Unsuspected Protumorigenic Signaling Role for the Oncometabolite GABA in Advanced Prostate Cancer

Renea A. Taylor1,2 and Matthew J. Watt3

Elucidating the events that underpin the transition from androgen-dependent to castrate-resistant prostate cancer (CRPC) remains a clinical challenge. In this issue of Cancer Research, Gao and colleagues identify that the γ-aminobutyric acid (GABA) shunt is upregulated with the onset of CRPC, via phosphorylation and activation of glutamate decarboxylase (GAD) 65. Overproduction of GABA, an oncometabolite, can directly regulate nuclear androgen receptor signaling to drive tumorigenesis, thereby providing a link between aberrant metabolism and protumorigenic signaling in advanced prostate cancer. The findings from this study support exploring the GABA shunt, GAD65 in particular, as a molecular target in the treatment of CRPC.

See related article by Gao et al., p. 4638

Despite significant advances in diagnostic tools and curative treatments, prostate cancer remains a lethal condition for those who develop advanced disease. Hence, the transition from androgen-dependent to castrate-resistant prostate cancer (CRPC) remains a major clinical challenge, with novel insights into the pathogenesis of disease providing new molecular targets for therapeutic development. Gao and colleagues present a tour de force of molecular biology, cell biology, and metabolic analyses to unravel the essential elements of the γ-aminobutyric acid (GABA) shunt that is upregulated with the emergence of CRPC (1). The major conceptual advancement of this study was that overproduction of GABA, a metabolic intermediate or oncometabolite, can directly regulate nuclear androgen receptor (AR) signaling to drive prostate cancer, thereby providing a link between aberrant metabolism, protumorigenic signaling, and advanced disease.

There has been an explosion of interest in deciphering the intricacies of metabolism in cancer, first to understand the pathways that are invoked to support tumorigenesis but also to identify so-called metabolic vulnerabilities that could be exploited as an adjunct to improve efficacy of existing therapeutics. Prostate cancer is characterized by marked metabolic remodeling, highlighted by increased dependency of prostate tumors on de novo lipogenesis (2), fatty acid uptake and metabolism (3), and glutamine uptake (4). While this is well-demonstrated in early-stage hormone-sensitive prostate cancer, the metabolic changes in advanced castrate-resistant disease were less well-characterized.

GABA is a four-carbon nonprotein amino acid metabolite, best known as the major inhibitory neurotransmitter in the mammalian brain (5) and negative regulator of glucose-dependent GABA and forming succinic semialdehyde, which eventually results in the production of succinate and NADH for energy production.

Gao and colleagues (1) demonstrate that the GABA shunt is upregulated through activation of glutamate decarboxylase (GAD) 65, which is one of two enzymes that catalyze the decarboxylation of glutamate to GABA. This key observation was made using clinically relevant patient-derived xenografts (PDX) of locally advanced or metastatic prostate tumors, experimentally manipulated to transition from androgen-dependent to CRPC. Activation of GAD65 was dependent on phosphorylation of serine 6, which appeared to result from increased PI3K-PKC signaling. The authors showed that GAD65 S6 phosphorylation was basically absent in tumors from men with untreated androgen-dependent prostate cancer but was detected in approximately 80% of cells in men with CRPC. This is consistent with previous studies that reported hyperactivation of PI3K/AKT/mTOR signaling in approximately 40% of early prostate cancer cases and 70%–100% in advanced disease (7). Moreover, PKCe levels are markedly elevated in prostate cancer compared with benign prostatic epithelia, and its overexpression has been associated with disease recurrence. While PKCe plays an important role in regulating the growth and survival of LNCaP prostate cancer cells (8), its role in human CRPC remains unknown.

Critically, the increase in GAD65 activity in the study by Gao and colleagues (1) was linked to increased GABA production and accumulation in CRPC PDXs, and the importance of the GABA shunt was confirmed by silencing GAD65 activity in vitro, which attenuated cell proliferation in CRPC. Alterations in metabolism often confer a growth and survival advantage for malignant tumors. Intriguingly, silencing GAD65 did not affect substrate
metabolism or bioenergetics in CRPC, suggesting that activation of the GABA shunt and GABA accumulation induced tumorigenesis through a mechanism independent of altered energy metabolism.

