**ABSTRACT**

High-grade prostatic intraepithelial neoplasia (HGPIN) is the most important predecessor of prostatic adenocarcinoma (PA), and frequently co-exists with PA. The aim of our study was to correlate percentage of PA-positive repeated biopsies with the number of initial biopsy specimens and their morphology.

Our case series consists of 254 patients with initial needle biopsies of the prostate. The patients were divided into four groups according to the number of biopsy specimens: group I, with 2-3 specimens (69 pts); group II, with 4-5 specimens (47 pts); group III, with 6-7 specimens (99 pts); and group IV, with 8-10 specimens (39 pts). PA was found in 101 cases (39.8%) and HGPIN in 98 cases (38.6%). In 28 patients with PA (out of 101 cases), HGPIN was co-existent with PA (27.7%). PA was found in 18 cases among the patients from group I (26.1%), and HGPIN in 17 cases (24.6%) from the same group. In group II, PA was found in 16 cases (34.0%) and HGPIN in 17 cases (36.2%). In group III, PA was found in 46 cases (46.5%), and HGPIN in 47 cases. In group IV, PA was found in 19 cases (48.7%), and HGPIN in 20 cases (51.3%). In 35 out of 98 patients (35.7%) with HGPIN, repeated biopsies were performed. Among 13 cases with HGPIN found in one biopsy specimen only, repeated biopsy discovered PA in three of them (23.1%). Among 14 cases with HGPIN found in two biopsy specimens, repeated biopsy discovered PA in six of them (42.9%). Finally, among 8 patients with HGPIN found in 3 initial biopsy specimens, repeated biopsy found PA in four of them (50.0%). Among 35 cases of HGPIN at initial biopsy, the most common histological types were tufting (60%) and micropapillary (25.7%), while the cribriform type was found in 11.4% of specimens, and the flat type in 2.9%. After repeated biopsy, PA was found the most frequently in patients with micropapillary (44.4%) and tufting (38.1%) types.

There was a significant correlation between the number of detected PAs and either the total number of specimens or the number of specimens with HGPIN at initial biopsy. Significant correlation was also found between tufting/micropapillary type of HGPIN and PA.

**Key Words:** high grade prostatic intraepithelial neoplasia, prostatic adenocarcinoma, biopsy of prostate.

**KORELATIVNA ANALIZA IZMEĐU BROJA UZETIH ISEČAKA PROSTATE I PROCENTA DIJAGNOSTIKOVANOG KARCINOMA PROSTATE U PONOVLJENIM BIOPSIJAMA: STUDIJA PRESEKA**

Ibrahim Prelijević1, Zorica Mihajlović2, Nedeljko Vezmar3 i Sanja Knežević4
1Department of Pathology, Health Center, Novi Pazar; 2Department of Pathology, Faculty of Medicine, University of Kragujevac; 3Department of Urology, Clinical Center, Kragujevac; 4Faculty of Medicine, University of Kragujevac

Received / Primljen: 17. 07. 2008. Accepted / Prihvaćen: 15.10.2008.

**ABSTRACT**

Visokostepenoj prostatičnoj intraepitelijskoj neoplaziji (VSPIN) je posvećena izuzetna pažnja kao najvažnijem prekursoru adenokarcinoma prostate (AP) i signifikantnom udruženosti sa AP. Cilj istraživanja je bio da se usredi uporedna analiza VSPIN nađenog na inicijalnoj biopsiji i AP dijagnostikovanog na ponovnoj biopsiji u odnosu na broj uzetih isečaka i morfološki izgled.

