Urinary prostate-specific antigen: predictor of benign prostatic hyperplasia progression?

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Introduction: *Urinary prostate-specific antigen (uPSA) can be used as additional parameter of benign prostatic hyperplasia (BPH) progression.*

Materials and methods: *From January 2001 to December 2011, uPSA was determined in 265 patients with benign prostate. Based on total prostate volume (TPV), the patients with benign prostate were divided in two groups: TPV < 31 mL and TPV ≥ 31 mL. Additional three groups were formed upon MTOPS study criteria: non- progressive BPH group (TPV < 31 mL, PSA < 1.6 ng/mL, age < 62 yrs), intermediate group (one, or two parameters {TPV, PSA, age} increased) and progressive BPH group (TPV ≥ 31 ml, PSA ≥ 1.6 ng/mL, age ≥ 62 yrs).*

Results: *Average uPSA values in the groups TPV < 31 mL and TPV ≥ 31 mL were 119.3 ± 124.5 and 255.5 ± 204.9 ng/mL, respectively and they were significantly different ($p < 0.0001$). Average uPSA values in the non-progressive BPH group, intermediate group and progressive BPH group were 86.8 ± 82.4 ng/mL, 166.6 ± 164.9 ng/mL and 274.9 ± 208.3 ng/mL, respectively and they were significantly different ($p < 0.0001$). The level of uPSA correlated significantly with TPV ($r = 0.32$, $p < 0.0001$). The cut off uPSA level of 150 ng/mL discriminates the patients with non-progressive BPH and progressive BPH with specificity of 0.83 and sensitivity of 0.67.*

Conclusion: *The level of uPSA reflects prostatic hormonal activity and correlates with TPV, PSA and age. UPSA level ≥ 150 ng/mL can be used as additional predictive parameter of BPH progression.*

Key Words: benign prostatic hyperplasia, prostate-specific antigen, urinary prostate specific antigen

Introduction

Benign prostatic hyperplasia (BPH) is one of the most common conditions associated with aging in men. BPH is the progressive disease, characterized with prostate growth, worsening of lower urinary tract symptoms (LUTS) and complications, like acute urinary retention (AUR) and BPH-related surgery.[1] However, it can be said that major biochemical characteristics of BPH are increased action of dihydrotestosterone (DHT) and increased synthesis of prostate-specific antigen (PSA).

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Today, BPH is the fourth most common treated disease.[2] Pan European Expert Project "Triumph" concluded that the incidence rate of LUTS/BPH increases linearly with age and reaches its maximum at the age of 79 years. The prevalence rate was lowest among males 45-49 years of age (2.7%) and increased with age until a maximum at the age of 80 years (24%).[3]

The major risk factors for BPH progression are age, prostate-specific antigen (PSA) and prostate volume (TPV). Other risk factors are symptom severity, decreased maximum urinary flow rate (Qmax) and increased post-void residual (PVR) urine volume. Enlarged prostate and high PSA are related with long term changes in symptom scores and flow rates and represent good clinical predictors of AUR and BPH-related surgery.[4-7] Multicentric study Medical Therapy

of Prostatic Symptoms (MTOPS) defined the most important predictors of BPH progression, after long term follow up of 3047 men with LUTS. These are: TPV ≥ 31 mL, PSA ≥ 1.6 ng/mL, age ≥ 62 years, Qmax < 10.6 mL/s and PVR ≥ 39 mL.[8] Another multicentric study on BPH progression, Combination of Avodart and Tamsulosin (CombAT) included 4844 patients followed up during 4 years. The study included the patients older than 50 years, with moderate to severe BPH symptoms, PSA levels of ≥ 1.5 ng/mL and ≤ 10 ng/mL, and a TPV ≥ 30 mL. After 2 years, the conclusion of the study was that in patients with TPV ≥ 30 mL combination therapy provides a significantly greater degree of benefit than tamsulosin or dutasteride monotherapy.[9] However, after 4 years, the study concluded that the greater reduction of clinical progression and the complications was achieved in men with TPV ≥ 40 mL, using combined therapy versus monotherapy.[10]

