Asian Journal of Andrology (2017) 19, 463–467
www.asiaandro.com; www.ajandrology.com

Open Access

ORIGINAL ARTICLE

Prostate Disease

Influence of age on seven putative prostate tumor markers: a cohort study in Chinese men

Wei-Gui Sun[1], Chao-Zhao Liang[2], Qi-Chuan Zheng[1], Xiao-Wu Hu[3], Zhi-Zhen Li[3], Ping Wu[3]

The accuracy and sensitivity of prostate-specific antigen (PSA) for prostate cancer diagnosis is often poor; however, the reasons for its inaccuracy have rarely been investigated, especially with respect to age. In this study, 476 healthy males, aged 10–89 years, were stratified into eight age groups, and levels of seven markers were determined: total PSA (tPSA), free PSA (fPSA), %fPSA, isoform [-2]proPSA (p2PSA), p2PSA/tPSA, %p2PSA, and the prostate health index (PHI). Both tPSA and fPSA levels increased with age. The tPSA level was highest (1.39 ng ml^{-1}) at 70–79 years; %fPSA was highest (0.57 ng ml^{-1}) at 10–19 years; and %p2PSA was lowest (18.33 ng ml^{-1}) at 40–49 years. Both p2PSA and p2PSA/tPSA had relatively flat curves and showed no correlation with age ($P = 0.222$). PHI was a sensitive age-associated marker ($P < 0.05$), with two peaks and one trough. The coverage rates and radiance graphs of PHI and %p2PSA were more distinctive than those of tPSA and the other markers. In subjects older than 69 years, PHI and %p2PSA both began to decrease, approximately 10 years earlier than the decrease in tPSA. Our results suggest that the clinical diagnosis of prostate cancer using PSA should be investigated more comprehensively based on patient age. Moreover, %p2PSA and PHI could be considered as earlier markers that may be more suitable than PSA alone.

Asian Journal of Andrology (2017) **19**, 463–467; doi: 10.4103/1008-682X.175787; published online: 5 April 2016

Keywords: age factors; diagnosis; prostate cancer; tumor marker

INTRODUCTION

The use of the prostate-specific antigen (PSA) test for early diagnosis of human prostate cancer (PCa) has resulted in both overdiagnosis and overtreatment.[1,2] Recent reports suggest that new indicators such as %p2PSA (isoform [-2]proPSA) and the prostate health index (PHI) have better clinical value than conventional PSA alone.[3,4] However, there is a lack of studies on these potential markers, and the reasons for using %p2PSA and PHI have not been clarified.

Previous studies concerning prostate disorders have focused on elderly men; however, various prostate diseases appear in men at different ages, such as prostatitis, benign prostate hyperplasia (BPH), and PCa. A comprehensive study of dynamic changes and detailed characteristics of tumor markers in men of all age groups is yet to be published.

In previous studies, several potential markers, including total PSA (tPSA), free PSA (fPSA), %fPSA, p2PSA, p2PSA/tPSA, %p2PSA, and PHI, have either been individually investigated or separately compared. These seven markers have yet to be assessed together to identify mutual relationships among them, and to examine their usefulness as markers. Moreover, the possible reasons underlying the lack of sensitivity and specificity of PSA for diagnosis of PCa, such as the confounding effects of age, also require more detailed examination. Last, previous studies have mostly focused on clinical PCa patients, and few studies have assessed healthy men. Levels of these prostate markers in healthy men can provide normal reference levels and certain fundamental characteristics, such that abnormal changes and results in PCa patients can be judged more accurately.

This study was performed in an age-based population of healthy men in China to investigate the characteristics of these prostate-specific markers.

PATIENTS AND METHODS

The study comprised 476 healthy Chinese men of various ages randomly selected from men who enrolled in a community health physical survey. The mean age of the subjects was 51.87 (range, 11–89) years. Participants were stratified into eight groups according to age decade (**Figure 1**). Any subject suspected of having PCa or who had a history of any cancer was excluded from the analysis. The study complied with the Code of Ethics of the World Medical Association.

