Commentary

Simple and Efficient Stratification of Invasive Bladder Cancer Patients

Jerome Zoidakis

Proteomics Laboratory, Biomedical Research Foundation Academy of Athens, Athens 11527, Greece

ARTICLE INFO

Bladder cancer (BC) is the 4th most common cancer in men and the 11th most common in women (Kamat et al., 2016). The majority of bladder carcinomas (~90%) are non-invasive papillary tumors that are treated by transurethral resection followed by intravesical BCG installations or chemotherapy (Kiselyov et al., 2016). The superficial papillary tumors rarely progress to invasive carcinomas but they have a high propensity for recurrence. This necessitates a lifetime of invasive surveillance by repeated cystoscopy.

On the other hand, non-papillary tumors have a high tendency for aggressive invasive growth and are associated with poor 5-year survival ranging from 30 to 50% (Kiselyov et al., 2016). Current therapeutic options in muscle-invasive BC are limited. Apart from cisplatin-based chemotherapy and cystectomy, only one other treatment (anti-PDL1) has been approved in the past 30 years. Moreover, reliable biomarkers for patient stratification are not currently available. Ongoing clinical trials and targeted therapy studies are aiming to appropriately develop novel treatments and assess the performance of prognostic and predictive biomarkers for patients suffering from invasive BC (Anon, 2014; Mbeutcha et al., 2016; Knollman et al., 2015).

Invasive BC is heterogeneous at the molecular level and can be grouped into 2 main subtypes (basal and luminal) with distinct clinical behaviors and responses to chemotherapy. The basal subtype expresses a signature similar to an undifferentiated basal layer of normal urothelium with frequent features of squamous differentiation along with up-regulation of markers characteristic of epithelial-to-mesenchymal transition (EMT). They also show overexpression of hypoxia induced factor 1 (HIF-1) and epidermal growth factor receptor (EGFR) with many of its ligands. The luminal subtype comprises papillary non-invasive tumors and a subset of invasive urothelial cancers. In the majority of these tumors, the PI-3 kinase/AKT pathway is altered (McConkey et al., 2015). Luminal tumors express markers characteristic of intermediate and luminal urothelial differentiation, high levels of

fibroblast growth factor receptor-3 (FGFR3) and activating FGFR3-, ERBB2-, ERBB3- and PIK3CA-mutations (Kamat et al., 2016; McConkey et al., 2015; Choi et al., 2014). BC patients with invasive basal tumors exhibit significantly lower survival rates compared to patients suffering from invasive luminal tumors (Choi et al., 2014). This observation is convincingly proven by Dadhania et al. by analysis of survival rates of BC patients in several large cohorts (Dadhania et al., 2016).

A luminal subtype termed "p53-like", is characterized by high levels of infiltrating stromal cells, particularly cancer-associated fibroblasts. These tumors are enriched with extracellular matrix (ECM) biomarkers and are resistant to neoadjuvant cisplatin-based chemotherapy. This chemoresistance is probably due to the rather "quiescent" state of those tumors compared to the other subtypes. A similar link between quiescence and chemoresistance has been observed in luminal breast cancers which share many features with BC "p53-like" tumors. There is also a basal aggressive subtype termed "claudin-low" with strong mesenchymal signature. This subtype is characterized by stem cell-like properties, down-regulation of claudins 3, 4 and 7 and up-regulation of EMT markers (Kamat et al., 2016; McConkey et al., 2015; Damrauer et al., 2014).

Dadhania et al. (Dadhania et al., 2016) report transcriptomics data that clearly define the basal and luminal subtypes on three large cohorts of fresh-frozen BC tissue samples. The authors demonstrate that the "p-53 like" transcriptomic signature is present in a subset of both luminal and basal tumors. They also show that the "claudin-low" subtype does not express luminal or basal markers, so they define it as "double negative".

Subsequently, the authors established a protocol based on IHC for easy assignment of BC tumors to the basal or luminal subtype using a cohort of formalin-fixed paraffin-embedded BC tissue samples. Basal tumors expressed high levels of Keratin 5/6 whereas luminal tumors were characterized by significant GATA3 overexpression. Additionally, the IHC analysis indicated that the "p53-like" molecular features do not originate from cancer cells but are due to stromal cells. Therefore the "p53-like" subtype does not appear to be an "intrinsic" BC subtype. Further experimental validation by laser capture microdissection and transcriptomics analysis of the cancer and stromal components of bladder sections is necessary in order to confirm this observation.

