UDP-glucuronosyltransferase Enzymes in Prostate Cancer Progression: Is Only Androgen Catabolism Involved?

Elahe A. Mostaghel^a, Ailin Zhang^a, Stephen Plymate^{b,*}

^a Division of Clinical Research, Fred Hutchinson Cancer Research Center, Seattle, WA, USA

^b Department of Medicine, University of Washington, Seattle, WA, USA

* Corresponding author. Department of Medicine, University of Washington, Harborview Research and Training Bldg, Box 359625, 325 9th Avenue, Seattle, WA 98104, USA. Tel. +1 253 2506749.

E-mail address: splymate@u.washington.edu (S. Plymate).

Mechanisms by which androgen metabolism impact the development and progression of prostate cancer (PCa) remain a critical question. The paper by Belledant et al [1] in this issue of *European Urology* adds to the growing body of data supporting an association between UDP-glucuronosyltransferase (UGT) steroid-metabolizing enzymes and PCa progression, while simultaneously raising questions as to whether their presumed mechanism of action (steroid inactivation) is responsible for this influence.

The UGT superfamily of enzymes comprises endoplasmic reticulum-bound proteins that catalyze the glucuronidation of numerous compounds including drugs, carcinogens, and hormones to inactive polar derivatives for excretion in bile and urine [2]. UGT1 enzymes (chromosome 2) are primarily involved in conjugation of bilirubins, phenols, and estrogens [3]. The larger UGT2B enzyme family (chromosome 4) is grouped into two subfamilies with highly homologous members. *UGT2A* genes are primarily expressed in olfactory epithelium and are involved in terminating odorant signaling [4]. UGT2B enzymes (including UGT2B4, UGT2B7, UGT2B15, UGT2B17, and UGT2B28) are widely expressed in liver and extrahepatic steroid target tissues and mediate the conjugation of diverse steroid substrates (Table 1) [3,4]. Sequence divergence in the substrate-binding domains encoded by exons 1 and 2 provide substrate specificity; the remaining exons code for conserved UDP-glucuronic acid-binding and transmembrane domains [5].

Genes encoding UGT enzymes are characterized by common polymorphisms and copy number variants (CNVs) that affect gene expression and enzymatic activity and have been associated with PCa risk and outcomes. Whole gene deletions for *UGT2B17* and *UGT2B28* occur in Caucasians at 27% and 13.5%, respectively, with 57% harboring a deletion in at least one of these genes [5]. A nonsynonymous single-nucleotide polymorphism (SNP) in *UGT2B15* (rs1902923: G>T, D85Y) increases the maximal velocity for dihydrotestosterone and androstanediol glucuronidation twofold, and occurs in 32% of Caucasians [6]. *UGT2B17* and *UGT2B28* CNVs are associated with lower levels of circulating androgen-glucuronides, while *UGT2B17* CNVs are also associated with variations in levels of intraprostatic and urinary androgen-glucuronides [2]. *UGT2B17* and *UGT2B8* gene deletions have been linked to a higher risk of biochemical PCa relapse, while deletion of *UGT2B17* and *UGT2B15* D85Y has been linked to higher PCa risk in some (but not all) studies [2].

UGT2B15 and UGT2B17 alterations in PCa development include lower UGT2B15 expression in castration-resistant PCa (CRPC) metastases in comparison to untreated PCa, but higher UGT2B17 expression [2]. Notably, these enzymes are repressed by androgen signaling, and a transient increase in their expression has been noted in prostate samples subjected to neoadjuvant hormone suppression [2]. Significantly, while the androgen receptor

(AR) is required for UGT2B17 expression, further transactivation of wild-type AR by ligands suppresses UGT2B17 expression [2]. The lower UGT2B15 expression in CRPC tumors is consistent with the AR gain of function that occurs in CRPC, whereas higher UGT2B17 expression is consistent with identification of UGT2B17 as a positively regulated gene target of the constitutively active AR splice variants present in many CRPC tumors [2].

The most straightforward hypothesis regarding the impact of UGT enzymes on PCa is that lower androgen exposure due to higher glucuronidation would be associated with a lower risk of PCa development and progression. Of interest, the high-activity UGT2B15 D85Y SNP (which would decrease exposure to active androgens) is less prevalent among Asians than Caucasians (18% vs 32%), whereas UGT2B17 CNVs (which would increase androgen exposure) are more prevalent among Asians than Caucasians (73% vs 27%), suggesting that decreases in androgen exposure due to glucuronidating enzymes are not overt mediators of the lower PCa risk observed in Asian populations [6,7].

The findings by Belledant et al [1] are not in complete accordance with the hypothesis that increases in androgen catabolism and the resulting decreases in local concentrations of potent androgens favorably impact PCa behavior. While lower local levels of androgens due to higher prostate UGT2B28 levels could result in lower PSA, smaller tumor volume, and fewer positive margins, the association of higher prostate UGT2B28 with higher Gleason score, positive nodal status, and higher risk of recurrence suggests that these lower androgen levels also lead to the emergence of more aggressive disease. These data stand in contrast to the prior observation by the authors' group that patients with *UGT2B28* deletion had a twofold higher risk of biochemical PCa relapse [7]. However, the literature does suggest an association between low androgen levels and aggressive disease [8], and as the authors point out, a nonlinear U-shaped association between serum steroid levels and high-risk PCa has been observed [9].