Blood samples were collected from participants during the same morning and promptly centrifuged at 4°C for analysis within 2 h. Seven markers, including tPSA, fPSA, %fPSA (fPSA/tPSA), p2PSA, %p2PSA (p2PSA/fPSA), p2PSA/tPSA, and PHI,[1,2] were determined. Human PSA (Roche, USA) kits and human p2PSA kits (Elabscience Biotechnology Co., Ltd., Wuhan, China) were used for serum analyses by immunometric assay (E170, ElectroChemiLuminescence, Roche). Other data from physical examination were also collected, including information related to the prostate and findings of urinary system ultrasonography and magnetic resonance imaging. Comparisons were made with the *t*-test and one-way analysis of variance using SPSS version 19.0 (IBM Corp., Armonk, NY, USA).

[1]Research Institute of Urology, Ma'anshan People's Hospital, Affiliated to Anhui Medical University, Ma'anshan 243000, China; [2]Department of Urology, The First Affiliated Hospital of Anhui Medical University, Hefei 230000, China; [3]The Center of Clinical Molecular Biology Laboratory, Ma'anshan 243000, China.
Correspondence: Dr. WG Sun (docswg@sina.com)
Received: 07 June 2015; Revised: 05 September 2015; Accepted: 17 December 2015

RESULTS

The tPSA levels of 22 (4.62%) men showed an abnormal level (above 10 ng ml^{-1}), and those of 12 (2.52%) men were found to be in the diagnostic gray zone (4.0–10.0 ng ml^{-1}).[3,4] These 34 men were excluded from the present study and were referred for further extensive medical diagnostics or long-term follow-up. Thus, the final number of healthy subjects enrolled in this study was 442 (92.86%).

The mean values of the seven markers are displayed in **Tables 1** and **2**, and the age-related distribution curves are shown in **Figure 2a–2c**. In general, the tPSA curve increased with increasing age in healthy men ($P < 0.001$). Before 70 years of age, the tPSA curve showed two rapidly increasing levels: at the ages of 10–19 years and 60–69 years. tPSA increased to a peak of 1.00 ng ml^{-1} at 40–49 years and showed its highest peak (1.39 ng ml^{-1}) at 70–79 years. Subsequently, the tPSA curve demonstrated a gradual decrease. The fPSA curve also increased with increasing age ($P < 0.001$) but with a shallower slope and slower rate than tPSA. Moreover, fPSA continued to increase in the age group of 70–79 years, unlike tPSA, which showed a decrease. The %fPSA curve showed a gradual decrease with age and was not associated with any specific age ($P > 0.5$). The %fPSA curve was highest at 10–19 years (%fPSA = 0.57) and lowest at 50–59 years (%fPSA = 0.37).

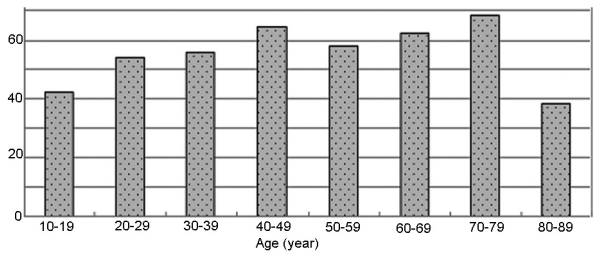

Figure 1: The age distribution and eight groups.

The features of the p2PSA curve were similar to those of fPSA. p2PSA/tPSA provided a relatively flat curve with fewer fluctuations, and no specific association with age ($P = 0.222$). The %p2PSA curve showed certain distinct features. At the age of 40–49 years, when tPSA increased to 1.00 ng ml^{-1}, %p2PSA simultaneously decreased to its lowest value (18.33). Subsequently, %p2PSA rapidly increased to its highest value (47.54) at 60–69 years, then decreased rapidly. In contrast, the decrease in tPSA occurred only after 79 years of age (**Figure 2a**), 10 years after the age-related decline in %p2PSA.