Usually, BPH is described as a pathological proliferation of prostatic fibroblasts and epithelial cells. Age, androgens, epithelial-mesenchymal transition (EMT) and TGF-beta are all contributing factors to the pathogenesis of BPH.[11,12] Hyperplasia occurs in the transitional zone, which normally constitutes only 5% of glandular prostate, but never in the peripheral zone, which constitutes 70% of the glandular prostate.[13] Androgens play an essential role in prostate development, prostate growth and the pathogenesis of BPH. Although testosterone is the main androgen secreted from the testes, DHT is the major androgen in the prostate cells. Enzymes 5 alpha-reductase (5AR), type 1 and 2, convert testosterone to DHT; 5AR-2 is the predominant isozyme expressed in the stromal cells.[14] During relatively slow, genomic process, testosterone and DHT molecules bind to androgen receptor (AR), enter the nucleus and together with AR bind to specific DNA sequences, termed androgen-responsive elements (AREs) ,[15] where they stimulate genes responsible for cell growth and survival and PSA synthesis.[16,17] The stimulation of PSA synthesis could be carried on by cell- membrane steroid receptor, as well; this process is faster and lasts 1-30 minutes.[18]

The action of DHT is markedly increased in BPH, most likely due to increased DHT tissue concentration, although there is no consensus about that.[19-22] Elderly men often have elevated serum DHT concentration; the treatment with testosterone usually decreases DHT level in these patients.[23,24]

The secretion and excretion of PSA

The concentration of PSA in prostatic tissue differs significantly in benign and malignant tissue: it is the lowest in the poor differentiated malignant cells, higher in the normal tissue and the highest in BPH glands.[25-32]

The synthesis of PSA takes place in the secretory epithelial cells. After the synthesis, PSA molecules are contained in 1μ- large prostatic secretory granules (PSG), which fulfill the cytosol.

During the process of "apocrine decapitation", PSG are thrown out the cell, in the lumen of the prostatic duct, where PSA molecules are released.[33] Within seminal fluid, PSA molecules leak through excretory ducts to reach prostatic urethra. Additional PSA molecules originate from "minor prostatic glands", urethral epithelial secretory cells that contain PSG.[34] In the urethral lumen, PSA molecules accumulate until the next micturition, when they are expelled from the body.

All excreted PSA molecules originate from the prostate and the urethra. Complexed PSA molecules are too large to pass glomerular membrane, and they were never discovered in the urine from nephrostomy catheters.[35] In addition, during heart catheterization, venous blood samples noted no decrease in the PSA concentration across the renal circulation.[36] Therefore, PSA is not the natural ingredient of the urine and the accustomed term "urinary PSA" is not quite precise.

During ejaculation, massive amount of PSA molecules is injected in the prostatic urethra.[37] After ejaculation, the first post-ejaculatory urine sample contains high concentration of PSA, while second and following post-ejaculatory samples contain baseline amounts of PSA.[38]

Clinical importance of determination of excreted PSA

The amount of excreted PSA can be measured by prostatic massage, or, usually, in the voided urine when it is called urinary PSA. The level of uPSA depends of age, hormonal status and the presence of BPH. While most males younger than 40 years have uPSA < 20 ng/mL, average uPSA in the BPH patients measures around 200 ng/mL.[39-41] Urinary PSA in healthy females is very low, close to zero, like in prepubertal boys and castrated males.[42-44] Prostate cancer and BPH have similar uPSA values, although some authors found lower uPSA in prostate cancer than in BPH patients.[45-50] After radical prostatectomy, almost all patients have measurable levels of uPSA, with average of 20 ng/mL. The level of uPSA cannot discover cases with local recurrence, because all PSA molecules after radical prostatectomy originate from urethral glands.[51-53]

Materials and methods

From January 2001 to December 2011, urinary PSA was determined in 265 patients with benign prostate, in Urological Clinic, Clinical Centre of Serbia. All patients

had long term follow up with at least two annual digital rectal examinations (DRE) and TPV measurement using abdominal, or transrectal ultrasonography (TRUS) and at least two annual records of PSA and uPSA. The patients were explained to collect the first stream of the first morning urine and to fill urine container (80 mL) to the top. All patients had to bring urine sample by 9:00 a.m. to the laboratory. The same day the patients underwent blood extraction for serum PSA analysis. Total prostate volume was measured using abdominal probe of 5.7 MHz, based on standard formula for ellipsoid (A x B x C x $\pi/6$). All ultrasound and physical examinations and measurements were done by the same urologist. In the cases with elevated PSA and/or suspect finding on DRE and/or TRUS, TRUS- guided prostate biopsy was performed. The cases with proven prostate cancer were excluded from the study. The values of PSA and uPSA that were included in the statistical analysis were the latest, or before any treatment.