Analizirano je 254 pacijenta sa urađenom iglenom biopsijom prostate. Primijenjene su patohistološke metode istraživanja. Pacijenti su na osnovu broja uzetih iglenih biopsija prostate svrstani u četiri grupe: I grupa 2-3 uzorka (69 sl.), II grupa 4-5 (47 sl.), III grupa 6-7 (99 sl.) i IV grupa 8-10 uzorka biopsije (39 sl.). U 101 sl. (39.8%) nađen je AP a VSPIN u 98 sl. (38.6%). U 28/101 sl. (27.7%) AP nađen je u rebiopsiji. U grupi II je nađen u 16. sl. (34.0%) i VSPIN u 17. sl. (26.1%). U grupi IV je nađen u 16. sl. (34.0%) i VSPIN u 17. sl. (36.2%). U III grupi je nađen u 46 sl. (46.5%), a u 47 sl. (47.5%) VSPIN. U IV grupi je nađen u 19 sl. (48.7%) i VSPIN u 20 sl. (51.3%). U 35/98 sl. (35.7%) sa VSPIN u rebiopsiji je odbijen. Koeficijent uključenosti u rebiopsiji je u 3 sl. (23.1%) nađen AP. Kod 14 sl. sa 2 isečaka nađen je AP u 6 sl. (42.9%), i kod 8 sl. sa 3 isečaka nađen je AP u 4 sl. (50.0%). Od 35 sl. VSPIN na inicijalnoj biopsiji najčešći histološki tip bio je resiţni (60%) i micropapilarni (25.7%), a cribriformni tip je nađen u 11.4% i pljosnat u 2.9%. Na rebiopsiji je AP najčešće nađen kod micropapilarnog (44.4%) i resiţastog tipa (38.1%).

Postoji značajna korelacija u detekciji VSPIN i AP u odnosu na broj uzetih isečaka, broj izdah ukošenih VSPIN na inicijalnoj biopsiji i na rebiopsiji, kao i korelacija između histološkog resiţastog/micropapilarnog tipa VSPIN prema AP.

**Klijurne reči:** Visoko stepeni PIN, adenokarcinom prostate, biopsija prostate.

**SAŽETAK**
INTRODUCTION

Prostatic adenocarcinoma (PA) is one of the most important human cancers, with rising incidence. It is more frequent in men of advanced age (1, 2, 3). Among men, PA is the cancer with the highest incidence, and the second most common cause of death from malignant diseases (after lung cancer). The mortality rate from PA is constantly rising (2).

According to epidemiological, histological, molecular and experimental studies, High Grade Prostatic Intraepithelial Neoplasia (HGPIN) is the most significant predictor of PA; it is considered to be the pre-malignant phase of PA (1, 3, 4). Four histological types of HGPIN have been described: tufting, micropapillary, cribriform and flat (1). Some studies have shown the high predictive value of HGPIN, which precedes PA by ten years (5, 4, 6, 7). Once discovered in a biopsy specimen, isolated HGPIN suggests the necessity for a clinical follow-up of the patient, including repeated biopsy of the prostate (8, 3, 6).

Some of the studies have shown that the detection rate of malignant lesions in the prostate increases with the number of specimens taken (9, 10, 11). Additionally, the correlation between the number of HGPIN-positive specimens taken at initial biopsy, their histological type and detection rate of PA at repeated biopsies was demonstrated (9, 12). On the other hand, there are studies that failed to show such a correlation (13). Due to these controversies and a relatively small number of studies in this area, further research efforts are necessary (13, 9, 6).

Our primary objective was to investigate the correlation between the detection rate of HGPIN and PA, and the number of tissue specimens taken by needle biopsy of the prostate.

Our secondary objective was to investigate correlations between the number of HGPIN-positive initial biopsy specimens or the incidence of certain histological types of HGPIN at initial biopsy, and the detection rate of PA at repeated biopsies.

MATERIALS AND METHODS

We have included 254 patients of the Clinical Centre “Kragujevac”, Kragujevac, Serbia in our study. All the subjects received needle biopsies of the prostate between January 1st, 2003 and June 1st, 2007.

The tissue specimens obtained by needle biopsy of the prostate were fixed in 4% buffered formalin, inserted into paraffin blocks and cut by microtome to preparations 4-6 micrometers thick. Both standard haematoxylin-eosin and histochemical (Alcian blue, Masson) staining methods were used. The results were compared by Chi-square test.

The tissue specimens of the prostate (2-10 per patient) were taken by a needle from both the right and left lobes of the gland. According to the number of tissue specimens taken, the patients were classified into four groups: group I with 2-3 specimens (69 pts), group II with 4-5 specimens (47 pts), group III with 6-7 specimens (99 pts) and group IV with 8-10 specimens taken (39 pts).