PHI presented with the most distinctive characteristics of all the markers (**Figure 2c**). PHI showed the strongest fluctuations with age ($P < 0.05$). The PHI curve had certain marked characteristics, including two peaks and one trough. The first peak was observed at 30–39 years (PHI = 31.83 ± 10.29, 95% confidence interval [95% CI] = 10.29–53.37). The second peak was the highest lifetime peak and was observed at 60–69 years (PHI = 42.85 ± 9.06, 95% CI = 23.96–61.75). A deep trough was observed at

Table 1: PSA levels of 442 healthy men

Age (year)	*n*	*tPSA (ng ml^{-1})*		*fPSA (ng ml^{-1})*		*%fPSA*	
		Mean±s.d.	*95% CI*	*Mean±s.d.*	*95% CI*	*Mean±s.d.*	*95% CI*
10–19	42	0.43±0.06	0.30–0.56	0.17±0.03	0.12–0.23	0.57±0.14	0.27–0.86
20–29	54	0.90±0.10	0.70–1.10	0.32±0.02	0.27–0.37	0.41±0.03	0.35–0.46
30–39	56	0.96±0.11	0.73–1.20	0.33±0.04	0.25–0.40	0.49±0.11	0.26–0.71
40–49	64	1.00±0.10	0.79–1.21	0.34±0.03	0.28–0.39	0.49±0.10	0.29–0.69
50–59	58	1.15±0.10	0.95–1.34	0.41±0.03	0.34–0.47	0.37±0.02	0.33–0.42
60–69	62	1.13±0.14	0.85–1.42	0.37±0.04	0.28–0.46	0.45±0.10	0.25–0.65
70–79	68	1.39±0.16	1.07–1.71	0.40±0.04	0.33–0.48	0.42±0.09	0.24–0.60
80–89	38	1.31±0.24	0.81–1.81	0.46±0.09	0.28–0.65	0.38±0.02	0.33–0.43
Total	442	1.05±0.05	0.96–1.15	0.41±0.02	0.35–0.01	0.45±0.03	0.39–0.51
F		4.376		3.923		0.468	
P		0.000		0.000		0.857	

s.d.: standard deviation; PSA: prostate-specific antigen; tPSA: total prostate-specific antigen; fPSA: free prostate-specific antigen; %fPSA: percentage of free prostate-specific antigen; CI: confidence interval

Table 2: p2PSA, %p2PSA, p2PSA/tPSA and PHI levels

Age (year)	*n*	*p2PSA (pg ml^{-1})*		*%p2PSA*		*p2PSA/tPSA*		*PHI*	
		Mean±s.d.	*95% CI*	*Mean±s.d.*	*95% CI*	*Mean±s.d.*	*95% CI*	*Mean±s.d.*	*95% CI*
10–19	42	3.44±0.58	2.22–4.66	22.41±3.57	14.91–29.92	9.11±1.23	6.53–11.68	17.20±4.09	8.64–25.76
20–29	54	4.85±0.52	3.78–5.91	18.18±2.60	12.82–23.53	6.76±1.02	4.67–8.85	15.83±3.43	8.66–23.00
30–39	56	6.86±1.07	4.66–9.07	19.36±3.05	12.80–34.87	6.65±1.21	4.16–9.15	31.83±10.29	10.29–53.37
40–49	64	5.24±0.55	4.13–6.36	18.33±2.39	13.45–23.21	6.49±0.79	4.86–8.11	9.16±2.05	8.19–16.76
50–59	58	9.93±0.88	8.13–11.74	26.65±3.20	20.10–33.20	9.71±1.16	7.33–12.08	30.47±3.83	22.42–38.52
60–69	62	9.07±0.80	7.44–10.69	47.54±14.12	18.74–76.33	8.65±0.90	6.79–10.50	42.85±9.06	23.96–61.75
70–79	68	10.46±1.29	7.84–13.07	35.48±10.44	14.36–56.60	8.15±1.07	5.98–10.32	32.11±6.12	18.99–45.23
80–89	38	13.33±2.55	8.04–18.63	31.68±6.56	18.08–45.27	9.14±1.22	6.61–11.67	31.17±5.19	20.08–42.27
Total	442	8.11±0.45	7.22–9.01	32.44±3.88	24.79–40.09	8.05±0.38	7.29–8.80	26.33±3.93	17.03–35.62
F		2.733		0.963		1.363		2.536	
P		0.000		0.624		0.222		0.013	

s.d.: standard deviation; CI: confidence interval; p2PSA: [-2]pro prostate-specific antigen; %p2PSA: percentage of [-2]pro prostate-specific antigen; tPSA: total prostate-specific antigen; PHI: prostate health index