Serum and urinary PSA were determined in the Central laboratory of Clinical Centre of Serbia. From January 2001 to September 2009, PSA and uPSA were determined using AxSYM Total PSA assay (Abbott Laboratories, Illinois, USA), a Microparticle Enzyme Immunoassay (MEIA). From September 2009, ARCHITECT Total PSA assay (Abbott Laboratories, Illinois, USA), a Chemiluminescent Microparticle Immunoassay (CMIA) was used. ARCHITECT Total PSA assay results are comparable to results from the AxSYM Total PSA assay (correlation coefficient r = 0.987). The samples were spun in the centrifuge at 2000 rpm for 10 minutes. Aliquots of 500 µL samples were frozen at -20ºC and stored. Urine samples were thawed only once prior to PSA determination.

Taking the prostate volume of 31 mL as a cut off point, the whole group of 265 patients was divided in two groups: 1) TPV < 31 mL (126 patients) and 2) TPV ≥ 31 mL (139 patients).

In addition, using MTOPS study criteria, the same group with 265 patients was divided in three other groups: 1) non-progressive BPH (TPV < 31mL, PSA < 1.6 ng/mL, age < 62 yrs) , 2) intermediate group (one or two parameters {TPV, PSA, age} increased) and 3) progressive BPH (TPV ≥ 31mL, PSA ≥ 1.6ng/mL, age ≥ 62 yrs.)

TABLE 1. **Average values of parameters**

Characteristics	Average ± SD	Range
Age (yrs)	64.9 ± 10.6	25-85
TPV (mL)	37.05 ± 21.02	12-120
PSA (ng/mL)	2.19 ± 1.96	0.2-9.9
uPSA (ng/mL)	190.8 ± 184.2	1.8-870.5

TPV = total prostate volume; PSA = prostate-specific antigen; uPSA = urinary prostate-specific antigen

Statistical analysis

Average values and standard deviations (SD) were calculated using Microsoft Excel 2007 program. The significance of observed variances, T-test, correlations and ROC analysis were done using the scientific calculator at VassarStats website for statistical computation.

Results

Average values of age, TPV, PSA and uPSA

The average values ± standard deviations (SD) and ranges of age, TPV, PSA and uPSA in the whole group are presented in Table 1.

The distribution of patients based on TPV of 31 mL as a cut off

The average values of age, PSA and uPSA in the groups TPV < 31 mL and TPV ≥ 31 mL were significantly different ($p < 0.0001$), Table 2.

The distribution of patients based on MTOPS criteria

The average uPSA in the non-progressive BPH group (86.8 ± 82.4 ng/mL) was significantly lower than the average uPSA in the intermediate group (166.6 ± 164.9 ng/mL, $p < 0.0001$). In addition, the average uPSA in the intermediate group was significantly lower than the

TABLE 2. **Two groups based on total prostate volume**

TPV (mL)	n	Age (yrs)	PSA (ng/mL)	uPSA (ng/mL)
< 31	126	62.3 ± 13.1	1.27 ± 1.19	119.3 ± 124.5
≥ 31	139	67.4 ± 6.8	3.03 ± 2.14	255.5 ± 204.9
		$p < 0.0001$	$p < 0.0001$	$p < 0.0001$

TPV = total prostate volume; PSA = prostate-specific antigen; uPSA = urinary prostate-specific antigen

TABLE 3. **Groups based on MTOPS criteria**

Group	n	Age (yrs)	TPV (mL)	PSA (ng/mL)	uPSA (ng/mL)
Non-progressive BPH (TPV < 31 mL, PSA < 1.6 ng/mL, age < 62 yrs)	41	47.2 ± 10.9 p < 0.0001	19.3 ± 5.5 p < 0.0001	0.62 ± 0.33 p < 0.0001	86.8 ± 82.4 p < 0.0001
Intermediate (one or two parameters {TPV, PSA, age} increased)	135	66.7 ± 6.9 p < 0.0001	31.2 ± 14.1 p < 0.0001	1.77 ± 1.58 p < 0.0001	166.6 ± 164.9 p < 0.0001
Progressive BPH (TPV ≥ 31 mL, PSA ≥ 1.6 ng/mL, age ≥ 62 yrs)	89	70.5 ± 4.8	54.3 ± 22.5	3.55 ± 2.09	276.8 ± 208.7

TPV = total prostate volume; PSA = prostate-specific antigen; uPSA = urinary prostate-specific antigen; BPH = benign prostatic hyperplasia

average uPSA in the progressive BPH group (276.8 ± 208.7, p < 0.0001). Likewise, the average values of age, TPV and PSA were significantly different in the neighboring groups, Table 3.