In groups III and IV, 46 and 31 biopsies, respectively, were taken with the aid of transrectal ultrasound imaging.

Among the 98 patients with an HGPIN-positive initial biopsy, thirty-five were subjected to repeated biopsy of the prostate. The remaining 53 patients with HGPIN were lost to follow-up. According to the number of HGPIN-positive specimens at the initial biopsy, the patients were classified into three groups: group I – one HGPIN-positive specimen, group II - 2 HGPIN-positive specimens and group III – three HGPIN-positive specimens. The results were compared by student’s T-test for independent samples.

RESULTS

Among the 254 patients subjected to needle biopsy of prostate in our study, one of the following clinical signs was always present: positive digital rectal examination of the prostate (DRE), elevated PSA or suspicious transrectal ultrasound imaging of the prostate.

The highest percentage of the study sample (Figure 1) consisted of patients in their eighth (123 pts, or 48.0%) or seventh (88 pts, or 34.7%) decade of life. Thirty-one patients (12.2%) were in their sixth, and 13 patients (5.1%) were in their ninth decade of life.

Table 1 shows the histological findings of the study samples. One hundred and one patients, with average age of 70.3 years, had PA (39.8%). Pre-malignant lesions were found in 117 patients (46.1%), and 98 of
them had HGPIN (38.6%, average age 69.5 years). The 28 patients with PA (101) had concomitant HGPIN (27.7%).

**Table 1.** Histological lesions from needle biopsies of prostate in 254 patients.

<table>
<thead>
<tr>
<th>Histological lesions</th>
<th>No cases</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prostatic adenocarcinoma</td>
<td>101</td>
<td>39.8</td>
</tr>
<tr>
<td>HGPIN</td>
<td>98</td>
<td>38.6</td>
</tr>
<tr>
<td>Atypical adenomatous hyperplasia</td>
<td>13</td>
<td>5.1</td>
</tr>
<tr>
<td>Atypical small acinar proliferation</td>
<td>6</td>
<td>2.4</td>
</tr>
<tr>
<td>Florid hyperplasia of basal cells</td>
<td>7</td>
<td>2.8</td>
</tr>
<tr>
<td>Clear cells cribriform hyperplasia</td>
<td>3</td>
<td>1.2</td>
</tr>
<tr>
<td>Benign prostatic hyperplasia</td>
<td>64</td>
<td>25.2</td>
</tr>
</tbody>
</table>

Atypical adenomatous hyperplasia (AAH) was found in 13 cases (5.1%), and atypical small acinar proliferation (ASAP) in 6 cases (2.4%). Sixty-four patients (25.2%) had benign hyperplasia of the prostate (BHP), seven patients (2.8%) had rich hyperplasia of basal cells and there was bright cellular cribriform hyperplasia in three cases (1.2%).

Table 2 shows the comparison of detection rates of PA and HGPIN with the number of prostate tissue specimens taken at initial biopsy. In the group of 69 patients with 2-3 specimens, PA was found in 18 cases (26.1%) and HGPIN in 17 cases (24.6%). In the group of 47 patients with 4-5 specimens, PA was found in 16 cases (34.0%), and HGPIN in 17 cases (36.2%). In the group of 99 patients with 6-7 specimens, PA was found in 46 cases (46.5%), and HGPIN was found in 47 cases (47.5%). In the group of 39 patients with 8-10 specimens, PA was found in 19 cases (48.7%) and HGPIN was found in 20 cases (51.3%). It is obvious from the table that the detection rates of PA and HGPIN increase with the number of tissue samples taken (p<0.05). However, the detection rates are similar in the groups with 6-7 and 8-10 tissue samples per biopsy (p>0.05).

**Table 2.** Comparison of detection rates of PA and HGPIN with number of prostate tissue specimens taken at initial biopsy.

