Increased Serum Level of Early Prostate Cancer Antigen (EPCA) Is Associated with Subsequent Cancer Risk in Men with High Grade Prostatic Intraepithelial Neoplasia

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Running Title: EPCA Associated with PCa Onset in HGPIN

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Abstract

Early prostate cancer antigen (EPCA) has been recently suggested as a novel biomarker in malignant and premalignant lesions of the prostate. This study was to examine serum expression of EPCA, and to further clarify the relationship between initial serum EPCA levels and the presence of subsequent cancer in the individuals with isolated high-grade prostatic intraepithelial neoplasia (HGPIN). An indirect ELISA test was used for initial serum EPCA measurement in 112 men with isolated HGPIN, who were enrolled and completed a follow-up of ≥5 years. All patients had a detectable concentration of EPCA in the initial serum, with a mean of 0.64±0.13 absorbance at 450 nm. Thirty-three patients had an initial serum EPCA level of ≥1.10, in which 31 cases were subsequently identified as having PCa on follow-up. However, in the remaining 79 cases, serum EPCA levels were all <1.10 and none was diagnosed with cancer later. Statistical analysis showed a significantly higher serum ECPA level in isolated HGPIN with subsequent cancer than those without (P<0.001). The area under the receiver operating characteristic (ROC) curves showed that serum EPCA level had better predictive accuracy of cancer onset on follow-up than PSA velocity and abnormal DRE findings. Furthermore, univariate and multivariate Cox regression analyses demonstrated the independently predictive performance by initial serum EPCA ≥1.10 absorbance (relative risk: 3.32, 95%CI: 2.62 - 5.03, P <0.001). These preliminary findings firstly show the potential of serum EPCA to serve as a significant predictor for subsequent cancer in isolated HGPIN.
**Keywords:** Early prostate cancer antigen (EPCA); Prostatic intraepithelial neoplasia (PIN); Prostate cancer (PCa); Enzyme linked immunosorbent assay (ELISA); Tumor markers
Introduction


The alterations in the protein components of the nuclear structure have been associated with carcinogenesis in a variety of tissues, including prostate. Thus, nuclear matrix proteins have been initially identified with which it is possible to differentiate between normal prostate and prostatic carcinoma. Early prostate cancer antigen (EPCA), a nuclear matrix protein recently discovered by Dhir et al. (2004), was identified as being expressed throughout the prostate of patients with PCa but not in those without the disease. A recent enzyme linked immunosorbent assay (ELISA) for plasma EPCA showed, using a cutoff of 1.7 absorbance at 450 nm, that EPCA had a sensitivity of 92% (11/12) for patients with PCa, a specificity of 100% (16/16) for healthy donors, and an overall specificity of 94% (32/34) for entire donor controls (Paul et al. 2005). Most recently, EPCA-2.22, one of three sequenced epitopes of the EPCA-2 (another nuclear structural protein which is unrelated to EPCA), has been
identified as having a 92% specificity for healthy men and men with benign prostatic hyperplasia and 94% sensitivity for overall PCa patients using a cutoff value of 30 ng/ml (0.8 absorbance at 630 nm) (Leman et al. 2007). EPCA-2.19, the second epitope of EPCA-2, also has a specificity of 94% and a sensitivity of 91% in separating normal men with PSA < and >2.5 ng/ml, and men with BPH from those with PCa at a cutoff of 0.5 ng/ml and above (Leman et al. 2009). These data have shown the potential of EPCA and EPCA-2 to serve as promising serum biomarkers for the detection of PCa. The positive immunostaining for EPCA protein was noted not only in the benign glands of the negative biopsies from men who, as much as 5 years later, were eventually detected as having prostate carcinoma, but also in HGPIN in foci adjacent to PCa (Dhir et al. 2004, Uetsuki et al. 2005). These findings suggest that up-regulation of EPCA may be involved in the presumably earlier stage of prostatic carcinogenesis and that EPCA could be used as a potential predictive marker. However, to the best of our knowledge, there are no published reports on serum EPCA expression and the relationship between serum EPCA levels and the clinical outcome in men with a diagnosis of isolated HGPIN. Therefore, we examined the serum levels of EPCA by using an indirect ELISA approach in 112 cases with isolated HGPIN diagnosed on initial biopsies, and to further clear whether serum EPCA level is a significant predictor of subsequent malignancy onset in those patients during a follow-up period of ≥5 years.