Correlations

There were weak, but significant correlations between various parameters of BPH progression in the group of patients with benign prostate. Urinary PSA correlated significantly with TPV (r = 0.32, p < 0.0001) and less significantly with PSA (r = 0.18, p = 0.0013) and patient age (r = 0.14, p = 0.009). However, the strongest correlation was noted between serum PSA and TPV (r = 0.51, p < 0.0001), Table 4.

Cut off values, sensitivity and specificity

In the whole group of 265 patients, 147 (55.5 %) had uPSA ≤ 150 ng/mL, 59 (22.3 %) had uPSA between 150 ng/mL and 300 ng/mL, while 59 patients (22.3%) had uPSA > 300 ng/mL. In the same uPSA ranges, the distribution of patients differed significantly between groups TPV < 31mL and TPV ≥ 31 mL, Table 5 and Figure 1.

In the whole group, uPSA cut off value of 150 ng/mL discriminates the patients with TPV < 31 mL and the patients with TPV ≥ 31 mL with sensitivity (Sn) of 0.61 and specificity (Sp) of 0.74. The ROC analysis revealed AUC of 0.726. In the groups formed upon MTOPS criteria, the comparison was calculated between non-progressive BPH group and progressive BPH group, while intermediate group was excluded due to its inhomogeneity. Urinary PSA cut off value of 150 ng/mL discriminates the patients from non- progressive and progressive BPH group with Sn of 0.67 and Sp of 0.83. The ROC analysis revealed AUC of 0.814.

TABLE 4. **The correlations between parameters**

	TPV	PSA	Age
uPSA	r = 0.32 (p < 0.0001)	r = 0.18 (p = 0.0013)	r = 0.14 (p = 0.009)
PSA	r = 0.51 (p < 0.0001)	-	r = 0.23 (p < 0.0001)

TPV = total prostate volume; PSA = prostate-specific antigen; uPSA = urinary prostate-specific antigen

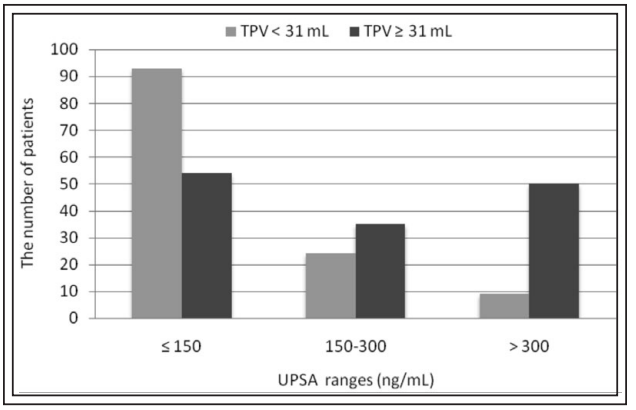

Figure 1. The number of patients in various uPSA ranges.

TABLE 5. **UPSA ranges and TPV**

uPSA range (ng/mL)	TPV < 31 mL	TPV ≥ 31 mL
≤ 150	93	54
150-300	24	35
> 300	9	50
Total	126	139

TPV = total prostate volume; PSA = prostate-specific antigen; uPSA = urinary prostate-specific antigen

Discussion

Total prostate volume (TPV) has always been the most important characteristic of BPH. However, TPV alone is not useful in the estimation of disease severity. Additional subjective and objective parameters are needed to estimate the risk of BPH progression. Various multicentric studies, like MTOPS, CombAT, ALTESS and others, defined PSA, patient age, IPSS, Qmax and PVR as most important additional parameters of BPH progression.

Numerous studies confirmed significant correlation between TPV and PSA.[54-56] However, in the PSA- based BPH follow up, there is a problem how to differentiate the benign and malignant increase of PSA: the rise of PSA ≥ 1.6 ng/mL suggests BPH progression, but PSA level > 2.5 ng/mL could also indicate prostate cancer.

Theoretically, the amount of excreted PSA reflects the secretory capacity of the prostate and local hormonal milieu and represents the real marker of BPH and prostatic intracrine activity. On the other hand, serum PSA is the marker of prostatic tissue damage and prostatic hormonal activity. Nevertheless, although the presence of PSA in the urine was proven early, not many issues have been written about it.

