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Genomic Correlates to the Newly Proposed Grading Prognostic Groups for Prostate Cancer

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Abstract

Recommendations by the International Society of Urologic Pathology and 2016 World Health Organization blue book propose the use of a five-tiered prostate cancer (PCa) grading system. The five prognostic grade groupings (PGGs) ranging from 1 to 5 are defined as Gleason grades $\leq$6, 3 + 4, 4 + 3, 8, and >8, respectively. Recent work suggests that each group is associated with a distinct risk of biochemical PCa recurrence. In this study, we sought genomic support for PGGs using whole-exome and whole-genome sequencing data for 426 clinically localized PCas treated by radical prostatectomy. After adjustment for tumor purity for the sequencing data, we observed a significant frequency increase in genomic amplifications and deletions ($p = 0.013$) and in nonsynonymous point mutations ($p = 0.008$) with increasing risk group. Interestingly, PGG1 (low risk) was entirely haploid, whereas PGG2–5 exhibited increasing polyploidy frequency. Principal component analysis of genomic profiles revealed that PGG1, PGG2, and PGG3 represent distinct classes, but PGG4 and PGG5 exhibit genomic similarity. Together, these observations for the largest PCa genomic data set to date provide support for increasing genomic alterations with increasing PGG. This is the first genomic correlation of the PGG system. Future work will need to explore the clinical utility of PGGs in prospective studies with long-term follow-up.

Patient summary: Gleason grading for prostate cancer provides important information for guiding clinical care. A new proposal by leading pathologists favors translating Gleason grades into five risk categories. In this study, a comprehensive analysis of the largest genomic data set on prostate cancer to date, we demonstrate molecular support for this new five-tiered system.

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The Gleason grading system was empirically developed to correlate acinar tumor growth patterns with patient clinical outcomes [1]. This system is consistently one of the best independent predictors of clinical outcome for prostate cancer (PCa). Contemporary clinical practice has reduced the system to distinguishing two key patterns: tumors with and without Gleason pattern 4 or 5 (high-grade) disease, associated with the worst clinical outcomes. The system originally used a Gleason range of 2–10, going from low (2–6), to intermediate (7) and high (8–10) Gleason grade. More recently, the scale has shrunk to scores that now range from 6 to 10; it is exceptionally rare to see Gleason scores of 2–5 with contemporary grading. In addition to educational improvements in Gleason scoring, the advent of immunohistochemistry for basal cells has virtually eliminated diagnosis of Gleason patterns 1 and 2 [2]. In 2013, Epstein and colleagues [3] originally proposed reducing this system to five prognostic risk categories. More recently, a larger validation study supported the distinct risk of PSA biochemical recurrence based on each risk category [4]. Epstein and others performed a retrospective analysis of more than 19 000 consecutive men with localized PCa treated by radical prostatectomy between 2005 and 2014 at the Cleveland Clinic, Memorial Sloan Kettering Cancer Center, Johns Hopkins Hospital, University of Pittsburgh, and Karolinska Institute. They reported 5-yr biochemical risk-free survival for prognostic grade groups (PGGs) of 97.5% for PGG1, 93.1% for PGG2, 78.1% for PGG3, 63.6% for PGG4, and 48.9% for PGG5. On the basis of these data and discussions by expert genitourinary pathologists, urologists, oncologists, and other clinicians who care for PCa patients, a consensus group convened by the International Society of Urological Pathology proposed these risk categories for the new 2016 World Health Organization PCa reporting guidelines [2]. The PGG system complements the Gleason score and helps to emphasize where scores fit in a proposed risk stratification schema. Most importantly, the PGG system emphasizes that a Gleason score of 6 corresponds to low-risk cancer. This will have important implications for patients as they consider treatment options among surgery, radiation therapy, and active surveillance.

It follows that PGGs with increasing risk could also have genomic correlates. With the emergence of large, publicly available genomic data sets for PCa, we asked whether these proposed risk strata correspond to distinct genomic groups. To this end, we compiled a data set comprising 426 tumor/normal pairs from men with clinically localized PCa treated with radical prostatectomy as monotherapy. The tumors were subjected to either whole-exome or whole-genome sequencing [5–7]. Supplementary Table 1 lists demographic data and the Gleason score breakdown. This data set is the largest genomic PCa data set to date.