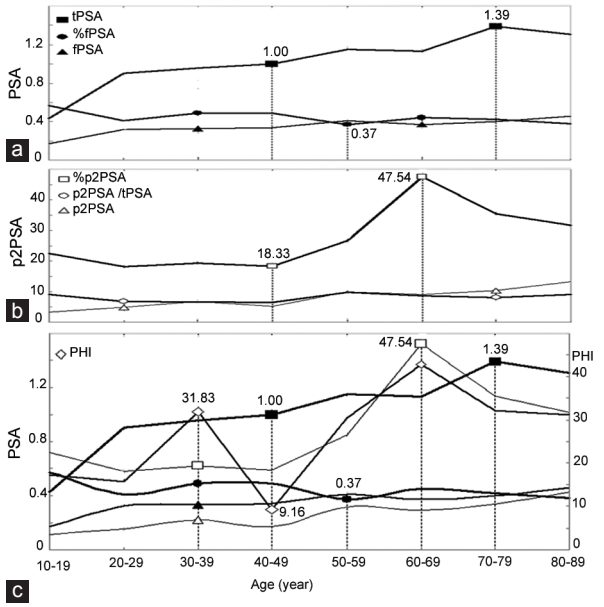

Figure 2: The age-related distribution curves of PSA (**a**), p2PSA (**b**) and PHI (**c**).

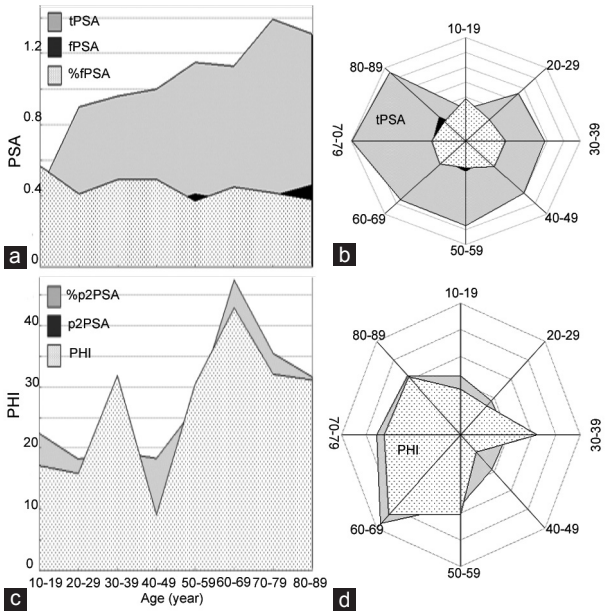

Figure 3: (**a**) The relationships between PSA and ages. (**b**) The outstanding clinical values of tPSA. (**c**) The relationships between PHI, p2PSA and ages. (**d**) The potential clinical values of PHI.

40–49 years (PHI = 9.16 ± 2.05, 95% CI = 8.19–16.76), concordant with the trough of %p2PSA when tPSA increased to 1.00 ng ml^{-1}.

%p2PSA and PHI both reached their lifetime peaks at 60–69 years (47.54 and 42.85, respectively), whereas tPSA reached its lifetime peak at 70–79 years (1.39). After the age of 69 years, %p2PSA and PHI began to decrease, but tPSA continued to increase until 70–79 years and then decreased (**Figure 2c**). Therefore, after 69 years of age, similar to %p2PSA, PHI also decreased approximately 10 years before tPSA (**Figure 2a–2c**). As the normal physiological reduction in %p2PSA and PHI occurred 10 years before the decrease in tPSA, these two parameters may be potentially used as earlier markers of PCa, approximately 10 years before PSA. It is worth noting that there is a very high incidence of PCa in men aged 60–79 years.