Except during ejaculation, there is a constant PSA leakage from prostatic acini to prostatic urethra. In addition, urethral secretory cells secrete additional PSA along whole urethra. These PSA molecules collect in the urethra during the interval between two micturitions; the next voiding washes out all PSA from the urethra. Therefore, the term "urinary PSA" is incorrect, as it suggests the urinary origin of PSA. The authors of this issue recommend the use of more precise terms, like "excreted PSA", or "voided PSA".

In this study, PSA was determined in the first 80 mL of the first morning voiding. The volume of the urine sample is a bit bigger than in other studies,[47] (80 mL is the volume of the urine containers in our institution) but it is less important: it is important that all patients provide the same sample volume in which voided PSA concentration will be calculated. In addition, all patients had to bring the urine sample to the laboratory by 9.00 a.m. to provide fresh urine and the similar time of sampling. The authors believe that first urinary stream reflects the maximal PSA excretion[51,52] through the longest period of continence during the night. The results were expressed as PSA concentration in the voided urine sample, rather than the total amount of voided PSA. This way is usual in the literature and provides the comparison of the results. The authors believe that there is no reason for expressing PSA concentration per mmol of creatinine.[47]

In order to record average uPSA values for the single patient, it is very important to obtain multiple uPSA determinations. This is also important in the follow up of the patients with BPH and for the measurement of the uPSA response on 5ARI.

The majority of patients in the study had relatively homogenous uPSA values during follow up, while a number of patients had uPSA variations, which can be the consequence of few phenomena. At first, inaccurate urine sampling, like low urine volume, or the sampling of the inadequate portion of the urinary stream. Second, the different intervals between the last evening and the first morning voiding result in different amounts of excreted PSA. Lastly, morning erections, sexual excitement and recent sexual intercourse could be associated with increased PSA excretion.

In the older study, in the group of 42 patients with BPH, DeVere White found mean urinary PSA level of 216 ng/mL, which did not correlate with prostate size.[57] In the group of patients with PSA between 2.5 ng/mL and 10 ng/mL, Bolduc et al found lower average uPSA in the group of 29 patients with prostate cancer, than in the group of 35 patients with BPH (52.6 ng/mL and 123.2 ng/mL, respectively). Urinary PSA cut off value of 150 ng/mL was found to differentiate the patients with BPH and prostate cancer, with Sn of 92.5%.

The authors of this study found that average uPSA concentration was 190.8 ± 184.2 ng/mL in the group of 265 patients with normal prostate or BPH and a significant correlation between uPSA and prostate size. Moreover, men with smaller prostate (TPV < 31 mL) and bigger prostate (TPV ≥ 31 mL) had significantly different uPSA values. Similarly with Bolduc, the authors found uPSA cut off level of 150 ng/mL, but that value separated the patients within benign prostate group. If only one parameter (TPV > 31 mL) was used, specificity and sensitivity were moderate (Sp = 0.74, Sn = 0.61); however, when additional parameters of BPH progression (age > 62 years, PSA > 1.6 ng/mL) were introduced, statistical parameters were higher (Sp = 0.83, Sn = 0.67). However, the authors did not find statistical difference between average uPSA in BPH and prostate cancer patients: in the unpublished study, 51 patients with prostate cancer in various stages of untreated prostate cancer had similar average uPSA (286.9 ± 300.4 ng/mL) like the patients in the progressive BPH group (p = 0.42). Mean uPSA in the 46 patients after radical prostatectomy was 16.4 ± 36.7 ng/mL.

Conclusion

The main conclusions of this study are that uPSA level correlates with prostate volume and reflects prostatic

increased hormonal activity in BPH. Urinary PSA > 150 ng/mL is independent marker of BPH and BPH progression and could be used together with other known parameters.

In the diagnosis and follow up of patients with BPH, uPSA have some advantages over PSA: the test is simple to perform and can be repeated many times. Second, the diagnostic range of uPSA is very wide in BPH, 1-2 ng/mL to over 800 ng/mL, which is not the case with serum PSA.

