# CAN PLOIDY OF PROSTATE CARCINOMA DIAGNOSED ON NEEDLE BIOPSY PREDICT RADICAL PROSTATECTOMY STAGE AND GRADE?

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ABSTRACT

Purpose: Deoxyribonucleic acid ploidy correlates with the biological behavior of prostate carcinoma. However, the usefulness of ploidy on needle biopsies that show prostate cancer has not been established to our knowledge.

Materials and Methods: We retrospectively determined ploidy on needle biopsies of 159 men with prostate carcinoma treated surgically at Johns Hopkins Hospital. Ploidy was determined by image analysis of Feulgen stained slides. Needle ploidy and Gleason score were compared as prognostic tools in the prediction of grade and stage of subsequent prostatectomy.

Results: Of the 159 cases 98 (62%) were diploid, 16 (10%) tetraploid and 45 (28%) aneuploid. Of the diploid, tetraploid and aneuploid tumors 69, 50 and 44%, respectively, proved to be organ confined. Tetraploid and aneuploid tumors were grouped for the remaining analysis. Needle ploidy correlated significantly with pathological stage (p = 0.003). However, needle Gleason score correlated even more strongly (p <0.001), and on multivariate analysis ploidy was not further predictive of pathological stage once Gleason score was considered. Needle ploidy and Gleason score were predictive of prostatectomy Gleason score (6 or less versus 7 or greater), and on multivariate analysis ploidy was an independently significant predictor of this parameter (p = 0.04). In 13 cases (8%) there was an important grading discrepancy, in which needle ploidy would have accurately predicted prostatectomy grade. However, in 33 cases (21%) needle and prostatectomy Gleason scores were congruent, and needle ploidy did not accurately predict prostatectomy Gleason score.

Conclusions: With accurate needle Gleason grading, ploidy is not helpful in predicting prostatectomy findings. However, ploidy correlates with prostatectomy stage and grade, and may be useful if accurate Gleason grading is a concern.

KEY WORDS: prostatic neoplasms, DNA, ploidies, prognosis, neoplasm staging

Ploidy of prostate carcinoma has been shown to correlate with progression of disease and survival.[1–3] However, it remains to be established whether ploidy analysis provides information in addition to that of routinely reported parameters, such as Gleason score and extent of tumor. Ploidy is not currently recommended for routine clinical use.[4] Several studies have suggested that ploidy of prostate cancer on needle biopsy may be superior to Gleason grading in the prediction of radical prostatectomy grade and stage.[5, 6] We attempt to validate the usefulness of this parameter on prostate needle biopsy by performing ploidy retrospectively on a series of tumors from men with matched needle biopsies and radical prostatectomy specimens.

## MATERIALS AND METHODS

The study group consisted of 165 men who underwent sextant needle biopsy of the prostate at the Johns Hopkins Hospital between 1994 and 1997. In all patients 1 or more biopsy parts were positive for adenocarcinoma. Radical prostatectomy with bilateral lymph node dissection was performed in each case at the same institution. Each prostate was inked, fixed in formalin and submitted in its entirety. Representative sections of seminal vesicles and vasa deferentia, and the entire lymphadenectomy specimen were examined. Tumors were reported as organ or nonorgan confined. The latter category included tumors which showed focal or established extraprostatic extension, seminal vesicle involvement or lymph node metastases.

Gleason scores of all materials and pathological stages of prostatectomy specimens were noted from the surgical pathology reports at Johns Hopkins Hospital. Depending on weekly rotation, 1 of 7 general surgical or a urological pathologist signed out the radical prostatectomy specimens. We did not review or reassign grades. All materials were signed out without knowledge of ploidy.

Ploidy was determined retrospectively at Albany Medical College on the tumors on needle biopsies, without any knowledge of Gleason scores of needle biopsy or prostatectomy material, or pathological stage following surgery. An unstained 5 $\mu$m. section from the positive biopsy was used for ploidy analysis. The highest Gleason score was used in cases with more than 1 part positive for carcinoma. If several parts had identical Gleason score tumor, the part with the greatest extent of tumor was chosen.