The value of PCa risk assessment combining PSA, %p2PSA, and PHI in patients aged 69–79 years might be inferred from the data in **Table 3**. If PSA, %p2PSA, and PHI values show declining trends, the possibility of PCa can be ruled out. If PSA shows a mild increase every year, but %p2PSA and PHI show decreases, this may be considered an uncertain result. This is because at this time, PCa development or recurrence may not necessarily be present, and the finding may only be an artifact of an age-related increase in PSA. Investigation of clinical symptoms and radiological imaging are required to determine the exact diagnosis. When necessary, follow-up monitoring and regular scheduled examinations may also be suggested. If PSA decreases but %p2PSA and PHI increase every year, this is a paradoxical result and the patient should be reexamined and regularly followed up. If PSA, %p2PSA, and PHI all show increased levels every year, the possibility of PCa should be highly suspected. At this time, clinical symptoms and radiological imaging should be used in combination. If necessary, biopsy should be performed as soon as possible to confirm the diagnosis.

The coverage and radiance of tPSA as a marker included almost all ages except childhood, suggesting that tPSA is more representative of prostate function than fPSA and %fPSA, especially at older ages (**Figure 3a** and **3b**). The highest covering power and radiometric force of tPSA were noted in the age group of 70–79 years, indicating that tPSA can be used as the main marker of prostate function.

Table 3: PCa risk for 69–79 years old

PSAV	*%p2PSAV*	*PHIV*	*PCa possibility*
–	–	–	Normal
+	–	–	Dubious
–	+	+	Follow-up
+	+	+	High suspicion

–: decreased velocity; +: increased velocity; PSAV: prostate-specific antigen velocity; %p2PSAV: percentage of [-2]pro prostate-specific antigen velocity; PHIV: prostate health index velocity; PCa: prostate cancer

However, the coverage rate and radiance of PHI and %p2PSA markers surpassed those of the other markers including tPSA, fPSA, %fPSA, and p2PSA, suggesting that PHI and %p2PSA are possibly superior to the other markers. The most representative and valuable age-points were 30–39 years for PHI and 60–69 years for %p2PSA (**Figure 3c** and **3d**).

DISCUSSION

In general, elderly patients experience a higher incidence of PCa, which has become the second leading cause of death in some countries.[3,4] As a result, research efforts toward screening and early detection of PCa have increased greatly. PSA has become the principal biomarker for detecting PCa and guiding clinical decisions. This is unfortunate because the PSA test lacks both adequate sensitivity and specificity to differentiate PCa from BPH. Patients with benign diseases are often subjected to unnecessary examinations, overtreatment, biopsies, or radical prostatectomies.[5] The flaws of PSA testing as a diagnostic tool have been frequently reported in older patients (>40 years old).[6,7]

This study was designed to compare seven key PCa markers and their respective relationships with age in healthy subjects. Our intent was to acquire preliminary data to further our research by next comparing these same parameters in patients with PCa and BPH. The serum samples in our study were collected and processed in a single day to reduce sample degradation.[8,9] Samples were analyzed by electrochemical luminescence immunoassay, instead of previously reported enzyme-linked immunoassay techniques.[10,11] In addition, 34 men were not included in our analysis as they had elevated baseline tPSA levels.

Given the changes in testicular function and androgen levels observed with male aging, our study demonstrated that tPSA is one of the most influential markers ($P < 0.001$). Patient age may be one of the main factors influencing the sensitivity and specificity of tPSA in clinical diagnosis. Therefore, the age of the patient should always be considered when reviewing PSA results, and an age-based reference standard should be employed.