<table>
<thead>
<tr>
<th>Number of prostate tissue specimens</th>
<th>No cases</th>
<th>%</th>
<th>HGPIN – 98 No of cases</th>
<th>%</th>
<th>PA - 101 No of cases</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-3 specimens</td>
<td>69</td>
<td>27.2</td>
<td>18 26.1</td>
<td>17</td>
<td>24.6</td>
<td></td>
</tr>
<tr>
<td>4-5 specimens</td>
<td>47</td>
<td>18.5</td>
<td>16 34.0</td>
<td>17</td>
<td>36.2</td>
<td></td>
</tr>
<tr>
<td>6-7 specimens</td>
<td>99</td>
<td>39.0</td>
<td>46 46.5</td>
<td>47</td>
<td>47.5</td>
<td></td>
</tr>
<tr>
<td>8-10 specimens</td>
<td>39</td>
<td>13.3</td>
<td>19 48.7</td>
<td>20</td>
<td>51.3</td>
<td></td>
</tr>
</tbody>
</table>

In the patients with HGPIN, repeated biopsy of the prostate was infrequently performed. Out of 98 HGPIN-positive patients at initial biopsy, only 35 were subjected to repeated biopsy (35.7%), in 29 cases one repeated biopsy was done, and in 6 cases two, in a 6-12 month interval. The detection rate of PA at repeated biopsy was 37.1% (13 cases out of 35).

Table 3 shows the correlation between the number of HGPIN-positive initial biopsy specimens and the detection rate of PA at repeated biopsies. In the group of 13 patients with one HGPIN-positive specimen, repeated biopsy detected PA in 3 cases (23.1%). In the group of 14 patients with two HGPIN-positive tissue specimens, repeated biopsy detected PA in 6 cases (42.3%); 5 cases in the first repeated biopsy and 1 case in the second repeated biopsy). In the third group of 8 patients with 3 HGPIN-positive tissue specimens, repeated biopsy detected PA in 4 cases (49.0%); 3 cases in the first repeated biopsy and 1 case in the second repeated biopsy). Out of 13 patients in total with PA detected by repeated biopsies, eleven patients were subjected to one (84.6%) and two patients (15.4%) to two biopsies. There are significant differences in the detection rate of PA at repeated biopsies among the groups with different number of HGPIN-positive initial biopsy specimens (p<0.05).

**Table 3.** Correlation between number of HGPIN-positive initial biopsy specimens and detection rate of PA at repeated biopsies.

<table>
<thead>
<tr>
<th>Number of HGPIN-positive needle biopsies</th>
<th>No of cases</th>
<th>The first repeated biopsy with PA No of cases</th>
<th>%</th>
<th>The second repeated biopsy with PA No of cases</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>One</td>
<td>13</td>
<td>3</td>
<td>23.1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Two</td>
<td>14</td>
<td>5</td>
<td>35.7</td>
<td>1</td>
<td>7.2</td>
</tr>
<tr>
<td>Three</td>
<td>8</td>
<td>3</td>
<td>37.5</td>
<td>1</td>
<td>12.5</td>
</tr>
</tbody>
</table>

Table 4 shows the comparison of histological subtype of HGPIN with the detection rate of PA at repeated biopsies. Among the 35 cases of HGPIN that were biopsied, the most frequent histological type was tufting (21/35 pts, or 60%), followed by micropapillary (9/35 pts, or 25.7%). Cribriform type was found in 4/35 cases (11.4%), and flat type in only one case (2.9%). After repeated biopsy, PA was the most frequent in the micropapillary (4/9 pts, or 44.4%) and tufting type (8/21 pts, or 38.1%). For the cribriform type, PA was found in 1/4 cases (25%). However, due to the small number of cases in each of the histological groups, the observed differences were not significant.

**Table 4.** Comparison of histological subtype of HGPIN and detection rate of PA at repeated biopsies.

<table>
<thead>
<tr>
<th>HGPIN – 35 cases.</th>
<th>PA at repeated biopsy – 13 cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Histological type</td>
<td>No of cases</td>
</tr>
<tr>
<td>Tufting</td>
<td>21</td>
</tr>
<tr>
<td>Micropapillary</td>
<td>9</td>
</tr>
<tr>
<td>Cribriform</td>
<td>4</td>
</tr>
<tr>
<td>Flat</td>
<td>1</td>
</tr>
</tbody>
</table>