Each slide was Feulgen stained and analyzed as described previously.[6] To account for nuclear fragmentation a previously published mathematical algorithm to correct for variations in section thickness was used.[7] A histogram with a minimum of 100 cells was developed for each specimen. Tumor areas with the highest apparent histological grade were chosen for analysis. Coefficients of variation for the G0G1 peaks of nontumoral prostate acinar cells analyzed by this

method varied from 9 to 23%. To accommodate this relatively wide coefficient of variation for normal prostate cells a deoxyribonucleic acid (DNA) index of 0.77 to 1.23 was considered diploid. Cases with G0G1 peaks in the diploid range and G2M components in the tetraploid range of less than 5% of the total tumor cell population were considered diploid. Tumors with a DNA index greater than 1.23 that featured a G2M component of greater than 5% of cells were considered tetraploid and those that lacked such a G2M component were considered aneuploid.

Statistical software was used for data analysis. The Pearson chi-square test was used to correlate needle biopsy Gleason score (6 or less, 7 or greater) and needle ploidy with organ confined status at radical prostatectomy. Similarly, the chi-square test was used for testing needle biopsy Gleason score (6 or less, 7 or greater) and needle ploidy with Gleason score (6 or less, 7 or greater) of the prostatectomy specimen. We performed a multivariate logistic regression analysis to determine if needle ploidy provided prognostic information in addition to that of needle Gleason score in the prediction of prostatectomy stage and grade.

## RESULTS

Of the patients 6 had needle biopsy material unsatisfactory for ploidy analysis, with less than 100 tumor cells present or suboptimal Feulgen staining. Therefore, the final group analyzed comprised 159 men with a median age at biopsy of 61 years (range 45 to 72). Median Gleason score of the tumors on needle biopsy was 6 (range 4 to 9) and median number of biopsy parts positive for carcinoma was 2. Tumor was present on more than 1 part in 105 cases (66%).

Pathological stage was organ confined disease in 96 prostatectomy specimens (60%), focal extraprostatic tumor extension in 26 (16%), established extraprostatic tumor extension in 30 (19%) and seminal vesicle invasion or lymph node metastases in 7 (4%). Gleason scores of the radical prostatectomy specimens ranged from 3 to 9 (median 6). Ploidy analysis of needle biopsies revealed 98 diploid (62%), 16 tetraploid (10%) and 45 aneuploid (28%) tumors (table 1). Representative histograms of tumors of different ploidy are shown in the figure. The rates of organ confinement at prostatectomy for the diploid, tetraploid and aneuploid groups were 69, 50 and 44%, respectively. Tetraploid and aneuploid cases were grouped as nondiploid for data analysis.

Needle ploidy (diploid versus nondiploid) correlated significantly with organ confinement (Pearson's chi-square test p = 0.003). Needle Gleason score (6 or less versus 7 or greater) also correlated with organ confinement (p <0.001). When needle biopsy grade was included in multivariate analysis the prediction of organ confinement by needle ploidy was reduced to just below the level of significance (p = 0.09). Lower Gleason score tumors (6 or less) on needle biopsy and diploid tumors on biopsy showed similar rates of organ confinement (70 and 69%, respectively, table 2). However, higher Gleason score tumors (7 or greater) on biopsy were more predictive of nonorgan confinement (68%) than nondiploid tumors on biopsy (54%).