Na *et al*. have also previously reported that tPSA values in patients with PCa correlated with age ($P = 0.008$).[12] However, previous studies did not include subjects younger than 40 years of age; therefore, their comparisons and age-related curves were difficult to draw and thus could not be compared with our data. The tPSA values in healthy men from Syria were reported as 1.7 ng ml^{-1} at 40–49 years and 5.8 ng ml^{-1} at 70–79 years.[13] In Canada, the 75th percentiles for the median tPSA level in healthy men aged 40–49, 50–59, 60–69, and 70–79 years were 1.1, 1.4, 2.6, and 3.6 ng ml^{-1}, respectively.[14] However, for healthy Chinese men in Beijing, tPSA was reported as 1.0 ± 0.6, 1.0 ± 0.7, 1.1 ± 0.7, and 1.3 ± 1.1 ng ml^{-1} in men aged 60-, 70-, 80-, and 90-year-old, respectively,[15] showing values similar to our data. Thus, Syrian and Canadian men possibly have higher baseline PSA values compared with Chinese men, which could explain the lower morbidity in China related to PCa.[16]

In our study, after the age of 79 years, fPSA continuously increased, while tPSA declined (**Figure 2a**). Therefore, after 79 years of age, if a patient presents with a dynamic increase in tPSA and decrease in fPSA, the possibility of PCa should be suspected. On the other hand, tPSA combined with fPSA can have greater diagnostic value than tPSA alone, which supports our conclusions that fPSA could reflect prostate volume better than tPSA.[17] Moreover, the lowest level of %fPSA (0.37) occurred at 50–59 years. Thus, a cutoff of %fPSA <0.37 should be considered a signal for the possibility of PCa in Chinese men. In light of the features of the %fPSA curve, %fPSA velocity values should be monitored along with PSA velocity.[18,19]

Fossati *et al*. reported that values for p2PSA and %p2PSA of PCa patients under the age of 60 years were significantly higher than those without cancer ($P < 0.0001$).[20] An Italian study based on 264 cases of diagnosed PCa patients showed a mean p2PSA and %p2PSA of approximately 15.0 pg ml^{-1} and 2.1 pg ml^{-1}, respectively.[21] Our data also indicated that the p2PSA and %p2PSA values in healthy men in China change with age. At 40–49 years, when tPSA increased to 1.00 ng ml^{-1}, %p2PSA decreased to its lowest point. After this age, %p2PSA approached its peak (47.54 ± 14.12) at 60–69 years. Therefore, it can be inferred that monitoring %p2PSA could better reflect prostate volume hyperplasia as well as distinguishing PCa and cases with poor prognosis.[22–24]

While some men over the age of 40 years may experience some early signs of BPH, such as urine voiding issues, constant urination stimulation, frequent nocturia, and delayed urine voiding, previous reports have sometimes attributed these symptoms to chronic prostatitis. In fact, treatment with certain alpha-1 adrenoceptor blockers often provided satisfactorily results. Moreover, these men have been found to have minor BPH with middle lobe gland hyperplasia on transurethral cystoscopy. Our study also suggested the importance of the age of 40–49 years, when the mean level of tPSA increased to 1.00 ng ml^{-1}, and PHI and %p2PSA curves simultaneously dropped to their lowest levels, suggesting that 40–49 years could be a critical age point. This is supported by histological data showing that BPH usually occurs after the age of 40 years.[25]

After the age of 40–49 years, in spite of gradually decreasing androgen levels resulting from the age-related decline in the function of the hypothalamic-pituitary-gonadal axis,[26] PHI and %p2PSA levels increased more quickly than PSA values, which could be attributed to functional compensation of the prostate through hypertrophy and interstitial hyperplasia as described above, and based on histological data.

Recent reports suggest that %p2PSA and PHI may have better clinical value than conventional PSA alone.[3,4] Fossati *et al*. reported that PHI was more accurate than PSA in predicting the possibility of PCa in men aged <60 years.[20] Unfortunately, their study did not include men of all ages, and the detailed merits of these two factors and the reasons underlying their significance remain unknown. The present study not only suggested that %p2PSA and PHI are probably better markers than tPSA in the age group of 60–79 years but also indicated the possible reasons underlying this advantage. This is of particular significance since the highest incidence of PCa occurs at 60–79 years. After 69 years of age, %p2PSA and PHI showed age-related decreases in their values, i.e., approximately 10 years earlier than the decrease in tPSA, which occurred after 79 years of age. From the perspective of age, %p2PSA and PHI may represent earlier indicators of PCa than tPSA, predating it by approximately 10 years. Simultaneous comparison of PSAV, %2PSAV, and PHIV would probably enable improved sensitivity and specificity of PCa screening and more accurately identify non-PCa patients to avoid unnecessary biopsy and/or repeat biopsy.