**Materials and methods**

**Patients**
Between January 2000 and May 2003, 687 men underwent transrectal ultrasound-guided needle biopsy of the prostate at our institution by using a standardized 12-cores biopsy scheme. This scheme was defined by two consecutive sets of sextant biopsies from the peripheral zone on the right and left. One hundred and forty-three (20.8%) men were histopathologically diagnosed with HGPIN and no focus of concurrent cancer on initial biopsy, i.e. isolated HGPIN. No patients had a previous biopsy, a previous diagnosis of PCa, and ever received androgen deprivation therapy, chemotherapy, or radiation therapy. Of those 143 patients with isolated HGPIN, 112 men had one blood draw on the morning of the scheduled day of prostate biopsy after a overnight fast at our Department and the volume of samples was sufficient for the determination of serum EPCA and total prostate specific antigen (PSA) levels. Thus, a total of 112 isolated HGPIN patients were enrolled into this study. The blood samples were collected in evacuated tubes. Serum was separated by centrifugation at 1,200g for 10 min at room temperature, and then stored immediately at –80 °C until analysis. Serum total PSA was measured by radioimmunoassay (Hybritech Inc., San Diego, CA) in each case. All patients provided a signed study-specific informed consent form prior to having the blood sampled, and the Institutional Review Board approved this study protocol.

**Indirect ELISA**

A commercially available anti-EPCA polyclonal antibody (Onconome Inc., Seattle, Washington) was used, and an indirect ELISA for EPCA was performed as described recently by Paul *et al*. (2005). Here, serum sample from each patient was plated in
triplicate in 96-well Nunc Immunoplate MaxiSorb plates (Nunc, Wiesbaden, Germany). When the reaction was stopped, the plates were read at 450nm on the LabSystems Multiskan Multisoft microplate reader (LabSystems, Helsinki, Finland). The mean of the three measurements for each sample was used for data analyses. In the present study, all assays were performed by an experienced laboratory expert at our institute (G.Z), who had no knowledge of the case status of the samples.

**Follow-up study**

All 112 men with isolated HGPIN were followed continuously for a minimum of 5 years from the date of the initial diagnosis. During this period, all cases had the first 12-core follow-up biopsy of prostate within 18 months of the diagnosis of HGPIN alone on their initial biopsy. The patients with persistently isolated HGPIN on the first follow-up biopsy had an immediate second 12-core biopsy when a raised PSA or a suspicious digital rectal examination (DRE) finding or both was present, and those without raised PSA levels and/or abnormal DRE findings underwent the second 12-core biopsy at the end of the study at our department. Before each re-biopsy, patients underwent serum total PSA assays with radioimmunoassay (Hybritech Inc., San Diego, CA) and DRE. The PSA velocity during the year prior to diagnosis was determined from at least three serum PSA measurements between the first and the last measurements, and was calculated as the linear regression slope of the PSA levels.

**Statistical analysis**

Student’s $t$-test was used to compare the geometric means of serum EPCA absorbances between isolated HGPIN cases with and without subsequent cancer. The
receiver operating characteristic (ROC) curves were constructed, and predictive accuracy of parameters for the presence of subsequent cancer was quantified by using the area under the curve (AUC). The differences of AUCs among parameters were assessed by the Mann-Whitney U test. Univariate and multivariate Cox regression models were developed to evaluate the relative risk of subsequent cancer onset on follow-up. For all analyses, a $P$-value of <0.05 was considered to be statistically significant. The data analyses and interpretation were independently reviewed.

Results

The characteristics of isolated HGPIN patient are outlined in Table 1. All eligible patients were followed up until one was subsequently diagnosed with PCa or until the study terminated in June 2008. No patient was lost to follow-up. The median length of follow-up was 80.7 months, with a range of 60-100 months. In total, 31 patients were diagnosed with subsequent cancer on follow-up study, in which 19 cases were identified with a Gleason score of 6 and the others with a score of 7. All 31 cancers were clinically localized tumors as determined by bone scan, computerized tomography of the abdomen and pelvis, chest radiography and serum enzymatic prostatic acid phosphatase testing. All these patients had a radical prostatectomy as cancer treatment and all had organ-confined pT2a–pT2c disease with negative surgical margins.