Ploidy on needle biopsy correlated strongly with Gleason grade (6 or less versus 7 or greater) in the prostatectomy specimen (p <0.001). Gleason grade on needle biopsy (6 or less versus 7 or greater) also correlated with prostatectomy grade (p <0.001). On multivariate analysis ploidy added
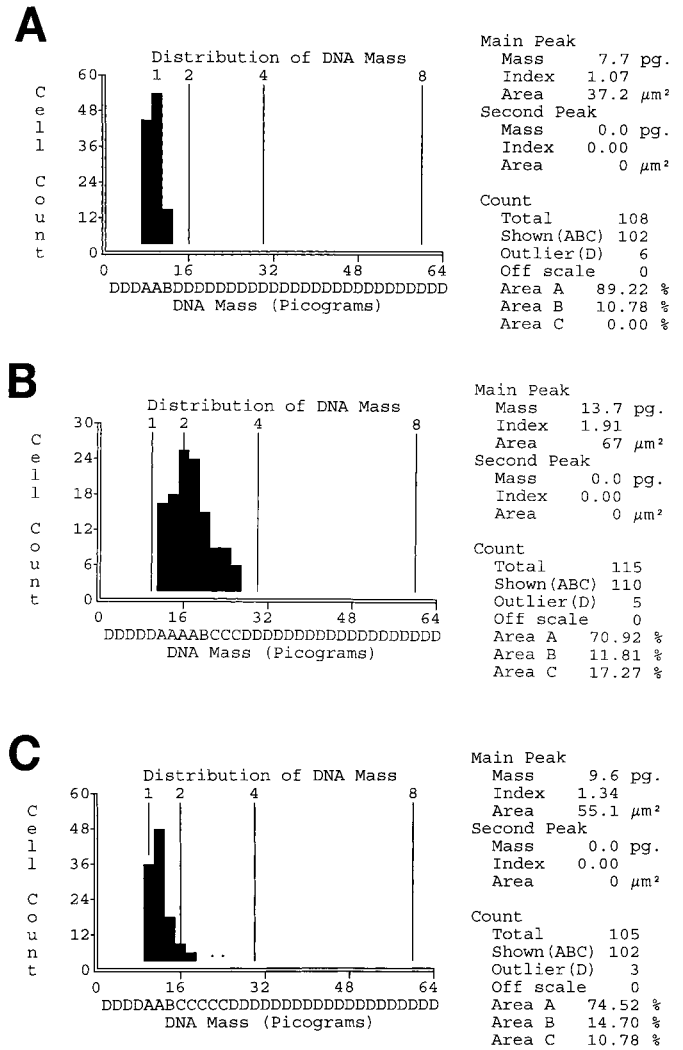


Representative histograms of diploid (*A*), tetraploid (*B*) and aneuploid (*C*) tumors.

marginal information (p = 0.04) to needle grade (p <0.0001) in the prediction of prostatectomy grade.

Table 3 shows the cases divided by which of the 2 factors on needle biopsy (Gleason score and ploidy) accurately predicted prostatectomy grade. In 97 cases (61%) both factors were predictive and in 16 (10%) neither factor was predictive. In 33 cases (21%) needle Gleason grade did and ploidy did not accurately predict prostatectomy grade, and in the remaining 13 (8%) needle ploidy was and needle Gleason grade was not predictive.

## DISCUSSION

Multiple studies have demonstrated a relationship between DNA ploidy and tumor behavior in prostate carcinoma. Studies cited in this discussion are not all inclusive but reflect some of the findings in this area of research. Investigators at the Mayo Clinic have shown that in radical prostatectomy patients who have nodal metastases ploidy of

TABLE 1. *Needle biopsy ploidy and status of subsequent radical prostatectomy*

| Needle Biopsy Ploidy | No. Cases (%) | % Organ Confined Disease |
|---|---|---|
| Diploid | 98 (62) | 69 |
| Tetraploid | 16 (10) | 50 |
| Aneuploid | 45 (28) | 44 |

TABLE 2. *Prediction of organ confinement by Gleason score and ploidy on needle biopsy*

| Needle Biopsy Findings | Pathological Stage % |
|---|---|
| Gleason score 6 or less | Organ confined (70) |
| Diploid | Organ confined (69) |
| Gleason score 7 or greater | Not organ confined (68) |
| Nondiploid | Not organ confined (54) |

