Application of liquid biopsies to identify genomic factors associated with therapy resistance in castration resistant prostate cancer

Daniel J. Crona\textsuperscript{1,2}, Young E. Whang\textsuperscript{2,3} \\
\textsuperscript{1}Division of Pharmacotherapy and Experimental Therapeutics, Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, North Carolina, USA; \textsuperscript{2}Lineberger Comprehensive Cancer Center, University of North Carolina, Chapel Hill, North Carolina, USA; \textsuperscript{3}Division of Hematology and Oncology, Department of Medicine, School of Medicine, University of North Carolina, Chapel Hill, North Carolina, USA

Correspondence to: Young E. Whang, MD, PhD. Division of Hematology/Oncology, Physicians Office Building, 3rd Floor, 170 Manning Drive, CB# 7305, Chapel Hill, NC 27599-7305, USA. Email: ywhang@med.unc.edu.

Submitted Oct 04, 2016. Accepted for publication Oct 08, 2016. 

doi: 10.21037/atm.2016.10.39

View this article at: http://dx.doi.org/10.21037/atm.2016.10.39

For over 70 years, androgen deprivation therapy (ADT) has been a fundamental treatment paradigm for advanced prostate cancer. Despite initial effectiveness, castration resistant prostate cancer (CRPC) develops in virtually all patients while on ADT. Until recently, additional available treatment options were limited for CPRC patients. Recognition of the critical role of reactivated androgen receptor (AR) signaling in CRPC led to the development of two novel AR axis-targeting agents, abiraterone and enzalutamide. Abiraterone is a selective inhibitor of the enzyme CYP17A1 that catalyzes a critical step in androgen biosynthesis (1). Inhibition of CYP17A1 reduces available ligands that stimulate AR in CRPC tumor cells. Enzalutamide is a potent inhibitor of ligand binding to AR and leads to loss of AR-induced transcriptional activation (2). Treatment with abiraterone or enzalutamide improved CPRC clinical outcomes (3,4), but CRPC remains a terminal disease due to resistance to these agents. Approximately 20–40% of chemotherapy-treated patients, and at least 10% of chemotherapy-naïve patients present with primary resistance to these agents (i.e., no initial decrease in prostate specific antigen (PSA) within three months of initiating treatment) (3,5). Additionally, CRPC patients who experience an initial PSA response to either abiraterone or enzalutamide will eventually develop secondary resistance (6). Also, despite differing mechanisms by which these medications affect the AR signaling axis, there is likely cross-resistance between abiraterone and enzalutamide (7,8), which further complicates the treatment landscape for CRPC. Clearly, there is an urgent need to identify and validate predictive biomarkers of treatment response or resistance that can effectively guide selection of therapeutic agents.

Mechanisms mediating resistance to abiraterone and enzalutamide are likely complex (9). Multiple etiologies that underlie primary and secondary resistance have been postulated. They include systemic and intratumoral androgen biosynthesis up-regulation, AR nonsynonymous point mutations, increased AR copy number, AR overexpression, expression of AR splice variants, and increased transcriptional co-activator expression. Glucocorticoid receptor overexpression, neuroendocrine differentiation and immune system deregulation have also been implicated in medication resistance in CRPC (10,11). These mechanisms have been characterized primarily in pre-clinical models and in many cases will require further validation in clinical specimens.

Recently, Romanel et al. provided new insights into mechanisms of abiraterone resistance through the use of circulating tumor DNA (ctDNA) to interrogate the AR genomic landscape in CPRC and identify AR variants associated with abiraterone resistance (12). After ctDNA samples were extracted from plasma, next-generation sequencing of the AR coding regions was performed on a total of 274 plasma samples from 97 CPRC patients treated with abiraterone. A total of 217 plasma samples from 80 of 97 patients (82%) possessed sufficient ctDNA (defined as tumor fraction ≥0.075) to determine AR copy number
accurately. The investigators analyzed samples collected prior to the patients initiating abiraterone, while on abiraterone, and after disease progression (12).

AR copy number gain was found in 81 of 217 samples (37%) and in 32 of 80 patients (40%) starting abiraterone. Somatic AR nonsynonymous point mutations were detected in 41 plasma samples (15%) collected from 16 patients. A significant inverse correlation between AR copy number and AR nonsynonymous point mutations was shown (P=0.004), and this result suggests that AR gain and AR point mutations may be mutually exclusive pathways. There was no change in AR copy number status from pre-treatment to progression. However, they demonstrated that two AR nonsynonymous point mutations (L702H and T878A) were associated with resistance to abiraterone (12). These two AR point mutations are activated by non-androgenic ligands (e.g., glucocorticoids and progesterone) present in patients treated with abiraterone (12-14). The investigators reported that patients with aberrant AR (AR gain, or L702H or T878A point mutations) were 4.9 and 7.8 times less likely to experience ≥50% and ≥90% PSA decline. Patients with aberrant AR also experienced significantly decreased overall survival (OS) (with hazard ratio of 7.33; P=1.3X10⁻⁶) and progression free survival (PFS) (hazard ratio of 3.73; P=5.6x10⁻⁷), when compared to patients with normal plasma AR. Finally, among the patients who had previously received treatment with enzalutamide or orteronel (another CYP17A1 inhibitor), 50% had AR gain and only 3 (of 23) patients experienced a ≥50% PSA decline after abiraterone treatment, which supports the hypothesis of a common mechanism of cross-resistance among AR axis-targeting agents (12).