A hallmark of CRPC is reactivation of the AR transcription factor, which occurs via several mechanisms including mutation, amplification, and rearrangement of the AR gene, transcriptional compensation by alternative steroid receptors (e.g., estrogen and glucocorticoid receptors), mutation or copy number alteration of genes encoding AR coregulators, and the elevated expression of glucocorticoid receptors, mutation or copy number alteration of genes encoding AR coregulators, and the elevated expression of glucocorticoid receptors). This provides novel insight into a mechanism by which AR transactivation is maintained in CRPC. Moreover, while it is known that alterations in AR signaling induce changes in cancer metabolism that are associated with poor prognosis, these data provide a new, reciprocal perspective that alterations in metabolism alter AR signaling to promote CRPC.

This comprehensive and intriguing study raises multiple points for therapeutic intervention of this pathway for patients with advanced prostate cancer. These include: (i) direct inhibition of GAD65 protein content, for example, by RNA interference; (ii) preventing phosphorylation of GAD65 at serine 6 or upstream kinase activity (i.e., PKCε) to reduce GAD65 activity; (iii) further considering the role of GABA-degrading enzymes, such as GABA-transaminase and succinic semialdehyde dehydrogenase, to decrease GABA accumulation; (iv) preventing GABA binding to AR by targeting the molecular docking site, and (v) targeting ZNHIT3 binding to the hinge region of AR, which is essential for AR nuclear retention in CRPC. In addressing the first option, inducible knockdown of GAD65 markedly reduced growth of CRPC PDX tumors in vivo, characterized by reduced cellularity, vascularity, proliferation, and incidence of metastases. In the androgen-dependent model, depletion of GAD65 prevented the transition to CRPC. These preclinical data indicate that GAD65/GABA-targeted interventions could be effective when given concurrently with androgen deprivation therapy to prevent or delay the onset of CRPC, or after CRPC ensues. Of note, the PDX models were indispensable in identifying GABA as a target and the therapeutic window of GAD65 interventions, reinforcing the utility of this important experimental model in cancer research, beyond immortalized cells lines.

An unaddressed consideration is the identification of patients with increased susceptibility to GAD65/GABA therapies. Studies in metastatic prostate cancers showed that all tumors have elevated phosphorylation of GAD65 in S6 (i.e., the activated form) compared with androgen-dependent tumors, although to varying degrees. GAD65 activity was also increased in PDXs during subsequent passaging in castrated mice, in clinical cohorts of patients treated with neoadjuvant hormone therapy, and in CRPC cases. No direct biomarkers or oncogenic drivers have been linked to the upregulation of GAD65, which could help to select or stratify patients in the future.

Another key consideration for the clinical development of this metabolic target is the likelihood of detrimental off-target effects, most notably altered neurotransmission in the central nervous system and impaired β-cell function in the pancreas. As with most metabolic targets, approaches to restrict the intervention to the tumor itself are most likely required to make this a viable approach in cancer treatment.

In summary, these advances in our understanding of how cancer metabolism is regulated in prostate cancer cells as they survive androgen deprivation have identified a possible new molecular target for CRPC. A dual role for the GAD65, including direct regulation of nuclear receptor signaling, as well as altered energy metabolism, highlights the need to better understand metabolic flux and oncometabolite signaling in cancer, and how it can be exploited for therapeutic targeting.

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were disclosed.

Received July 15, 2019; accepted July 19, 2019; published first September 13, 2019.

References
Unsuspected Protumorigenic Signaling Role for the Oncometabolite GABA in Advanced Prostate Cancer

Renea A. Taylor and Matthew J. Watt


Updated version  Access the most recent version of this article at: http://cancerres.aacrjournals.org/content/79/18/4580

Cited articles  This article cites 8 articles, 2 of which you can access for free at: http://cancerres.aacrjournals.org/content/79/18/4580.full#ref-list-1

E-mail alerts  Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions  To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions  To request permission to re-use all or part of this article, use this link http://cancerres.aacrjournals.org/content/79/18/4580.

Click on "Request Permissions" which will take you to the Copyright Clearance Center's (CCC) Rightslink site.