CONCLUSION

This comprehensive analysis of seven prostate-related markers, namely of tPSA, fPSA, %fPSA, p2PSA, %p2PSA, and PHI, contributes to the general understanding of prostate maturation, function compensation, and decompensation changes in healthy men over their lifetimes. The age of 40–49 years is an important age point for tPSA, %p2PSA, and PHI, suggesting that these may represent important clinical parameters that warrant further examination. In addition, %p2PSA and PHI may have high clinical value as earlier markers than tPSA by approximately 10 years. Further studies are needed to compare the findings for these seven markers in patients diagnosed with PCa and in healthy subjects.

AUTHOR CONTRIBUTIONS

WGS designed the project, performed the statistical analysis, and wrote the paper; CZL provided overall technique directions; QCZ and XWH provided some support in each department; and WGS, ZZL, and PW carried out all the experiments. All authors have read and approved the final manuscript.

COMPETING INTERESTS

The authors declare no competing interests.

ACKNOWLEDGMENTS

This study was supported by Anhui Medical University, Tongji Medical College, Ma'anshan Municipal Science and Technology Bureau (Technique subject number: 2012-K-Z-02; 2013-Y-5).

REFERENCES

1 Tosoian JJ, Ross AE, Sokoll LJ, Partin AW, Pavlovich CP. Urinary biomarkers for prostate cancer. *Urol Clin North Am* 2016; 43: 17–38.
2 Huang YQ, Sun T, Zhong WD, Wu CL. Clinical performance of serum [-2]proPSA derivatives, %p2PSA and PHI, in the detection and management of prostate cancer. *Am J Clin Exp Urol* 2014; 2: 343–50.
3 Loeb S. Time to replace prostate-specific antigen (PSA) with the prostate health index (PHI)? Yet more evidence that the PHI consistently outperforms PSA across diverse populations. *BJU Int* 2015; 115: 500.
4 Mirandola L, Figueroa JA, Phan TT, Grizzi F, Kim M, *et al*. Novel antigens in non-small cell lung cancer: SP17, AKAP4, and PTTG1 are potential immunotherapeutic targets. *Oncotarget* 2015; 6: 2812–26.