The authors of this paper noticed fast and abundant response of uPSA following the treatment with 5ARI. In the unpublished study, in the group of 48 patients, average uPSA showed the highest decrease after 4 months of treatment with 5ARI (57.6%), compared to PSA (26.9%) and TPV (11.9%). In addition, elevated uPSA in younger patients could present the earliest sign of BPH awakening. These are the reasons for optimism about the clinical use of uPSA; nevertheless, further studies and some modifications in methodology are needed for the routine utilization of uPSA. □

References

1. Fitzpatrick JM. The natural history of benign prostatic hyperplasia. *BJU Int* 2006;97(Suppl 2):3-6; discussion 21-22.
2. Naslund MJ, Issa MM, Grogg AL, Eaddy MT, Black L. Clinical and economic outcomes in patients treated for enlarged prostate. *Am J Manag Care* 2006;12(4 Suppl):S111-S116.
3. Verhamme KM, Dieleman JP, Bleumink GS et al; Triumph Pan European Expert Panel. Incidence and prevalence of lower urinary tract symptoms suggestive of benign prostatic hyperplasia in primary care--the Triumph project. *Eur Urol* 2002;42(4):323-328.
4. de la Rosette JJ, Alivizatos G, Madersbacher S et al. EAU Guidelines on Benign Prostatic Hyperplasia. *Eur Urol* 2001;40(3):256-263.
5. Roehrborn CG, Boyle P, Bergner D et al. Serum prostate-specific antigen and prostate volume predict long-term changes in symptoms and flow rate: results of a four-year, randomized trial comparing finasteride versus placebo. PLESS Study Group. *Urology* 1999;54(4):662-669.
6. Roehrborn CG. BPH progression: concept and key learning from MTOPS, ALTESS, COMBAT, and ALF-ONE. *BJU Int* 2008;101(Suppl 3):17-21.
7. Roehrborn CG. The utility of serum prostatic-specific antigen in the management of men with benign prostatic hyperplasia. *Int J Impot Res* 2008;20(Suppl 3):S19-S26.
8. Crawford ED, Wilson SS, McConnell JD et al; MTOPS RESEARCH Group. Baseline factors as predictors of clinical progression of benign prostatic hyperplasia in men treated with placebo. *J Urol* 2006;175(4):1422-1426.
9. Roehrborn CG, Siami P, Barkin J, Damião R, Major-Walker K, Morrill B, Montorsi F; CombAT Study Group. The effects of dutasteride, tamsulosin and combination therapy on lower urinary tract symptoms in men with benign prostatic hyperplasia and prostatic enlargement: 2-year results from the CombAT study. *J Urol* 2008;179(2):616-621; discussion 621.
10. Roehrborn CG, Barkin J, Siami P et al. Clinical outcomes after combined therapy with dutasteride plus tamsulosin or either monotherapy in men with benign prostatic hyperplasia (BPH) by baseline characteristics: 4-year results from the randomized, double-blind Combination of Avodart and Tamsulosin (CombAT) trial. *BJU Int* 2011;107(6):946-954.
11. Timms BG, Hofkamp LE. Prostate development and growth in benign prostatic hyperplasia. *Differentiation* 2011;82(4-5):173-183.
12. Alonso-Magdalena P, Brössner C, Reiner A et al. A role for epithelial-mesenchymal transition in the etiology of benign prostatic hyperplasia. *Proc Natl Acad Sci U S A* 2009;106(8): 2859-2863.