A moderate increase in median prostate-specific antigen (PSA) at diagnosis was observed with increasing risk category (linear regression, $p = 0.009$), whereas no association with median age at diagnosis was detected (linear regression, $p = 0.3189$). After adjustment for tumor polyploidy and purity on a sample basis [8] to allow fair comparison across tumors with diverse cellularity, we analyzed tumor genomic characteristics across risk groups (Supplementary Table 2). The overall number of nonsynonymous somatic mutations slightly increased across risk strata, reaching higher statistical significance when comparing lower (PGG1 and PGG2) with higher (PGG3, PGG4, and PGG5) risk groups (Wilcoxon-Mann-Whitney test, $p = 1.01 \times 10^{-4}$). Overall, more abundant somatic copy number alterations (gains and losses) were observed with increasing PGG (Fig. 1).

To investigate aberration frequency across PGGs on a gene basis, we focused on a set of 880 cancer genes (Supplementary material). For each aberrant gene we tested possible increasing, decreasing, invariant and mixed (or parabolic) trends (Supplementary material). Examples are shown in Figure 2A, where *MYC* amplification, *TP53* deletion, and 21q22.3 deletion near *TMPRSS2* represent increasing, mixed, and invariant frequencies, respectively. Of the 389 recurrently amplified genes (ie, increased somatic DNA copy number) in the data set, 91% exhibit increasing DNA copy number amplification frequency across PGGs; thus, their incidence correlates with higher PGG. This includes *NCOA1* (9–50% for PGG1–5) and *MYC* family members, with *MYC* reaching the highest frequency in PGG5 (45.5%), followed by *MYCN* (13.6%) and *MYCL* (6.8%). A similar situation was observed for 35% of the 756 recurrently deleted genes, whereas a large fraction (56%) exhibited an invariant trend across risk groups (Fig. 2B). Genomic regions with the steepest increase in deletion frequencies across PGGs included areas on 13q32 and 18q21. Interestingly, of the few recurrent somatic point mutations in hormone-naïve PCa, only *TP53* incidence increased across the PGGs (PGG1, 0%; PGG2, 7%; PGG3, 8%; PGG4, 10%, and PGG5, 9%). Complete results for the frequency trend analysis are listed in Supplementary Table 3. There was similar incidence across the groups for the most common aberrations in localized PCa, *ERG* rearrangement in terms of structural variants and *SPOP* mutations for single base changes. Frequency trend analysis was also explored for gene aberration within subsets of tumors defined by *ERG* rearrangement or *SPOP* mutation. The results are in line with the findings for the whole data set (Supplementary Figs. 1 and 2). Consistent with previous studies, while the number of aberrant genes slightly increased with patient age (linear regression using age quartiles, $p = 0.027$), *ERG* rearrangement was significantly enriched in the youngest group (58% for the lowest and 32% for the highest age quartile; hypergeometric test, $p = 0.0045$) [9].

PGG1, defined as Gleason score $\leq$6 (3 + 3), is the critical class to understand at a molecular level. Clinically, PGG1 patients with PSA $<$10 ng/ml and negative digital rectal examination (DRE) results are suitable candidates for active surveillance. The genomic analysis of PGG1 clearly confirms the presence of somatic aberrations (eg, the incidence of interstitial deletion between *TMPRSS2* and *ERG* is $\sim$20%) but reveals that none of the tumors exhibited polyploidy, a characteristic finding in 1.3% of PGG2, 3.1% of PGG3, 3.5% of PGG4, and 9.1% for PGG5 tumors (linear regression, $p = 0.02$), and hardly any amplification (Fig. 1).