In the patients with HGPIN, repeated biopsy of the prostate was infrequently performed. Out of 98 HGPIN-positive patients at initial biopsy, only 35 were subjected...
DISCUSSION

The detection rate of HGPIN after needle biopsy of the prostate is higher than 25%, increases with age and is associated with PA in 85% of patients (3). In a large study by Cheng (3), presence of HGPIN at initial biopsy bears a risk of 35% for detection of PA at repeated biopsies. Therefore, detection of HGPIN at initial biopsy is an indication for repeated biopsy, especially if the patient is in good general health. Lowe (2) has shown an incidence of 36% during a 6-year study period. Schonfeld (14) has followed 100 patients, detecting PA in 39%, HGPIN in 22% and associated PA and HGPIN in 26% of patients. After repeated biopsy in patients with isolated HGPIN (22 pts), PA was detected in 34% of them. In the same study, a higher number of HGPIN-positive tissue specimens were associated with a higher detection rate of PA at repeated biopsy.

HGPIN is associated with PA, and the incidence of HGPIN is higher in patients with PA, compared with healthy patients. Davidson (4), in his study of 100 patients with HGPIN and 112 patients without HGPIN, has detected prostate cancer in 35% of repeated biopsies in the first group, and in 13% of repeated biopsies in the second group. Other studies have had similar results (8, 6). All of these studies recommend strict follow-up of the patients with an HGPIN-positive initial biopsy. Kronz et al. (6) had analysed 245 patients with HGPIN-positive initial biopsies; in this group, repeated biopsy detected PA in 32.2% of cases, which is about one-third of all patients.

The results of our study fit well with already published data. One-hundred-and-one patients were PA-positive (39.8%), with an average age of 70.3 years. In total, premalignant lesions were found in 117 patients (46.1%), and HGPIN was found in 98 patients (38.6%), with an average age of 69.5 years. Additionally, in our study, HGPIN was significantly associated with PA. Twenty-eight patients with PA (27.7% of 101 patients) also had HGPIN. On the other hand, 35 patients with an HGPIN-positive initial biopsy (out of 98 patients in total) were subjected to repeated biopsy, and PA was found in 13 cases (37.1%). Therefore, the total association rate of HGPIN with PA in our study was 41/114 patients (36.0%). However, one should bear in mind that only 35.7% of patients with HGPIN were subjected to repeated biopsy, which is an unacceptably low rate.

According to other studies (10), the detection rate of PA increases with the number of tissue specimens taken. The detection rate of PA is higher with 2 or more biopsy specimens containing HGPIN (11, 15, 16).

Roscigno and associates (12) followed 47 patients with an HGPIN-positive initial biopsy for 11.4 months on average, and repeated biopsies (average number of biopsy specimens was 11.5); PA was detected at repeated biopsy in 21 patients (44.6%).

In our study, the detection rate of PA or HGPIN increased with the number of tissue samples taken (p < 0.05). The highest detection rate was observed in groups with 6-7 and 8-10 tissue specimens.

Roscigno (12) found that PA is more frequent in patients with multi-focal HGPIN (70%) than in patients with mono-focal HGPIN (10%). The patients with 10 to 12 tissue specimens at initial biopsy, with mono-focal or multi-focal HGPIN, have a 45% detection rate of PA at repeated biopsy. Therefore, multi-focal HGPIN at initial biopsy is highly predictive for prostate cancer (14, 17, 18).

Abde-Khalek and associates (8) tried to find other significant predictors of PA among HGPIN-positive patients. Eighty-three patients with previous HGPIN-positive sextant biopsies (number of tissue specimens was 11) were followed, and the biopsies were repeated in 31 of them. Enrollment criteria for the second biopsy were an increase in PSA and/or abnormal findings from a digital rectal examination. The repeated biopsies detected PA in 30/83 patients (36%). The PSA level, digital rectal examination findings and transrectal ultrasound examination were not useful predictors of PA. However, the patients’ age, PSA density and number of HGPIN-positive tissue specimens were independent predictors of PA (p < 0.001).