Our ELISA analysis of 112 individuals with isolated HGPIN showed that all had a detectable concentration of EPCA in the initial sera, with a mean of $0.64 \pm 0.13$ absorbance at 450 nm. Figure 1 clearly showed that all 31 subsequent cancers had
their initial serum-EPCA levels ≥1.10 absorbance. In addition, the ELISA assay also detected serum-EPCA levels above 1.10 absorbance in 2 of 81 cases who were subsequently diagnosed without PCa. However, none of the individuals who had an initial serum-EPCA level of <1.10 had progressed to clinically significant cancer during the follow-up period of 60–100 months. Thus, if using a cutoff value of 1.10, an overall specificity for this group with isolated HGPIN was 97.5%, whereas the sensitivity for identifying subsequent malignancy was 100%. The positive predictive values (PPV) and negative predictive values (NPV) were 93.9% and 100%, respectively (Table 2). Intriguingly, the circulating EPCA level in the initial serum from patients with subsequent PCa was significantly higher than that from those without (1.62 ± 0.37 versus 0.57 ± 0.11, \( P<0.001 \)). The posthoc analysis showed that men with isolated HGPIN who presented with an increased EPCA level in the initial serum (≥1.10) were significantly more likely to develop a clinically significant cancer after the initial diagnosis than those who had an absorbance of <1.10 for serum EPCA (\( P<0.001 \)).

There is no statistically significant correlation between the Gleason scores and the EPCA levels in 31 isolated HGPIN men with cancer because all of the Gleason grade was either 6 or 7 ( \( T \) test, \( P=0.34 \)). Eighteen of 112 men had abnormal DRE findings noted at study entry, in which only two cancers were identified on the follow-up biopsies. Not surprisingly, DRE findings were not significantly associated with the subsequent development of cancer in isolated HGPIN (chi-square test, \( P=0.47 \)). Although 11 of 31 cases with subsequent cancer had the elevated PSA levels and the
rest had a decline in PSA levels at the time to diagnosis when compared to their initial
PSA concentrations, the elevated, declined, and overall PSA levels were all
statistically insignificant (3.46 ± 0.6 ng/ml versus 2.78 ± 0.3 ng/ml, \( P = 0.11 \); 2.85 ±
0.5 ng/ml versus 3.03 ± 0.6 ng/ml, \( P = 0.27 \); and 3.16 ± 0.6 ng/ml versus 2.91 ± 0.4
ng/ml, \( P = 0.13 \), respectively). In addition, PSA velocity was also found to be
statistically insignificant between isolated HGPIN patients with subsequent cancer
and without (0.51 ng/ml/year versus 0.47 ng/ml/year, \( P = 0.18 \)). Thirteen of 112 men
had PSA velocity >0.75 ng/ml/year, in which 4 cases were identified as having cancer
on follow-up. Table 2 gives the calculated specificity, sensitivity, PPV and NPV of
PSA velocity >0.75 ng/ml/year, abnormal DRE findings and initial serum-EPCA level,
respectively.

The ROC curves for PSA velocity, abnormal DRE findings and initial
serum-EPCA level are plotted in Figure 2, and the calculated AUCs and 95%
confidence intervals (CI) are presented in Table 3. Mann-Whitney U test showed that
the AUC was significantly greater for serum-EPCA level than for PSA velocity and
abnormal DRE findings (\( P<0.001 \) and \( P<0.001 \), respectively). Furthermore, univariate
and multivariate Cox regression models were developed to evaluate the relative risk
of subsequent cancer onset on follow-up. As shown in Table 4, the results confirmed
that the increased serum-EPCA levels (\( \geq 1.10 \)) carried a significantly higher risk of
the presence of subsequent cancer in isolated HGPIN over a period of \( \geq 5 \) years after
the initial diagnosis (Relative risk: 3.32, 95%CI: 2.62-5.03, \( P<0.001 \)) whereas
elevated serum PSA concentrations, PSA velocity and abnormal DRE findings did
Discussion

