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The Grey Zone

Urothelial Cancer: What Is the Role of Expression-based Subtypes to Guide Neoadjuvant Therapy in Muscle-invasive Bladder Cancer?

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1. Introduction

Most epithelial tumors can be grouped by functional architecture. The bladder lining is a pseudostratified epithelium with the basal or renewal cells near the deeper or connective tissue layer and likely the source of progenitor epithelial cells. These basal cells express KRT5, KRT14, and CD44. In contrast, epithelial cells of the differentiated layers reside closer to the bladder lumen (hence, "luminal") [1]. Luminal cells express epithelial differentiation markers—UPK2 and GATA3. While not all tumors fit clearly into a subtype, expression-based subtyping with RNA sequencing or microarray is used to profile a tumor's transcriptome and assign a patient's tumor to a subtype.

2. Why are there so many subtyping systems? Do these matter? What are the limitations?

The degree of heterogeneity, mostly from immune cells and fibroblasts, can make subtype classification less than ideal, and placing these heterogeneous tumors into one subtype (eg, 51% basal and 49% luminal) makes precision therapy more challenging [2]. A tumor's transcriptome can change over time, and the subtype can evolve. For example, in a study of paired tumors treated with neoadjuvant chemotherapy, 41% of luminal tumors switched to a basal subtype [3]. In the VESPER trial, 48% of tumors had mixed subtypes and, regardless of their subtype, had a worse outcome (hazard ratio 2.0, 95% confidence interval 1.36–3.0) [4]. Thus, a significant limitation of subtyping is the presence of tumor heterogeneity and plasticity. At this point, subtypes are descriptive, and there are limited data to say which subtype call is a "ground truth" by which others can be standardized. There are many different methods for clustering tumors because there is no single "right" way to group them. The main characteristics of a subtyping system are that it (1) is reproducible with multiple approaches, (2) is methodologically defensible, and (3) identifies potential differences in biology. With more tumors, the ability to identify subtle similarities becomes feasible, and some of the rarer biological features can be identified.

3. What is the "holy grail" of subtyping?

The goal of subtyping is to reproducibly group tumors that result in a specific outcome (eg, response to therapy). In The Cancer Genome Atlas (TCGA), for example, in which no therapy was described, the outcome was prognosis. If we agree that TCGA or consensus-based classification is the best system, the goal would be to match each subtype with the best or most active therapy. In this way, tumor subtypes could be applied for precision oncology to select the best therapy for a patient. An alternative strategy is to provide a rational framework for intensification or deintensification based on the behavior of a subtype. An example of the limited frequency is advanced-stage (pT3+) luminal tumors [5].

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Yet, despite a similar rate of nodal metastasis in luminal and nonluminal subtypes, this strategy has not been adopted widely.

4. Is there any evidence that subtypes can guide response to perioperative therapy?

The answer is both yes and no. Tumors with more immune cells and more exhaustion markers (eg, luminal infiltrated and basal tumors) tend to respond better to immunotherapy. Multiple retrospective analyses of neoadjuvant chemotherapy trials have shown a similar rate of pathologic response across all subtypes. With differences in survival across subtypes, one tumor may have a less pronounced pathologic response but "improved" event-free or overall survival. A prospective trial was started using the Veracyte platform based on the TCGA 2017 subtyping (Gene expression subtypes of Urothelial carcinoma: Stratified Treatment and Oncological outcomes [GUSTO]) and is currently enrolling [6,7]. We developed a subtype-directed trial largely using immunotherapy combinations, but the muscle-invasive bladder cancer field moved so quickly that a "comparison" arm was not feasible, so the trial was ultimately not initiated.

5. With enfortumab vedotin and pembrolizumab as the new standard, does subtyping matter?

The answer to this question is unknown. There is some evidence that luminal tumors express more Nectin4 [8], and enfortumab may be more active in patients with luminal biology. Translational research from clinical trials demonstrating the efficacy of enfortumab vedotin and pembrolizumab (EVP) should evaluate pretreatment tumors for response by subtype. Unfortunately, almost no translational research has been performed, and the only biomarker research was performed by an investigator with samples and collections of samples [9]. With a complete pathologic response rate of 57% for muscle-invasive bladder cancer patients treated with EVP, is there any reason to consider subtyping [10]? Probably not as much, but can subtyping be used to decrease the number of neoadjuvant or adjuvant doses of perioperative therapy? Perhaps more responsive subtypes can be used to identify patients for bladder preservation. We implore sponsors of these groundbreaking trials to consider investment in biomarker evaluation.

6. Conclusions

Tumor subtypes originate from the heterogeneity of bladder cancer. This heterogeneity influences the response to treatment. A retrospective analysis of clinical trials should be conducted to identify which subtypes may respond better to perioperative therapy regimens. These findings could guide prospective, comprehensive biomarker trials to enhance the precision of perioperative therapy.

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