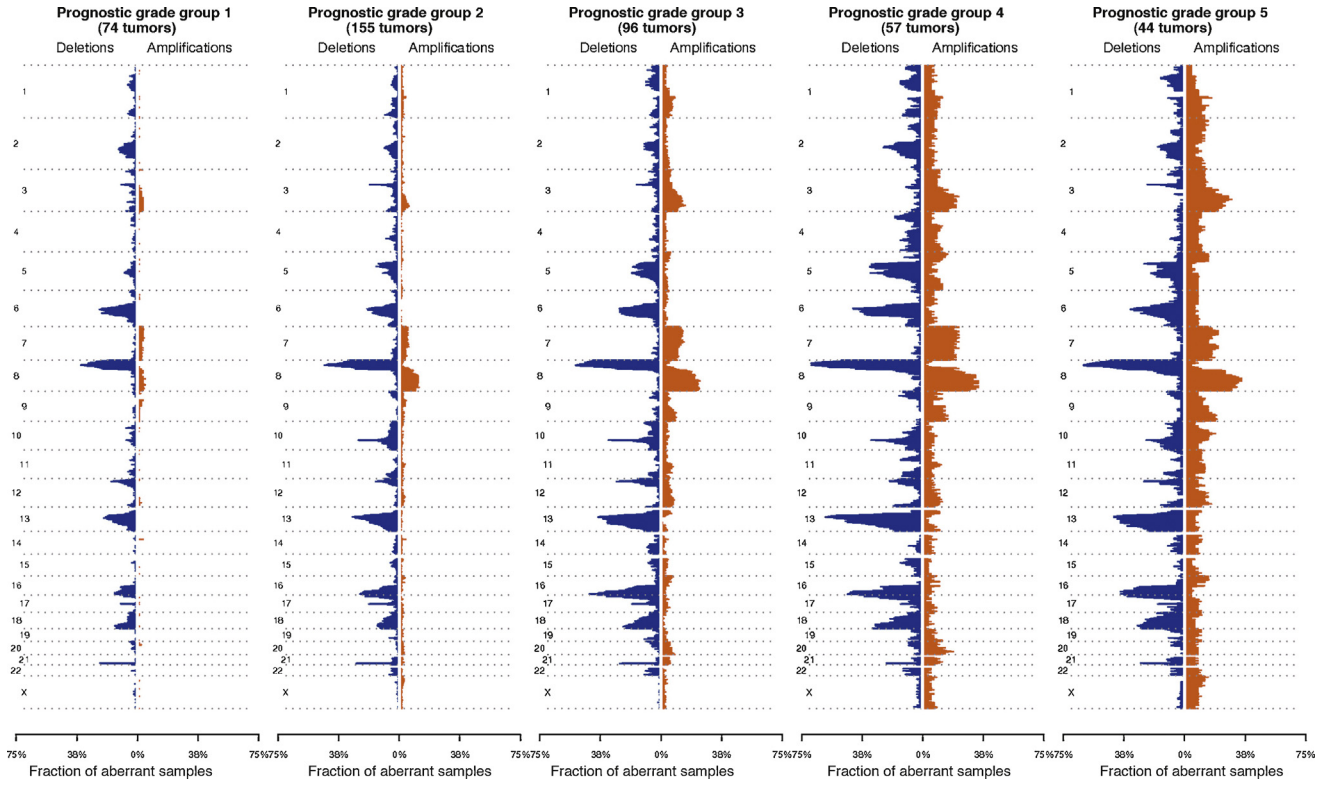

Fig. 1 – Landscape of somatic copy number alterations from 426 prostate cancer cases ordered by prognostic grading group from 1 (low) to 5 (high). Blue denotes deletions; red denotes amplifications.

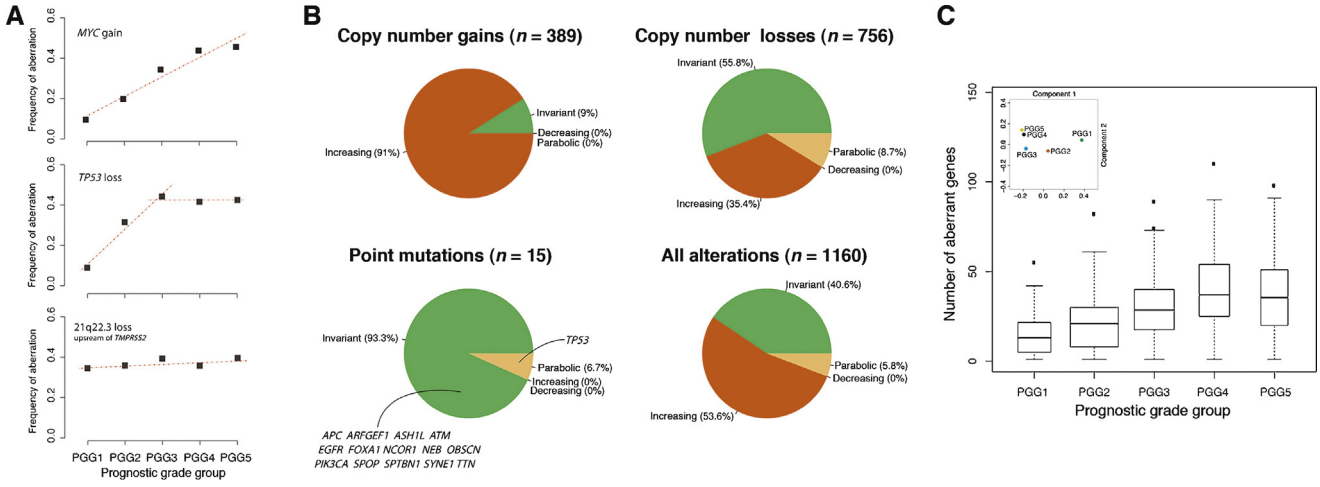

Fig. 2 – (A) Examples of aberration frequencies across prognostic grading groups (PGGs) on a gene basis. *MYC* amplification, *TP53* deletion and 21q22.3 deletion near *TMPRSS2* represent increasing, mixed, and invariant frequencies, respectively. (B) Summaries of frequency trends detected for copy number alterations on a gene basis across PGGs. (C) Number of aberrant genes per patient tumor across PGGs. Principal component analysis demonstrates that PGG4 and PGG5 have significant overlap (inset).