A traditional barrier to better understanding the genomic and mechanistic etiologies underlying resistance to abiraterone and other agents used in the treatment of CRPC has been the difficulty in obtaining serial tumor biopsies from metastatic sites, which include the bones and abdominal lymph nodes. However, identifying and validating predictive biomarkers of treatment efficacy and resistance will help guide clinician treatment decision-making. “Liquid biopsies” [e.g., ctDNA isolated from blood or analysis of circulating tumor cells (CTCs)] are an attractive option for clinicians because they are convenient and minimally-invasive to the patient, can help identify multiple potentially actionable genomic aberrations present in a patient, and can identify the presence of clinically-actionable variants and guide selection of targeted therapy agents. They are also advantageous because they are dynamic in their ability to track genomic changes across time, in response to treatment, and thereby may help elucidate important mechanisms of primary resistance or acquired secondary resistance to therapy.

In addition to the report by Romanel et al., other publications have highlighted the use of liquid biopsies to identify variants that impact AR axis signaling, therapy response and resistance (6,14-16). These studies demonstrate the ability of investigators to use ctDNA samples to detect AR gain and AR nonsynonymous point mutations, and then relate them to clinical outcomes. Azad et al. and Wyatt et al. utilized array comparative genomic hybridization to measured AR copy number gain (15,16), which differed from Romanel et al. who used next-generation sequencing. Azad et al. were able to detect AR copy number gain in 45% of their patients (28 out of 62), while Wyatt et al. were able to detect an increase in 30% of patients (19 out of 63). Similarly to Romanel et al., CRPC patients with AR copy number gain in these two studies also experienced significantly worse clinical outcomes (PSA response and PFS) (15,16).

In addition to AR gain and point mutations, the expression of AR splice variants, which lack the ligand binding domain and are constitutively active, offer an alternative mechanism of medication resistance in CRPC. Antonarakis et al. demonstrated that detection of AR-V7 mRNA expression in CTCs from CRPC patients predicts for resistance to enzalutamide and abiraterone (6). In this study, 31% of the enzalutamide-treated patients and 19% of the abiraterone-treated patients had detectable AR-V7 mRNA in CTCs. In both abiraterone-treated patients and enzalutamide-treated patients, AR-V7-positive patients experienced significantly lower PSA response rates and significantly shorter clinical or radiographic PFS and OS, when compared to AR-V7-negative patients. This small study demonstrates the viability of liquid biopsies to detect clinically significant splice variants, and also highlights a fundamental difference between ctDNA and CTCs. ctDNA is composed of nucleic acid fragments not associated with cells while CTCs are intact cells (17). Moreover, the investigators from that study noted a significant association between increased full-length AR mRNA and the presence of AR-V7 splice variant (6). It may be hypothesized that an increase in AR transcripts, due to AR amplification, could cause a subtle shift in mRNA splicing, leading to the production of splice variants in tumors with AR amplification. However, uncovering the mechanisms that underlie this association and also determining the relative
contribution of AR aberration detected by ctDNA versus AR splice variant (AR-V7 and potentially additional clinically-relevant splice variants) expression in CTCs, in mediating resistance to AR axis targeting agents, need to be addressed in future work. Finally, while ctDNA has been shown useful to elucidate potential mechanisms of resistance to abiraterone (12,15) and to enzalutamide (16), validation of these results in larger cohorts of CRPC patients is still needed. Furthermore, applications of liquid biopsies in identification and validation of predictive biomarkers of treatment response and/or resistance in novel agents in the drug development pipeline will be crucial in improving clinical outcomes for CRPC patients.

In conclusion, the article by Romanel et al. represents an important advancement in the application of liquid biopsies as an approach to develop clinically useful predictive biomarkers for guiding treatment selection in CRPC.

Acknowledgments

The authors would like to thank Dr. Lana Crona for her help in reviewing and editing this invited Guest Commentary.

Footnote

Provenance: This is an invited Guest Commentary commissioned by Section Editor Hui Kong, MD, PhD (Department of Respiratory Medicine, The First Affiliated Hospital of Nanjing Medical University, Guangzhou Road, Nanjing, China).

Conflicts of Interest: Y.E.W. received research funding from Astellas and Janssen and Tokai. D.J.C. stated no conflicts of interest.


References


Cite this article as: Crona DJ, Whang YE. Application of liquid biopsies to identify genomic factors associated with therapy resistance in castration resistant prostate cancer. Ann Transl Med 2016;4(Suppl 1):S64. doi: 10.21037/atm.2016.10.39