

The Periurethral Glands do not Significantly Influence the Serum Prostate Specific Antigen Concentration

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Abstract

Purpose: The periurethral glands are known to produce prostate specific antigen (PSA). With ultra-sensitive assays now routinely available, it is necessary to determine if the periurethral glands significantly influence serum PSA concentration after radical prostatectomy.

Materials and Methods: Serum PSA levels of 46 men, 51 to 89 years old (median age 67) who underwent radical cystoprostatectomy and total urethrectomy, were compared with those of 92 men 46 to 91 years old (median age 67) who underwent radical cystoprostatectomy only. All men had transitional cell carcinoma of the bladder without gross or microscopic evidence of prostate cancer and all underwent ileal conduit diversion. Serum was obtained at least 1 year postoperatively. Each specimen was analyzed using the Tosoh† Immulite‡ and Yu and Diamandis ultra-sensitive PSA assays with analytical detection limits of 0.02 ng./ml., 0.004 ng./ml. and 0.002 ng./ml., respectively.

Results: Median PSA for the radical cystoprostatectomy with urethrectomy group was 0.00 ng./ml. (range 0.00 to 0.14) for each of the 3 assays. For the radical cystoprostatectomy only group the median Tosoh and Immulite PSA assay levels were 0.01 ng./ml. (range 0.00 to 0.22), and median Yu and Diamandis PSA assay level was 0.00 ng./ml. (range 0.00 to 0.31).

Conclusions: The greatest difference in median PSA levels that could be found between men with and without periurethral glands when using 3 different ultra-sensitive assays was 0.01 ng./ml., indicating that the periurethral glands do not have a clinically significant effect on serum PSA concentration after radical prostatectomy. Thus, a serum PSA level above the residual cancer detection limit following radical prostatectomy, even if obtained with an ultra-sensitive assay, reflects either malignant or benign residual prostatic tissue, rather than the presence of periurethral glands.

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Key Words: prostate-specific antigen, urethra, bladder neoplasms, prostatic neoplasms

Several reports have suggested that extraprostatic sources of prostate specific antigen (PSA) production exist.[1-3] Kamoshida and Tsutsumi demonstrated PSA in periurethral glands using immunoperoxidase staining[2] as did Frazier et al.[3] Iwakiri et al characterized PSA in the urine of men before and after radical prostatectomy in an effort to determine the role of urinary PSA in detecting locally recurrent prostate cancer.[1] Thus, it is well known that the periurethral glands secrete PSA in the urine. To our knowledge the level of PSA secretion by the periurethral glands into the serum has not been characterized to date. However, with the advent of ultra-sensitive assays this issue may become important and clinically relevant.

Ultra-sensitive PSA assays have been developed in an effort to detect extremely low levels of PSA in the serum. The 3 assays used in our report are the Tosoh assay,[4] which is currently available for widespread clinical use, as well as the Immulite,[5] and Yu and Diamandis assays,[6] which are being used under investigational protocols. Analytical detection limits with these assays are 0.02 ng./ml., 0.004 ng./ml. and 0.002 ng./ml., respectively. The residual cancer detection limit, defined as 3 standard deviations above the mean of sera obtained from men known to be cured by radical prostatectomy, is 0.07 ng./ml. for the Tosoh assay,[4] unknown for the Immulite assay, and 0.09 ng./ml. for the Yu and Diamandis assay based on 30 serum specimens from Stanford University.

The facts that the periurethral glands produce PSA and low levels of PSA can be detected in the serum imply that detectable levels of PSA after radical prostatectomy may be from 1 of 3 sources: residual benign prostate tissue, persistent prostate cancer or the periurethral glands. This possible lack of specificity of serum PSA at low levels may become a significant issue in the future as newer treatment options for residual or persistent prostate cancer become available. Our study was designed to answer the clinically important question of whether the periurethral glands influence the serum PSA level in post-prostatectomy patients by comparing postoperative sera from large numbers of cystoprostatectomy patients with and without urethrectomy as suggested in a previous ultra-sensitive assay study.[4]

Material and Methods

Patient population

Two groups of patients were identified from the Mayo Clinic tumor registry: group 1--men who had undergone radical cystoprostatectomy and total urethrectomy muscle invasive transitional cell carcinoma of the bladder (age range 51 to 89 years at PSA determination, mean plus or minus standard deviation 73 plus/minus 8.7 and mean age at cystoprostatectomy 67) and group 2--age-matched controls who had undergone radical cystoprostatectomy only for muscle invasive transitional cell carcinoma of the bladder (age range 46 to 91 years, mean 73 plus/minus 9.3 and mean age at cystoprostatectomy 67). All patients underwent ileal conduit urinary diversion. Exclusion criteria were a simultaneous diagnosis of prostate cancer on pathological examination of the surgical specimen (determined by step-sectioning), urinary diversion with an orthotopic bladder or continent reservoir, or any patient receiving androgen deprivation therapy (bilateral orchiectomy, luteinizing hormone-releasing hormone agonist, estrogen or antiandrogen therapy). Men who had undergone alternative urinary diversions were excluded to control for the possible trophic effect of urine flow upon the periurethral glands. After internal review board approval was obtained, 170 men were contacted by mail and 138 (81 percent) agreed to participate in the study, with 46 in group 1 and 92 in group 2 (2 controls for each study patient). The patients were matched on the basis of age and year of cystoprostatectomy. The same surgeons performed the operations on all men in each group.