Finally, we investigated the potential of aberrant cancer genes to distinguish PGGs. Focusing on the merged gene set obtained from the top 100 aberrant genes per PGG (resulting in 147 genes; Fig. 2C) and applying principal component analysis to the whole data set, we observed no significant distinction between PGG4 and PGG5, whereas PGG1 appears to be clearly distinct from the other categories (Fig. 2C inset).

In summary, the proposed new risk categories take into account the reality of clinical practice in 2015. Although PGGs are not initially meant to replace Gleason scores, they provide a useful clinical perspective in discussing risk. Using emerging genomic data, the current study supports the notion that PGGs also correspond to genomic events. Importantly, the lowest risk group (PGG1) supports favorable genomic features such as a lack of polyploidy

and fewer driver mutations. PGG1 patients with low PSA and negative DRE represent the type of patients who may be ideal for active surveillance. Interestingly, PGG4 and PGG5 do not significantly differ from one another. However, recent clinical studies suggest that these groups do have different clinical outcomes [10]. It is possible that epigenetic changes or other alterations that are yet to be described are responsible for the clinical difference.

One major limitation in the development of the PGG system is that PSA biochemical recurrence was used to determine levels of risk. Recent work does not support PSA recurrence as a surrogate for disease progression [11]. Therefore, future studies will be needed in the setting of active surveillance with long-term observations to help in supporting the PGG system and potential genomic correlates of risk. Ultimately, if verified over time with additional studies, the PGG system could replace Gleason scores.

Author contributions: Mark A. Rubin and Francesca Demichelis had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

Study concept and design: Demichelis, Rubin.
Acquisition of data: Demichelis, Girelli.
Analysis and interpretation of data: Demichelis, Girelli, Rubin.
Drafting of the manuscript: Demichelis, Rubin.
Critical revision of the manuscript for important intellectual content: Demichelis, Girelli, Rubin.
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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.eururo.2015.10.040.

References

[1] Gleason DF. Histologic grading of prostate cancer: a perspective. Hum Pathol 1992;23:273–9.
[2] Moch H, Humphrey PA, Ulbright TM, Reuter VE, editors. WHO classification of tumours of the urinary system and male genital organs. ed. 4. Lyon: IARC; 2016 in press.
[3] Pierorazio PM, Walsh PC, Partin AW, Epstein JI. Prognostic Gleason grade grouping: data based on the modified Gleason scoring system. BJU Int 2013;111:753–60.
[4] Epstein JI, Zelefsky MJ, Sjoberg DD, et al. A contemporary prostate cancer grading system: a validated alternative to the Gleason score. Eur Urol 2016;69:428–35.
[5] Baca SC, Prandi D, Lawrence MS, et al. Punctuated evolution of prostate cancer genomes. Cell 2013;153:666–77.
[6] Barbieri CE, Baca SC, Lawrence MS, et al. Exome sequencing identifies recurrent SPOP, FOXA1 and MED12 mutations in prostate cancer. Nat Genet 2012;44:685–9.
[7] The Cancer Genome Atlas Network. The molecular taxonomy of primary prostate cancer. Cell 2015;163:1011–25.
[8] Prandi D, Baca SC, Romanel A, et al. Unraveling the clonal hierarchy of somatic genomic aberrations. Genome Biol 2014;15:439.
[9] Schaefer G, Mosquera JM, Ramoner R, et al. Distinct ERG rearrangement prevalence in prostate cancer: higher frequency in young age and in low PSA prostate cancer. Prostate Cancer Prostatic Dis 2013;16:132–8.
[10] Tsao CK, Gray KP, Nakabayashi M, et al. Patients with biopsy Gleason 9 and 10 prostate cancer have significantly worse outcomes compared to patients with Gleason 8 disease. J Urol 2015;194:91–7.
[11] Xia J, Trock BJ, Gulati R, et al. Overdetection of recurrence after radical prostatectomy: estimates based on patient and tumor characteristics. Clin Cancer Res 2014;20:5302–10.