However, these data also raise the possibility that UGT enzymes have a role in CRPC progression separate from their role in androgen inactivation. Specifically, altered UGT enzyme activity could modify the activity of steroidal or nonsteroidal metabolites with currently unrecognized pro- or anticarcinogenic effects. Knockdown of UGT2B17 in an endometrial carcinoma cell line resulted in lower cell viability due to an increase in apoptosis, in association with downregulation of the antiapoptotic protein Mcl-1 and upregulation of the proapoptotic target of Mcl-1, Puma [10]. Importantly, polymorphisms in *UGT2B15* and *UGT2B17* are quite common and have been associated with altered androgen levels and PCa risk, but were not characterized in this study, so the findings may also have been influenced by the compound effects of simultaneous polymorphisms in multiple UGT genes [11].

In conclusion, the current paper adds to evidence of an association between UGTs and PCa progression. It also emphasizes new avenues of investigation, including whether enzymatic functions vary with cytoplasmic or nuclear subcellular localization and if UGTs have activity independent of their catalytic actions, as suggested by recent findings in endometrial cancer. Importantly, a better understanding of the regulation of UGT expression is required to determine whether the UGT changes observed in PCa are an effect of altered AR signaling, or the steroid-metabolizing activities of UGTs are actual drivers of disease. Finally, although it is tempting to focus on the glucuronidating effects of UGTs on androgens and estrogens in PCa, these enzymes may well affect nonsteroidal pathways that impact PCa development and progression.

Conflicts of interest: The authors have nothing to disclose.

Acknowledgments: The authors acknowledge support from Pacific Northwest Prostate Cancer SPOR P50 (CA97186, E.A.M. and S.P.) and US Department of Defense CDMRP (W81XWH-12-1-0208, E.A.M.; W81XWH-13-2-0093 and PO1CA163227, S.P.).

References

1. Belledant A, Hovington H, Garcia L, et al. The UGT2B28 sex-steroid inactivation pathway is a regulator of steroidogenesis and modifies the risk of prostate cancer progression. *Eur Urol*. In press. <http://dx.doi.org/10.1016/j.eururo.2015.06.054>
2. Gauthier-Landry L, Belanger A, Barbier O. Multiple roles for UDP-glucuronosyltransferase (UGT)2B15 and UGT2B17 enzymes in androgen metabolism and prostate cancer evolution. *J Steroid Biochem Mol Biol* 2015;145:187–92.
3. Turgeon D, Carrier JS, Lévesque E, Hum DW, Bélanger A. Relative enzymatic activity, protein stability, and tissue distribution of human steroid-metabolizing UGT2B subfamily members. *Endocrinology* 2001;142:778–87.
4. Levesque E, Turgeon D, Carrier JS, Montminy V, Beaulieu M, Bélanger A. Isolation and characterization of the UGT2B28 cDNA encoding a novel human steroid conjugating UDP-glucuronosyltransferase. *Biochemistry* 2001;40:3869–81.
5. Guillemette C, Lévesque E, Harvey M, Bellemare J, Menard V. UGT genomic diversity: beyond gene duplication. *Drug Metab Rev* 2010;42:24–44.
6. Lampe JW, Bigler J, Bush AC, Potter JD. Prevalence of polymorphisms in the human UDP-glucuronosyltransferase 2B family: UGT2B4(D458E), UGT2B7(H268Y), and UGT2B15(D85Y). *Cancer Epidemiol Biomarkers Prev* 2000;9:329–33.
7. Nadeau G, Bellemare J, Audet-Walsh E, et al. Deletions of the androgen-metabolizing UGT2B genes have an effect on circulating steroid levels and biochemical recurrence after radical prostatectomy in localized prostate cancer. *J Clin Endocrinol Metab* 2011;96:E1550–7.
8. Nishiyama T, Ikarashi T, Hashimoto Y, Suzuki K, Takahashi K. Association between the dihydrotestosterone level in the prostate and prostate cancer aggressiveness using the Gleason score. *J Urol* 2006;176:1387–91.
9. Salonia A, Abdollah F, Capitanio U, et al. Serum sex steroids depict a nonlinear U-shaped association with high-risk prostate cancer at radical prostatectomy. *Clin Cancer Res* 2012;18:3648–57.
10. Hirata H, Hinoda Y, Zaman MS, et al. Function of UDP-glucuronosyltransferase 2B17 (UGT2B17) is involved in endometrial cancer. *Carcinogenesis* 2010;31:1620–6.
11. Menard V, Eap O, Harvey M, Guillemette C, Lévesque E. Copy-number variations (CNVs) of the human sex steroid metabolizing genes UGT2B17 and UGT2B28 and their associations with a UGT2B15 functional polymorphism. *Hum Mutat* 2009;30:1310–9.

Table 1 – Steroid conjugation by UGT2B enzymes

UGT2B enzyme ^a	Conjugation			
	Testosterone	Dihydrotestosterone	Androstenediol	Androsterone
UGT2B4	No	No	Yes	Yes
UGT2B7	Yes	No	Yes	Yes
UGT2B15	Yes	Yes	Yes	No
UGT2B17	Primary	Primary	Yes	Primary
UGT2B28	Yes	No	Yes	Yes

^a All enzymes except UGT2B7 are expressed in the prostate.