5 Lee AG, Choi YH, Cho SY, Cho IR. A prospective study of reducing unnecessary prostate biopsy in patients with high serum prostate-specific antigen with consideration of prostatic inflammation. *Korean J Urol* 2012; 53: 50–3.
6 Yang X, Monn MF, Liu L, Liu Y, Su J, *et al*. Incidental prostate cancer in Asian men: high prevalence of incidental prostatic adenocarcinoma in Chinese patients undergoing radical cystoprostatectomy for treatment of bladder cancer and selection of candidates for prostate-sparing cystectomy. *Prostate* 2015; 75: 845–54.
7 Arafa MA, Farhat K, Aqdas S, Al-Atawi M, Rabah DM. Assessment of lower urinary tract symptoms in Saudi men using the international prostate symptoms score. *Urol Ann* 2015; 7: 221–5.
8 Liu GR, Luo B, Zheng T, Yan XL, Li XJ, *et al*. Influence of serum storage on the laboratory results of prostate-specific antigen. *Zhonghua Nan Ke Xue* 2013; 19: 637–41.
9 Sağlam HS, Köse O, Ozdemir F, Adsan O. Effect of heamolysis on prostate-specific antigen. *ISRN Urol* 2012; 12: 729821.
10 Jiang YQ, Xie ZX, Liu Q, Liu TY, Ma ZR, *et al*. Comparison and analysis of two methods for serum free prostate-specific antigen (PSA) detection. *Zhonghua Shi Yan He Lin Chuang Bing Du Xue Za Zhi* 2012; 26: 316–8.
11 Wang Y, Zhang XF, Dai J, Zheng YC, Zhang MG, *et al*. Predictive value of serum bone sialoprotein and prostate-specific antigen doubling time in patients with bone metastasis of prostate cancer. *J Huazhong Univ Sci Technol Med Sci* 2013; 33: 559–62.
12 Na R, Wu Y, Xu J, Jiang H, Ding Q. Age-specific prostate specific antigen cutoffs for guiding biopsy decision in Chinese population. *PLoS One* 2013; 8: e67585.
13 Bakir MA, Abo-daher D. Age-specific reference ranges for prostate-specific antigen among healthy Syrian men. *Int J Biol Markers* 2012; 27: e152–9.
14 Capitanio U, Perrotte P, Zini L, Suardi N, Antebi E, *et al*. Population-based analysis of normal total PSA and percentage of free/total PSA values: results from screening cohort. *Urology* 2009; 73: 1323–7.
15 Shao Q, Guo Y, Guo H. A preliminary study of PSA level in 646 healthy men in Beijing. *Zhonghua Yi Xue Za Zhi* 2000; 80: 591–2.
16 Na R, Ye D, Liu F, Chen H, Qi J. Performance of serum prostate-specific antigen isoform [-2]proPSA (p2PSA) and the prostate health index (PHI) in a Chinese hospital-based biopsy population. *Prostate* 2014; 74: 1569–75.
17 Choi H, Park JY, Shim JS, Kim JH, Bae JH. Free prostate-specific antigen provides more precise data on benign prostate volume than total prostate-specific antigen in Korean population. *Int Neurourol J* 2013; 17: 73–7.
18 Sasaki M, Ishidoya S, Ito A, Saito H, Yamada S, *et al*. Low percentage of free prostate-specific antigen (PSA) is a strong predictor of later detection of prostate cancer among Japanese men with serum levels of total PSA of 4.0 ng/mL or less. *Urology* 2014; 84: 1163–7.
19 Vasarainen H, Salman J, Salminen H, Valdagni R, Pickles T, *et al*. Predictive role of free prostate-specific antigen in a prospective active surveillance program (PRIAS). *World J Urol* 2015; 33: 1735–40.
20 Fossati N, Lazzeri M, Haese A, McNicholas T, de la Taille A, *et al*. Clinical performance of serum isoform [-2]proPSA (p2PSA), and its derivatives %p2PSA and the prostate health index, in men aged<60 years: results from a multicentric European study. *BJU Int* 2015; 115: 913–20.
21 Lazzeri M, Haese A, de la Taille A, Palou Redorta J, McNicholas T, *et al*. Serum isoform [-2]proPSA derivatives significantly improve prediction of prostate cancer at initial biopsy in a total PSA range of 2-10 ng/ml: a multicentric European study. *Eur Urol* 2013; 63: 986–94.
22 Lazzeri M, Haese A, Abrate A, de la Taille A, Redorta JP, *et al*. Clinical performance of serum prostate-specific antigen isoform [-2]proPSA (p2PSA) and its derivatives, %p2PSA and the prostate health index (PHI), in men with a family history of prostate cancer: results from a multicentre European study, the PROMEtheuS project. *BJU Int* 2013; 112: 313–21.
23 Filella X, Giménez N. Evaluation of [-2]proPSA and prostate health index (phi) for the detection of prostate cancer: a systematic review and meta-analysis. *Clin Chem Lab Med* 2013; 51: 729–39.
24 Abrate A, Lazzeri M, Lughezzani G, Buffi N, Bini V, *et al*. Clinical performance of the prostate health index (PHI) for the prediction of prostate cancer in obese men: data from the PROMEtheuS project, a multicentre European prospective study. *BJU Int* 2015; 115: 537–45.
25 Pereira JA, Bilhim T, Duarte M, Rio Tinto H, Fernandes L, *et al*. Patient selection and counseling before prostatic arterial embolization. *Tech Vasc Interv Radiol* 2012; 15: 270–5.
26 Inci M, Rifaioglu MM, Inci M, Celik M, Demir M, *et al*. The investigation of total PSA, free PSA, and free/total PSA ratio in patients with liver cirrhosis patients according to Child-Pugh score. *Urology* 2013; 81: 617–22.