In the present study, we found a detectable circulating EPCA protein in the serum and a significant correlation between increased levels of serum-EPCA and risk of subsequent cancer onset on follow-up in a group of individuals with isolated HGPIN. In a recent study by Paul et al. (2005), PIN alone appears to be not sufficient to result in detectable serum levels of EPCA. Our data are not in accordance with this finding. This discrepancy may be related to the different sensitivity of the assay to the populations of various radical/ethnic and geographic characteristics, such as Chinese and American. In addition, for blood-based tests using antibodies, the antibodies, especially polyclonal antibodies, can show even minor crossreactivity with other antigens that circulate in the blood at various levels. Our follow-up studies revealed a statistically significant increase in serum-EPCA levels in initial samples obtained from isolated HGPIN patients with subsequent PCa as compared with samples from those without. Taken together with the previous immunostaining findings for EPCA (Dhir et al. 2004, Uetsuki et al. 2005, Hansel et al. 2007), these data imply that EPCA may be used as a potential predictive marker for PCa onset. Moreover, the results presented here do not reveal a correlation between initial serum-EPCA absorbance and Gleason score of subsequent tumor. This is in agreement with the results of Dhir et al. (2004), Paul et al. (2005) and Uetsuki et al. (2005), where tissue EPCA staining and plasma EPCA absorbances were not correlated with tumor stage in patients with PCa, supporting the suggestion that EPCA expression occurs early during the
development of cancer. In the present study, no statistically significant difference was found in regard to patients age between men with subsequent cancer findings and those without (64.2±1.0 versus 64.8±1.1, \(P= 0.28\)), suggesting that age probably does not influence serum levels of EPCA in patients with isolated HGPIN.

Although serum PSA and its isoforms have been of continuous interest with regard to early detection and screening for PCa (Crawford & Abrahamsson 2008, Schröder et al. 2008, Jansen et al. 2009), limitations in the sensitivity and specificity for the prediction of PCa are well understood (Welch et al. 2005, Steuber et al. 2008). Serum PSA level, PSA velocity and abnormal DRE findings as parameters of clinical importance have more recently been evaluated to predict the presence of PCa among men with HGPIN, however, the results were still conflict (Cheng et al. 2004, Moore et al. 2005, Herawi et al. 2006, Akhavan et al. 2007, Loeb et al. 2007). In this study, serum PSA changes, PSA velocity and abnormal DRE findings were not shown to be helpful in predicting the likelihood of cancer on the repeat biopsy in patients with isolated HGPIN. This is in agreement with the results of other authors (Cheng et al. 2004, Moore et al. 2005, Herawi et al. 2006, Akhavan et al. 2007) and our recent findings (Zhao & Shen 2007). Henshall et al. (2001) reported that overexpression of the cell cycle inhibitor p16INK4A in HGPIN is an independent predictor of PCa relapse after radical prostatectomy, which is the first prognostic marker identified in patients with HGPIN. Microvessel density as a molecular marker was found to distinguish HGPIN that have the potential of progressing to PCa from those that do not have (Sinha et al. 2004). Our recent studies revealed, using \textit{in situ} hybridization
analysis, that prostate stem cell antigen (PSCA) expression in HGPIN tissues on the initial biopsy of prostate was independently predictive of the onset of subsequent cancer over ≥5 years of follow-up in a group of 66 men with isolated HGPIN (odd ratio: 3.23; 95% CI: 1.57–6.15; \( P<0.001 \)) (Zhao & Shen 2007). Most recently, a prospective study showed that the prostate cancer gene 3 (PCA3) score was significantly higher in the urine of men with HGPIN versus those without, suggesting the potential of PCA3 in identifying men at risk of developing PCa (Haese et al. 2008). However, the identification of novel biomarkers in blood is still needed.