The simple IHC-based classification of BC tumors proposed by Dadhania et al. (Dadhania et al., 2016) has the potential to assist physicians in identifying patients suffering from highly aggressive basal tumors. Unfortunately, the two biomarkers reported cannot identify double negative tumors (5% of invasive BC cases) that exhibit poor survival rates. A subsequent step would be to develop a support-vector

machine (SVM) approach for combining the scores of additional IHC markers for efficient discrimination of double negative tumors.

Preclinical data have demonstrated that basal tumors are sensitive to EGFR inhibitors. However, EMT as a characteristic of basal tumors confers resistance to EGFR inhibitors in BC cells. Therefore, epigenetic agents (e.g. histone deacetylase inhibitors) could be used to reverse EMT and promote sensitivity to EGFR inhibitors in those tumors (McConkey et al., 2015). Additionally, there are many clinically available agents to inhibit HIF-1 pathway in basal tumors (e.g. inhibitors of the proteasome, mammalian target of rapamycin, vascular endothelial growth factor receptor-2 (VEGFR-2)). Recently, a combination of VEGF or VEGFR-2 inhibitors with conventional chemotherapy in patients with basal tumors yielded satisfactory results (McConkey et al., 2015).

For luminal tumors, small molecule and blocking antibody FGFR inhibitors are being evaluated in patients and clinical trials with pan-ERBB inhibitors are being planned (McConkey et al., 2015). Moreover, there is evidence that targeting the PI-3 kinase/AKT/mTOR pathway could produce strong clinical responses in patients with luminal tumors (Kiselyov et al., 2016; McConkey et al., 2015).

Up-regulation of cytotoxic T-lymphocyte-associated protein 4 (CTLA-4), programmed death-ligand 1 (PD-L1) and programmed cell death protein 1 (PD-1) in either the tumor itself or the surrounding immune cells, has been associated with BC's ability to escape the cell-killing effects of treatment (Kamat et al., 2016). Drugs targeting those molecules have demonstrated greater response rates and better tolerance than platinum-based chemotherapy. Patients with luminal "p-53 like" tumors responded better to anti-PD-L1 therapies than basal tumors (Kamat et al., 2016). As alluded to above, in May 2016, the anti-PD-L1 drug atezolizumab was approved by the FDA for the treatment of progressing metastatic BC during or after platinum-based chemotherapy, representing the first drug approval for muscle-invasive bladder cancer in decades (Kamat et al., 2016; Drake et al., 2016).

The data presented by Dadhania et al. (Dadhania et al., 2016) open the way for efficient stratification of BC tumors into luminal and basal subtypes. The clinical application of these findings is important since patients will benefit from recently developed targeted therapies.

Disclosure

The author declared no competing interests.

References

Anon, 2014. Comprehensive molecular characterization of urothelial bladder carcinoma. Nature 507 (7492), 315–322.

Choi, W., et al., 2014. Identification of distinct basal and luminal subtypes of muscle-invasive bladder cancer with different sensitivities to frontline chemotherapy. Cancer Cell 25 (2), 152–165.

Dadhania, V., et al., 2016. Meta-analysis of the luminal and basal subtypes of bladder cancer and the identification of signature immunohistochemical markers for clinical use. EBioMedicine 12, 105–117.

Damrauer, J.S., et al., 2014. Intrinsic subtypes of high-grade bladder cancer reflect the hallmarks of breast cancer biology. Proc. Natl. Acad. Sci. U. S. A. 111 (8), 3110–3115.

Drake, C.G., Bivalacqua, T.J., Hahn, N.M., 2016. Programmed cell death ligand-1 blockade in urothelial bladder cancer: to select or not to select. J. Clin. Oncol. http://dx.doi.org/10.1200/JCO.2016.68.4696.

Kamat, A.M., et al., 2016. Bladder cancer. Lancet http://dx.doi.org/10.1016/S0140-6736(16)30512-8.

Kiselyov, A., Bunimovich-Mendrazitsky, S., Startsev, V., 2016. Key signaling pathways in the muscle-invasive bladder carcinoma: clinical markers for disease modeling and optimized treatment. Int. J. Cancer 138 (11), 2562–2569.

Knollman, H., et al., 2015. Muscle-invasive urothelial bladder cancer: an update on systemic therapy. Ther. Adv. Urol. 7 (6), 312–330.

Mbeutcha, A., et al., 2016. Current status of urinary biomarkers for detection and surveillance of bladder cancer. Urol. Clin. North Am. 43 (1), 47–62.

McConkey, D.J., Choi, W., Dinney, C.P., 2015. Genetic subtypes of invasive bladder cancer. Curr. Opin. Urol. 25 (5), 449–458.