Kronz and associates (6) have shown that the only independent histological predictor of PA was the number of HGPIN-positive tissue specimens: the cancer risk was 30.2% in patients with 1-2 specimens, 40% in patients with 3 specimens and 75% in patients with more than three HGPIN-positive tissue specimens. Bishara and associates (9) followed 132 patients with an HGPIN-positive initial biopsy, and found PA at repeated biopsy in 28.8% of patients (89.5% were detected at the first two repeated biopsies). The patients with 2 or more HGPIN-positive tissue specimens at initial biopsy had PA at repeated biopsy in 35.9% of cases, and those with only one HGPIN-positive specimen had PA in 22% of cases. In total, PA was detected in 32% of patients with an HGPIN-positive initial biopsy. Therefore, the patients with multiple HGPIN-positive tissue specimens at initial biopsy bear a highest risk of PA.

Naya et al. (13) also investigated whether presence or number of HGPIN-positive tissue specimens at initial biopsy were predictors of PA at repeated biopsies. At initial biopsy, PA was detected in 33.8% of cases and HGPIN in 20.8%. Incidence of HGPIN among the patients with PA-positive initial biopsy was 29.7%. In 175 patients without PA, at least one biopsy was repeated (1 to 3, with an average interval of 3 months), and 47 were HGPIN-positive. In total, repeated biopsies detected PA in 18.3% of 175 patients. The number of initial biopsy tissue specimens was not associated with the probability of PA at repeated biopsy. The authors concluded that the number of HGPIN-positive tissue specimens was not predictive of PA. Scottoni et al. (17), in their recent publication, analysed the detection rate of PA in a much larger sample of patients, which were divided into groups with 12 or 18 tissue samples. There was not a significant
difference in the detection rate of PA among the groups (38.4% and 39.9%, respectively).

In our study, the number of HGPIN-positive tissue specimens at initial biopsy was associated with the detection rate of PA at repeated biopsy. In the group of 13 cases with one HGPIN-positive specimen, PA was detected in 23.1% of the first repeated biopsies. In the group of 14 cases with 2 HGPIN-positive specimens, PA was detected in 42.3% of repeated biopsies. In the group of 8 cases with 3 HGPIN-positive specimens, PA was detected in 49.0% of repeated biopsies. Out of 13 total patients with PA detected at repeated biopsies, eleven patients were subjected to one (84.6%) and two patients (15.4%) to two biopsies. The detection rate of PA at repeated biopsy increases with the number of HGPIN-positive tissue samples at initial biopsy (p<0.05).

Bishara and associates (9) stressed the association between isolated HGPIN and invasive prostatic cancer. They investigated whether histological types (tufting, micropapillar, cribriform or flat) of HGPIN or the number of HGPIN-positive tissue specimens at repeated biopsy were predictive of a higher risk of invasive prostatic cancer. There were 200 HGPIN-positive needle biopsies (average patient age was 66.4 years). The tufting type was present in 59% of cases, micropapillar in 34.3%, cribriform in 6.2% and flat in 0.5% of cases. The tufting and flat types were associated with PA at repeated biopsies in 31.9% of cases, while micropapillar and cribriform types were associated with PA in 22% of cases. They concluded that histological type is more than simply an informative characteristic.

Kronz et al. (6) has shown the following histological parameters from initial biopsy to be predictive of PA: presence of mitoses, number of HGPIN-positive tissue specimens, predominantly micropapillar or cribriform HGPIN and presence of a large, prominent nucleolus.

Among the 35 cases of HGPIN that were re-biopsied, the most frequent histological type was tufting (21/35 pts, or 60%), followed by micropapillar (9/35 pts, or 25.7%). Cribriform type was found in 4/35 cases (11.4%), and flat type was found in only one case (2.9%). After repeated biopsy, PA was most frequent in the micropapillar (4/9 pts, or 44.4%) and tufting types (8/21 pts, or 38.1%). There is stronger association between the tufting/micropapillar type and PA than between the cribriform/flat type and PA. However, the tufting and micropapillar types were the most prevalent, and the total number of repeated biopsies was small.

The results of our study suggest that the patients with multiple HGPIN-positive tissue specimens at initial biopsy bear the highest risk of invasive PA.

REFERENCES