Specimen collection

Venipuncture kits were sent to all men, and blood was drawn at local medical facilities and shipped to Stanford University on dry ice. Serum specimens were collected from April to July 1994, stored at -70F and analyzed in triplicate during a 3-day interval with the Tosoh, Immulite, and Yu and Diamandis ultra-sensitive PSA assays.

Assays

The Tosoh PSA assay is an automated 2-site immunoenzymometric assay that uses monoclonal antibody immobilized on a magnetic solid phase and an enzyme-labeled monoclonal antibody to bind PSA. The unbound enzyme labeled monoclonal antibody is then removed and the bound fraction is incubated with a fluorogenic substance. The amount of bound enzyme labeled antibody indicates the serum PSA concentration.

The intra-assay coefficient of variation is 14.1 percent and the inter-assay coefficient of variation is 26.3 percent at a PSA of 0.11 ng./ml. The analytical sensitivity of the Tosoh PSA assay is 0.02 ng./ml., which was determined by running the 0 calibrator 20 times in the ultra-sensitive format and represents the addition of 2 standard deviations above the mean.

The Immulite third generation PSA assay is a solid-phase chemiluminescent assay in which the solid phase, a polystyrene bead, is coated with a monoclonal antibody. The serum sample is incubated with the beads and unbound serum is removed by centrifugal washing. An alkaline phosphatase labeled polyclonal antibody is introduced. The sustained emission of light from the chemiluminescent substrate is quantified and is proportional to the serum level of PSA. The intra-assay coefficient of variation is 3.8 percent at a PSA of 0.08 ng./ml. and the inter-assay coefficient of variation is 5.2 percent at a PSA of 0.30 ng./ml. The analytical sensitivity of this assay is 0.004 ng./ml.[5]

The Yu and Diamandis PSA assay is an investigational immunofluorometric assay using a monoclonal capture antibody and a polyclonal detection antibody. Highly fluorescent complexes are formed with terbium and are quantified using a microsecond time-resolved fluorometer. The intra-assay coefficient of variation is 21.4 percent at a PSA of 0.016 ng./ml. and the inter-assay coefficient of variation is 14.5 percent at a PSA of 0.20 ng./ml.[6] The analytical sensitivity of this assay is 0.002 ng./ml.

Statistical analysis

The individual PSA values analyzed for each assay were based on the mean of 3 replicate readings for each specimen. The rank sum test was used to evaluate the difference between the PSA levels obtained for the 2 groups, that is men with and without periurethral glands. All tests were 2-sided with an alpha level of 0.05. The Wilcoxon signed rank test was used to compare PSA values among the 3 assays.

Results

Tosoh PSA assay

Median PSA level in group 1 was 0.00 plus/minus 0.02 ng./ml. (range 0.00 to 0.11) compared to 0.01 plus/minus 0.03 ng./ml. (range 0.00 to 0.22) in group 2 (see Table 1). More than 90 percent of men in both groups had a PSA level of less than 0.05 ng./ml.

	Tosoh PSA Assay		Immulite Assay		Yu and Diamandis Assay ⁶	
	Group 1	Group 2	Group 1	Group 2	Group 1	Group 2
PSA (ng./ml.):						
Mean ± SD	0.01 ± 0.02	0.02 ± 0.03	0.01 ± 0.03	0.01 ± 0.03	0.02 ± 0.03	0.02 ± 0.05
Median	0.00	0.01	0.00	0.01	0.00	0.00
Range	0.00-0.11	0.00-0.22	0.00-0.11	0.00-0.22	0.00-0.14	0.00-0.31
% Distribution:						
0.00 -0.01	79	67	91	77	78	76
0.02 -0.03	15	20	2	13	4	9
0.04 -0.05	2	8	0	4	2	2
More than 0.05	4	5	7	6	16	13

Table 1. PSA assay results

Immulite PSA assay

Median PSA level in group 1 was 0.00 plus/minus 0.03 ng./ml. (range 0.00 to 0.11) compared to 0.01 plus/minus 0.03 ng./ml. (range 0.00 to 0.22) in group 2 (see Table 1). Again, more than 90 percent of men in both groups had a PSA level of less than 0.05 ng./ml.

Yu and Diamandis PSA assay

When using the Yu and Diamandis assay, median serum PSA level in group 1 was 0.00 plus/minus 0.03 ng./ml. (range 0.00 to 0.14) compared to 0.00 plus/minus 0.05 ng./ml. (range 0.00 to 0.31) in group 2. With this assay more than 80 percent of men in both groups had a PSA of 0.05 ng./ml. or less (see Table 1).