13. McNeal JE. The zonal anatomy of the prostate. *Prostate* 1981;2(1):35-49.
14. Eicheler W, Tuohimaa P, Vilja P, Adermann K, Forssmann WG, Aumüller G. Immunocytochemical localization of human 5 alpha-reductase 2 with polyclonal antibodies in androgen target and non-target human tissues. *J Histochem Cytochem* 1994;42(5):667-675.
15. Balk SP, Ko YJ, Bubley GJ. Biology of prostate-specific antigen. *J Clin Oncol* 2003;21(2):383-391.
16. Zhu YS, Cai LQ, You X, Cordero JJ, Huang Y, Imperato-McGinley J. Androgen-induced prostate-specific antigen gene expression is mediated via dihydrotestosterone in LNCaP cells. *J Androl* 2003;24(5):681-687.
17. Zhu YS, Sun GH. 5α-Reductase Isozymes in the Prostate. *J Med Sci* 2005;25(1):1-12.
18. Kampa M, Papakonstanti EA, Hatzoglou A, Stathopoulos EN, Stournaras C, Castanas E. The human prostate cancer cell line LNCaP bears functional membrane testosterone receptors that increase PSA secretion and modify actin cytoskeleton. *FASEB J* 2002;16(11):1429-431.
19. Olsson M, Ekström L, Guillemette C, Belanger A, Rane A, Gustafsson O. Correlation between circulatory, local prostatic, and intra-prostatic androgen levels. *Prostate* 2011;71(9): 909-914.
20. Zhang Y, Ye L, Ding Q, Fang Z, Yao M, Shi D. Measurement of T and DHT contents in normal and diseased human prostate tissues. *Zhonghua Wai Ke Za Zhi* 2000;38(7):545-547.
21. Marks LS, Mostaghel EA, Nelson PS. Prostate tissue androgens: history and current clinical relevance. *Urology* 2008;72(2):247-254.
22. Van der Sluis TM, Vis AN, van Moorselaar RJ et al. Intraprostatic testosterone and dihydrotestosterone. Part I: concentrations and methods of determination in men with benign prostatic hyperplasia and prostate cancer. *BJU Int* 2012;109(2):176-182.
23. Trifiro MD, Parsons JK, Palazzi-Churas K, Bergstrom J, Lakin C, Barrett-Connor E. Serum sex hormones and the 20-year risk of lower urinary tract symptoms in community-dwelling older men. *BJU Int* 2010;105(11):1554-1559.
24. Gooren LJ, Saad F, Haide A, Yassin A. Decline of plasma 5alpha dihydrotestosterone (DHT) levels upon testosterone administration to elderly men with subnormal plasma testosterone and high DHT levels. *Andrologia* 2008;40(5):298-302.
25. Vesey SG, Goble M, Ferro MA, Stower MJ, Hammonds JC, Smith PJB. Quantification of prostatic cancer metastatic disease using prostate specific antigen. *Urology* 1990;35(6):483-486
26. Erickson DR, Hlavinka TC, Rockwood AP, Metter JD, Novicki DE, Fried MG. Prostatic acid phosphatase, beta-glucuronidase and prostate specific antigen assays in fine needle aspirates from benign and malignant prostates. *J Urol* 1991;146(5):1402-1406.
27. Denmeade SR, Sokoll LJ, Chan DW, Khan SR, Isaacs JT. Concentration of enzymatically active prostate-specific antigen (PSA) in the extracellular fluid of primary human prostate cancers and human prostate cancer xenograft models. *Prostate* 2001;48(1):1-6
28. Jung K, Brux B, Lein M et al. Molecular forms of prostate specific antigen in malignant and benign prostatic tissue: biochemical and diagnostic implications. *Clin Chem* 2000;46(1):47-54.

