperproliferation. Recently, according to a metabolic model, Costello et al. [22] proposed the new concept of “testosterone and PRL dependent model” for PCa. They supported the existence of a cross talk between testosterone and PRL pathways in prostate physiology with the former controlling the latter. The decrease of TT serum level should increase the effect of PRL pathway [22]. This hypothesis finds its biological bases in the old animal model [23]. Recently, a murine model study demonstrated the influence of testosterone in central nervous system dopamine pathway that represents the major PRL inhibitor [24]. These disorders can appear in particular aging male conditions such as obesity that is correlated with alteration of hormonal assessment and associated with lower TT and PRL levels in middle age men [25–27]. These changes, which form the basis of our hypothesis, can contribute to PCa induction and/or progression. Furthermore, recent evidence demonstrated the presence of androgen-independent progenitors of prostate luminal cells (LSC<sup>med</sup> cells) in the probasin-PRL transgenic mouse model prostate cells microenvironment, that may play an important role in the castration resistant-PCa progression [28, 29].

So far, physiopathological variations of both testosterone and PRL levels alter the steady testosterone-PRL system and induce changes that modify normal pathway systems and induce initiation and progression of cancer in prostate cells microenvironment

In a preceding analysis, we have shown that preoperative TT levels are directly associated with tumor grade in the surgical specimen [30]. In that study, we focused on evaluating associations of TT with disease presenting with or without organ confined PCa. In the present analysis, we investigated the associations of PRL preoperative levels with or without confined disease in patients presenting with clinically localized PCa. We did not find any significant associations of TT with the groups. Although the distribution of intermediate high-grade tumors was significantly higher in patients with non-organ confined disease, subjects with intraprostatic disease still showed considerable percentages of intermediate high-grade cancers in the specimen. This might explain why TT did not show any association with non-organ confined disease. So far, the results of the present investigation confirm our preceding findings showing that low PRL levels associate with aggressive PCa biology in the surgical specimen [10, 11]. The association between TT and PCa biology is a controversial topic that still remains unsettled. Indeed, low as well as high TT levels have been associated with aggressive PCa biology [31]. However, in a modern and large cohort of PCa patients including different risk classes, we have shown that increased TT levels are associated with the risk of positive surgical margins in the specimen suggesting association between TT and non-organ confined PCa [32]. The absence of any association between TT and non-organ confined disease in the present study can also be explained by the small cohort of patients.

In previous studies, we demonstrated the predictive value of PSA and BPC in terms of upgrading and upstaging in clinically low, intermediate, and high risk PCa [33–40]. On the basis of those findings, we evaluated in the present study the role of both in association with PRL. We have found a close association between preoperative low PRL and high PSA levels, high BPC, and non-organ confined disease. Indeed PSA and BPC can be an expression of major clinical disease extension. This further confirms the power of our model.

The TT-PRL dependent model could explain the negative results of the LFA102 phase I trial that investigated the potential therapeutic role of a humanized monoclonal antibody that binds and inhibits the PRL-R signaling in patients with PRLR-positive metastatic breast cancer or metastatic castration-resistant PCa [9]. By integrating our results with animal models and the TT-PRL dependent pathway, a controlled study investigating PRL-R inhibitors and androgen blockade can be proposed with the

aim of preventing the selection of castration-resistant cell lines. However, these theories need to be confirmed and the control of the entire male hormonal system could play a major role in PCa prevention.

The present study shows that low preoperative PRL levels associate with the risk non-organ confined PCa in clinically localized disease in pathological specimen. So far, low preoperative PRL levels are independent predictors of the more aggressive PCa biology risk when the disease is clinically confined to the prostate gland, and the power of PRL's predictive role increases with association with PSA and BPC. In clinical practice, the risk model including preoperative PRL levels might be an effective model for assessing the risk of PCa clinical under-staging that still represents a critical issue. To the best of our knowledge, our investigation represents the first clinical study addressing this subject and gives results that correlate with modern basic science findings.

Our study has certain limitations. First, as a retrospective study, it suffers all limitations related to these kinds of studies. Second, the body mass index and prostate volume were not evaluated. Instead PW was evaluated in the surgical specimen and did not show any association with PRL or non-organ confined disease. Third, intraprostatic measurements of PRL, PRL-R, and its mediators were not performed.

However, beyond these limits, our investigation gives evidence of the central role of PRL biology in the natural history of PCa. Moreover, we are confident that our investigation can form the basis for future clinical and pre-clinical trials exploring the role of PRL and its receptor in prostate microenvironment with the aim of finding new therapeutic targets for PCa care.

## Conclusion

The involvement of PRL, PRL-R, and its mediators in PCa pathogenesis is debated and unclear. This is the only study in the recent literature that provides and supports with new theory the evidence that low preoperative serum PRL levels are associated with non-organ confined PCa. Therefore, the preoperative serum PRL level is a useful tool in planning the patient care strategy, and it should be measured in every patient with PCa. Molecular biology clinical studies are needed to confirm the biological basis of this association and its implications in improvement of therapies of advanced PCa.

## Disclosure Statement

The authors declare that they have not conflict of interest.

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