Using the Tosoh assay, a small but statistically significant ($p = 0.003$) difference in median PSA between the 2 groups was noted. A similar observation was made when using the Immulite PSA assay ($p = 0.0005$). However, with the Yu and Diamandis assay no significant difference between the median PSA values of the 2 groups was observed ($p = 0.36$).

When the results obtained with the 3 assays were compared, the Tosoh and Immulite assays were similar (mean difference 0.004 ng./ml., p less than 0.0001). The Yu and Diamandis values averaged 0.006 ng./ml. higher than the Tosoh levels. However, the difference was not significant using the Wilcoxon signed rank test ($p = 0.73$). The Yu and Diamandis assay values averaged 0.010 ng./ml. higher than the Immulite PSA values ($p = 0.007$). For all 3 comparisons 88 to 97 percent of the differences found between assays were within 0.03 ng./ml. or less.

Discussion

PSA is the most useful tumor marker available to date for the treatment of prostate cancer,[7] particularly in the post-radical prostatectomy period when the serum PSA level should theoretically become undetectable.[8-10] Until now, a serum PSA of greater than 0.1 ng./ml. after curative resection has been considered by many to indicate recurrent or persistent cancer.[1,2,9-11] With the new ultra-sensitive PSA assays that detect PSA concentrations 1 order of magnitude lower than standard assays, recurrent or residual prostate cancer may possibly be detected sooner and at a lower PSA level, which will become even more important clinically as more treatment options for residual or recurrent prostate cancer emerge. However, this result is based on the assumption that all detectable PSA is produced solely by prostatic tissue. Several recent reports have indicated that PSA is not produced in an organ specific manner.

Kamoshida and Tsutsumi used indirect immunoperoxidase staining to demonstrate PSA in the periurethral glands of 3 men and 2 of 3 women.[2] Frazier et al evaluated 20 male urethral specimens for PSA and prostatic acid phosphatase (PAP) using immune staining.[3] Of 20 specimens 8 (40 percent) demonstrated strong staining for both markers localized to the periurethral glands. Five of 17 urethral biopsies (29 percent)

were positive for PSA and PAP staining, and all 3 whole-mount autopsy urethral specimens stained positively for PSA and PAP throughout the length of the urethra.

Iwakiri et al studied 48 patients to determine the role of urinary PSA as a potential tumor marker for locally recurrent prostate cancer after radical prostatectomy.[1] They found substantial urinary PSA levels in all patients studied, including those with prostate cancer who had undergone curative radical prostatectomy and those without prostate cancer who had undergone radical cystoprostatectomy with orthotopic bladder substitution. The difference in the preoperative and postoperative levels of urinary PSA was not statistically significant, which implied that the urethra may be the source of the PSA detected in the urine. They hypothesized that low levels of serum PSA detected in cured prostate cancer patients may be from the periurethral glands.[1]

If the periurethral glands contribute significantly to serum PSA at as low levels as detected by ultra-sensitive assays, the value of PSA determination in the posttreatment period could become less sensitive for indicating residual or recurrent disease. Our study was designed to address this important issue of how the periurethral glands influence serum PSA in patients who have undergone radical prostatectomy. In our investigation 61 to 83 percent of the serum samples from patients who had undergone radical cystoprostatectomy alone or with total urethrectomy and ileal loop diversion for transitional cell carcinoma had a serum PSA of 0.00 ng./ml. when analyzed by 3 different ultra-sensitive PSA assays, 67 to 91 percent had PSA of 0.01 ng./ml. or less and only 7 to 20 percent had PSA of 0.03 ng./ml. or greater. Median PSA values in the 2 groups differed by only 0.01 ng./ml. with the Tosoh and Immulite assays, and was 0.00 ng./ml. in both groups when using the Yu and Diamandis assay. With these ultra-sensitive assays the greatest difference in mean and median PSA values for men with and without periurethral glands was 0.01 ng./ml. The inter-assay and intra-assay coefficients of variation for these assays at as low PSA levels as detected in our study range from 3.8 to 26.3 percent, implying that some difference may be related to assay variability. Also, there is a possibility that some of these men with higher PSA values may have residual apical prostate tissue. While the difference in the serum PSA values for the 2 groups was statistically significant (p less than 0.003) in 2 (Tosoh and Immulite) of 3 ultra-sensitive assays, it appears that the periurethral glands do not have a clinically significant effect on serum PSA concentration.

Conclusions

A measurable serum PSA value following radical prostatectomy, even if obtained with an ultra-sensitive assay, is accurate and reliable, and reflects either malignant or benign residual prostatic tissue. With our results the clinician can feel comfortable attributing a detectable serum PSA level in a patient who has undergone radical prostatectomy to residual or recurrent disease rather than a false-positive result from the periurethral glands. However, before initiating adjuvant therapy based on a solitary PSA value, it is advisable in individual patients to seek a trend toward increasing PSA.

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