29. Ersev A, Ersev D, Turkery L et al. The relation of prostatic acid phosphatase and prostate specific antigen with tumor grade in prostatic adenocarcinoma: an immunohistochemical study. *Prog Clin Biol Res* 1990;357:129-134.
30. Bostwick DG. Prostate specific antigen and pathology of the prostate. *Eur Urol* 1995;27(Suppl 2):5.
31. Pretlow TG, Pretlow TP, Yang B et al. Tissue concentrations of prostate specific antigen in prostatic carcinoma and benign prostatic hyperplasia. *Int J Cancer* 1991;49(5):645-649.
32. Weir EG, Partin AW, Epstein JI. Correlation of serum prostate specific antigen and quantitative immunohistochemistry. *J Urol* 2000;163(6):1739-1742.
33. Cohen RJ, McNeal JE, Redmond SL et al. Luminal contents of benign and malignant prostatic glands: correspondence to altered secretory mechanisms. *Hum Pathol* 2001;32(2):242-243.
34. Cohen RJ, Garrett K, Golding JL, Thomas RB, McNeal JE. Epithelial differentiation of the lower urinary tract with recognition of the minor prostatic glands. *Hum Pathol* 2002;33(9): 905-909.
35. Kabalin JN, Hornberger JC. Prostate specific antigen is not excreted by human kidney or eliminated by routine hemodialysis. *Urology* 1991;37(4):308-310.
36. Agha AH, Schechter E, Roy JB, Culkin DJ. Prostate specific antigen is metabolized in the liver. *J Urol* 1996;155(4):1332-1335.
37. Wang MC, Papsidero LD, Kuriyama M, Valenzuela LA, Murphy GP, Chu TM. Prostate antigen: a new potential marker for prostatic cancer. *Prostate* 1981;2(1):89-96.
38. Sivananthan T, Bathur F, Jimenez M, Conway A, Idan A, Handelsman D. Objective non-intrusive markers of sperm production and sexual activity. *Asian J Androl* 2012;14(3):476-480.
39. Tremblay J, Frenette G, Tremblay R, Dupont A, Thabet M, Dube JY. Excretion of three major prostatic secretory proteins in the urine of normal men and patients with benign prostatic hypertrophy or prostate cancer. *Prostate* 1987;10(3):235-243.
40. Pejcic T, Hadzi-Djokic J, Acimovic M, Topuzovic C, Milkovic B, Janjic A. Urinary prostate specific antigen: is the clinical use likely? *Acta Chir Iugosl* 2005;52(4):69-74.
41. Pejčić TP. Prostata specifični antigen u urinu. Zadužbina Andrejević, Beograd, 2005.
42. Zaviacic M, Ruzickova M, Jakubovsy J, Danihel L, Babal P, Blazekova J. The significance of prostate markers in the orthology of the female prostate. *Bratisl Lek Listy* 1994;95(11):491-497.
43. Obiezu CV, Giltay EJ, Magklara A et al. Serum and urinary prostate-specific antigen and urinary human glandular kallikrein concentrations are significantly increased after testosterone administration in female-to-male transsexuals. *Clin Chem* 2000;46(6 Pt 1):859-862.
44. Sato I, Yoshikawa A, Shimizu K, Ishiwari A, Mukai T, Iwamoto T. Urinary prostate-specific antigen is a noninvasive indicator of sexual development in male children. *J Androl* 2007;28(1):155-157.
45. Breul J, Pickl U, Hartung R. Prostate-specific antigen in urine. *Eur Urol* 1994;26(1):18-21.
46. Hillenbrand M, Bastian M, Steiner M et al. Serum-to-urinary prostate-specific antigen ratio in patients with benign prostatic hyperplasia and prostate cancer. *Anticancer Res* 2000;20(6D):4995-4996.
47. Irani J, Millet C, Levillain P, Dore B, Begon F, Aubert J. Serum-to-urinary prostate-specific antigen ratio: its impact in distinguishing prostate cancer when serum prostate specific antigen level is 4 to 10 ng/mL. *J Urol* 1997;157(1):185-188.
48. Pannek J, Rittenhouse HG, Evans CL et al. Molecular forms of prostate-specific antigen and human kallikrein 2 (hK2) in urine are not clinically useful for early detection and staging of prostate cancer. *Urology* 1997;50(5):715-721.
49. Bolduc S, Lacombe L, Naud A, Gregoire M, Fradet Y, Tremblay RR. Urinary PSA: a potential useful marker when serum PSA is between 2.5 ng/mL and 10 ng/mL. *Can Urol Assoc J* 2007;1(4): 377-381.
50. Pejcić T, Hadzi-Djokić J, Marković B et al. Urinary PSA level and relative tumor volume after prostate biopsy. *Acta Chir Iugosl* 2009;56(2):17-21.
51. Iwakiri J, Granbois K, Wehner N, Graves HC, Stamey TA. An analysis of urinary prostate specific antigen before and after radical prostatectomy: evidence for secretion of prostate specific antigen by the periurethral glands. *J Urol* 1993;149(4):783-786.
52. Takayama TK, Vessella RL, Brawer MK, True LD, Noteboom J, Lange PH. Urinary prostate specific antigen levels after radical prostatectomy. *J Urol* 1994;151(1):82-87.
53. Pejcić T, Hadzi-Djokić J, Marković B, Lalić N, Glisić B. What are the possible reasons for urethral PSA varieties after radical prostatectomy? *Acta Chir Iugosl* 2010;57(2):31-35.
54. Berges R, Oelke M. Age-stratified normal values for prostate volume, PSA, maximum urinary flow rate, IPSS, and other LUTS/BPH indicators in the German male community-dwelling population aged 50 years or older. *World J Urol* 2011;29(2):171-178.
55. Vesely S, Knutson T, Damber JE, Dicuio M, Dahlstrand C. Relationship between age, prostate volume, prostate-specific antigen, symptom score and uroflowmetry in men with lower urinary tract symptoms. *Scand J Urol Nephrol* 2003;37(4):322-328.
56. Roehrborn CG, Boyle P, Gould AL, Waldstreicher J. Serum prostate-specific antigen as a predictor of prostate volume in men withbenign prostatic hyperplasia. *Urology* 1999;53(3):581-589.
57. DeVere White RW, Meyers FJ, Soares SE, Miller DG, Soriano TF. Urinary prostate specific antigen levels: role in monitoring the response of prostate cancer to therapy. *J Urol* 1992;147(3 Pt 2): 947-951.