With the observation that isolated HGPIN patients had the detectable circulating EPCA protein, another concern to us in this study is whether blood levels of EPCA can predict the progression to clinical cancer in these individuals. Thus, we evaluated the relationship of initial serum-EPCA levels and the subsequent PCa risk over a follow-up period of ≥5 years in 112 cases with isolated HGPIN. The results presented here firstly demonstrate a significant trend for those who had an increased initial serum level of EPCA (≥1.10 absorbance) to progress to a clinically significant cancer after the initial diagnosis of isolated HGPIN. Cancer diagnostic tests must be both specific and sensitive. Our ELISA test showed that initial serum-EPCA absorbance ≥1.10 does not only exhibit an overall specificity of 97.5% and a sensitivity of 100% for detection of subsequent cancer but also have a PPV of 93.9% and a NPV of 100% in this set of patients. Our results are similar to those of Paul et al. (2005) and Leman et al. (2007, 2009), where the blood-based ELISA for EPCA and EPCA-2 showed both high specificity and sensitivity for patients with PCa. Taken together, these data
support the potential of using the EPCA ELISA test to identify men with isolated HGPIN who do have carcinoma subsequently. The estimation of AUC values from ROC curves for initial serum-EPCA level, PSA velocity, and abnormal DRE findings showed that initial serum-EPCA level had the largest AUC among those parameters, demonstrating the most promising marker for predicting the presence of subsequent cancer in isolated HGPIN. Furthermore, univariate and multivariate Cox regression analyses confirmed that initial serum-EPCA level ≥1.10 carried a significantly increased risk for the development of subsequent PCa in the process of isolated HGPIN. Taken together, our findings presented here substantiate the possible role of EPCA as a serum marker for identifying men with isolated HGPIN who are at higher risk to subsequently develop cancers early in the disease process, even at an earlier interval than 5 years. Although a larger study is required for definitive conclusions and we are now in the process of setting up such a study, the preliminary results of this study may have significant clinical implications for managing a patient with isolated HGPIN. The individuals with isolated HGPIN who had initial serum-EPCA absorbances greater than or equal to 1.10 absorbance would be followed-up more closely than those who had initial serum-EPCA levels below 1.10, regardless of PSA changes and DRE findings.

In conclusion, this preliminary study, to the best of our knowledge, firstly demonstrated a detectable level of EPCA in the initial serum of men with isolated HGPIN by an ELISA test and a statistically significant difference of initial serum EPCA concentrations between isolated HGPIN individuals with subsequent PCa and
those without. The findings that those individuals with increased serum levels of EPCA (≥1.10 absorbance) are at higher risk to subsequently develop clinical cancer over a period of ≥5 years make EPCA potentially a very powerful serum biomarker for the early detection of PCa in patients with isolated HGPIN.
Acknowledgements

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Figure 1. Serum analysis of EPCA. Initial serum-EPCA levels were measured in 112 men with isolated HGPIN by using an indirect ELISA, of whom 31 were subsequently found with cancer and the others were not on a follow-up of ≥5 years. Each sample was done in triplicate and reported here as the mean. Representative of three independent experiments.
Figure 2: The receiver operating characteristic (ROC) curves for PSA velocity, DRE findings and initial serum-EPCA level. AUC = area under ROC curve.
Figure 1. Serum analysis of EPCA. Initial serum-EPCA levels were measured in 112 men with isolated HGPIN by using an indirect ELISA, of whom 31 were subsequently found with cancer and the others were not on a follow-up of ≥5 years. Each sample was done in triplicate and reported here as the mean. Representative of three independent experiments.

Figure 2. The receiver operating characteristic (ROC) curves for PSA velocity, DRE findings and initial serum-EPCA level. AUC = area under ROC curve.
Table 1. Clinical characteristics of patients with isolated HGPIN

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Eligible patients (n=112)</th>
<th>All patients (n=143)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yrs)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mean ± SD</td>
<td>64.6 ± 1.0</td>
<td>64.2 ± 1.0</td>
<td>0.37</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td></td>
<td></td>
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<tr>
<td>mean ± SD</td>
<td>67.3 ± 5.2</td>
<td>68.1 ± 5.3</td>
<td>0.34</td>
</tr>
<tr>
<td>Height (cm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mean ± SD</td>
<td>169.4 ± 4.4</td>
<td>169.7 ± 4.5</td>
<td>0.41</td>
</tr>
<tr>
<td>Body mass indices (BMI, kg/m2)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mean ± SD</td>
<td>24.6 ± 2.3</td>
<td>24.4 ± 2.3</td>
<td>0.28</td>
</tr>
<tr>
<td>Prostate volume (ml)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mean ± SD</td>
<td>43.3 ± 8.2</td>
<td>44.2 ± 8.4</td>
<td>0.30</td>
</tr>
<tr>
<td>Serum PSA (ng/ml)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0-2.5</td>
<td>64 (57.1%)</td>
<td>83 (58%)</td>
<td>0.11</td>
</tr>
<tr>
<td>2.6-4.0</td>
<td>29 (25.9%)</td>
<td>35 (24.5%)</td>
<td>0.17</td>
</tr>
<tr>
<td>4.1-10.0</td>
<td>15 (13.4%)</td>
<td>20 (14%)</td>
<td>0.16</td>
</tr>
<tr>
<td>&gt;10.0</td>
<td>4 (3.6%)</td>
<td>5 (3.5%)</td>
<td>0.21</td>
</tr>
<tr>
<td>DRE findings</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>normal</td>
<td>94 (83.9%)</td>
<td>124 (86.7%)</td>
<td>0.14</td>
</tr>
<tr>
<td>abnormal</td>
<td>18 (16.1%)</td>
<td>19 (13.3%)</td>
<td>0.26</td>
</tr>
</tbody>
</table>