TABLE 3. *Prediction of radical prostatectomy Gleason score by needle biopsy Gleason score and ploidy*

| Radical Prostatectomy Gleason Score | Needle Biopsy Gleason Score | Needle Biopsy Ploidy | No. Cases (%) | Needle Ploidy + Grade Concordance |
|---|---|---|---|---|
| 6 or Less | 6 or Less | Diploid | 76 (61) | Yes |
| 7 or Greater | 7 or Greater | Aneuploid | 21 | |
| | | | | |
| 7 or Greater | 6 or Less | Diploid | 11 (10) | Yes |
| 6 or Less | 7 or Greater | Aneuploid | 5 | |
| | | | | |
| 6 or Less | 6 or Less | Nondiploid | 25 (21) | No |
| 7 or Greater | 7 or Greater | Diploid | 8 | |
| | | | | |
| 7 or Greater | 6 or Less | Nondiploid | 10 (8) | No |
| 6 or Less | 7 or Greater | Diploid | 3 | |

the tumor provides prognostic information.[1] Furthermore, early hormonal therapy may offer a survival advantage in those patients with diploid tumors.[8] Ploidy can also help predict outcome in men with radiation refractory prostate cancer who have undergone salvage prostatectomy.[9] In a group of men treated surgically at Johns Hopkins Hospital nondiploid status of the radical prostatectomy specimen helped identify those with low grade tumors that progressed.[2] Borre et al studied ploidy in men diagnosed with cancer on transurethral resection or simple prostatectomy and treated without intent to cure.[3] Like the Hopkins group, they found that nondiploid status helped predict disease progression in a subset of men with low grade tumors.

In contrast, few studies have examined the usefulness of prostate needle biopsy ploidy. Song[10] and van den Ouden[11] et al analyzed ploidy on prostate needle biopsies of men treated nonsurgically. Both groups reported that needle ploidy correlated significantly with progression of disease. Ross et al studied needle biopsy ploidy in men treated with prostatectomy.[5] Aneuploid status on needle biopsy correlated with high histological grade on radical prostatectomy, extension of tumor outside the prostate and postoperative recurrence. In their study ploidy was more accurate than Gleason grade for predicting postoperative recurrence. However, the authors found no correlation between needle biopsy and radical prostatectomy specimen grades, and there was a high percentage of over grading of needle biopsy relative to the radical prostatectomy grade. These findings raise questions regarding the accuracy of the grade assigned to needle biopsy and radical prostatectomy.

Our study confirms the correlation between needle ploidy and stage. However, given accurate Gleason grading of the biopsy, ploidy did not provide additional information in the prediction of pathological stage. Virtually the same rate of organ confinement (69 versus 70%) was reported for tumors that were diploid or had lower Gleason score (6 or less) on needle biopsy. However, higher Gleason score (7 or greater) on needle biopsy was a better predictor of nonorgan confinement (68%) than nondiploid status (54%).

Preoperative prediction of the histological grade of prostate carcinoma within the radical prostatectomy specimen is also desirable as this grade independently correlates with adverse pathological conditions in the resection specimen and postoperative progression.[12, 13] However, sampling error and the difficulty in accurate Gleason grading of the small foci of tumors often seen on needle biopsy hinder the predictive power of needle biopsy grade. There are frequent discrepancies between needle and subsequent prostatectomy grades, with under grading of needle biopsies more common.[14, 15] A recent study by Ross et al demonstrated that needle biopsy ploidy can help predict "grade shifting" from needle biopsy to prostatectomy[6] but it is noteworthy that such discrepancies between needle and prostatectomy grades were much more common (34%) in their study than in ours (18%). In our study

ploidy as determined on needle biopsy added, albeit marginally, to needle Gleason score in the prediction of radical prostatectomy grade. In 61% of the cases both factors accurately predicted the grade at prostatectomy (6 or less versus 7 or greater). However, when the 2 factors were discordant, needle Gleason score was predictive (21%) more frequently than needle ploidy (8%) of the final grade.

## CONCLUSIONS

Although ploidy correlates with pathological stage and prostatectomy grade, with accurate grading of prostate cancer performance of DNA ploidy on needle biopsy does not add to the prediction of pathological stage or grade. Just as sampling error limits even the most careful needle Gleason grading, tumor heterogeneity and sampling error hinder ploidy on needle biopsy as a prognostic tool.[16, 17] Ploidy on biopsy may be useful in planning treatment when accurate Gleason grading of needle biopsy is a concern.

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