PSA, prostate specific antigen; DRE, digital rectal examinations; HGPIN, high-grade prostatic intraepithelial neoplasia; SD, standard deviation.
Table 2. The specificity, sensitivity, PPV and NPV of the parameters evaluated in the study

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Specificity (%)</th>
<th>Sensitivity (%)</th>
<th>PPV (%)</th>
<th>NPV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PSA velocity (&gt;0.75 ng/ml/year)</td>
<td>88.9</td>
<td>12.9</td>
<td>30.8</td>
<td>72.7</td>
</tr>
<tr>
<td>DRE findings (abnormal)</td>
<td>80.2</td>
<td>6.5</td>
<td>11.1</td>
<td>69.1</td>
</tr>
<tr>
<td>Initial serum-EPCA level (≥1.10)</td>
<td>97.5</td>
<td>100</td>
<td>93.9</td>
<td>100</td>
</tr>
</tbody>
</table>

PSA, prostate specific antigen; DRE, digital rectal examinations; EPCA, early prostate cancer antigen; PPV, positive predictive values; NPV, negative predictive values.
Table 3. The AUC values and 95% CI calculated from ROC curves for the parameters evaluated in the study

<table>
<thead>
<tr>
<th>Parameter</th>
<th>AUC values</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>PSA velocity</td>
<td>0.641</td>
<td>0.583-0.762</td>
</tr>
<tr>
<td>Abnormal DRE findings</td>
<td>0.575</td>
<td>0.417-0.664</td>
</tr>
<tr>
<td>Initial serum-EPCA level</td>
<td>0.892*</td>
<td>0.753-0.968</td>
</tr>
</tbody>
</table>

PSA, prostate specific antigen; DRE, digital rectal examinations; EPCA, early prostate cancer antigen; AUC, area under the curve; 95% CI, 95% confidence intervals; ROC, receiver operating characteristic curves.

*Statistically significant versus PSA velocity ($P<0.001$) and abnormal DRE findings ($P<0.001$), respectively, by the Mann–Whitney U test.
Table 4. Univariate and multivariate Cox regression analyses for relative risk of the subsequent PCa onset in isolated HGPIN

<table>
<thead>
<tr>
<th>Covariate</th>
<th>Relative risk</th>
<th>95%CI</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Univariate</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Increased serum EPCA level</td>
<td>6.84</td>
<td>5.34 – 8.76</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>(≥1.10 absorbance)</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Elevated serum PSA level</td>
<td>0.72</td>
<td>0.45 – 1.61</td>
<td>0.52</td>
</tr>
<tr>
<td>PSA velocity (&gt;0.75 ng/ml/year)</td>
<td>1.16</td>
<td>0.77 – 2.18</td>
<td>0.24</td>
</tr>
<tr>
<td>Abnormal DRE findings</td>
<td>0.45</td>
<td>0.26 – 1.15</td>
<td>0.86</td>
</tr>
<tr>
<td><strong>Multivariate</strong></td>
<td></td>
<td></td>
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<tr>
<td>Increased serum EPCA level</td>
<td>3.32</td>
<td>2.62 – 5.03</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>(≥1.10 absorbance)</td>
<td></td>
<td></td>
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<tr>
<td>Elevated serum PSA level</td>
<td>0.41</td>
<td>0.27 – 1.11</td>
<td>0.63</td>
</tr>
<tr>
<td>PSA velocity (&gt;0.75 ng/ml/year)</td>
<td>0.75</td>
<td>0.48 – 1.24</td>
<td>0.18</td>
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<tr>
<td>Abnormal DRE findings</td>
<td>0.78</td>
<td>0.16 – 0.75</td>
<td>0.88</td>
</tr>
</tbody>
</table>

PSA, prostate specific antigen; DRE, digital rectal examinations; EPCA, early prostate cancer antigen; HGPIN, high-grade prostatic intraepithelial neoplasia; 95% CI, 95% confidence intervals.