Long noncoding RNA-mediated activation of androgen receptor in prostate cancer

Gyorgy Petrovics, Shiv Srivastava

Asian Journal of Andrology (2014) 16, 418–419; doi: 10.4103/1008-682X.126398; published online: 28 March 2014

Remarkable progress has been made in molecular characterization of prostate cancer (PCa) with continued innovations in high throughput technologies evaluating human cancer. Since the completion of the Human Genome Project it has been estimated that only about 1.5%–2% of our genome codes for proteins. Various genome-wide approaches, e.g. the ENCODE project, revealed that a much larger percent of the genome is transcribed as non-protein coding (nc) RNA, including long noncoding (lnc) RNA (over 200bps long). Although the biological roles of lncRNA (the ‘dark matter of the genome’) are not nearly as well-understood as the protein coding mRNAs, it is increasingly clear that they play important roles in almost every aspects of biology, including cancer biology. This is exemplified by recent genome-wide association studies revealing that over 80% of cancer-associated single nucleotide polymorphisms (SNPs) are in noncoding regions of the genome.

Lnc RNAs can function through a variety of mechanisms, including chromatin remodeling (Xist, Hotair, Anrils), transcriptional co-activation/repression (H19, lncRNA-p21, SRA), posttranscriptional modifications (MALAT1), protein inhibition (TERRA) and decay elements (PTENP1). SRA has been identified as a steroid receptor coactivator lncRNA. However, the function and mechanism of most lnc RNAs remain unclear.

Surprisingly enough, early discoveries using differential display technologies described two lncRNAs, DD3/PCA3 and PCGEM1, which not only exhibited high prostate tissue specificity but also showed prostate tumor associated overexpression. Recent evaluations of PCa transcriptome have described several lncRNAs, including PCAT-1, a transcriptional repressor and target of Polycomb Repressive Complex 2, implicated in PCa progression and PRNCR1, a PCa susceptibility associated lncRNA. Interestingly, both PCAT-1 and PRNCR1 reside in the 8q24 PCa susceptibility locus, with some androgen receptor (AR) targets without affecting AR expression levels. Significantly, the truncated AR-V7 (75 kDa) splice variant, which can activate AR regulated gene expression (about 600 genes). Similarly, the recent discovery of PCGEM1, a prostate specific androgen regulated gene, provides a new target for development of novel therapeutics.

Hormonal induction of these associations was revealed by DHT treatment of LNCaP cells. Of note, previous studies have shown that PCGEM1 itself can be induced by androgen, which may further cooperate with AR activation especially when it is overexpressed in PCa. Antisense oligonucleotide based knockdown of PRNCR1 abolished both its own interaction with AR, and the association of PCGEM1 with AR. However, antisense oligonucleotide targeting of PCGEM1 abolished only the PCGEM1-AR association, suggesting for a PRNCR1 dependent recruitment of PCGEM1 to AR. In vitro binding studies mapped the PRNCR1 binding site to AR 549–623 region, and the PCGEM1 binding site to the N-terminal region of AR. The lncRNA-bound AR had specific posttranslational modifications: acetylation was required for association with PRNCR1 and methylation for the PCGEM1 binding. These promising novel observations will lead to further refinement of these complex interactions.

Chromatin isolation by RNA purification (ChIRP) revealed over 2000 PCGEM1 occupancy sites in the genome, about 80% of them colocalize with AR-bound sites. Global run-on sequencing (GRO-seq) revealed that knockdown of either lncRNAs by antisense oligonucleotide decreased AR target gene expression (about 600 genes). Similarly, shRNA against either PCGEM1 or PRNCR1 reduced the DHT-induced activation of AR targets without affecting AR expression levels. Significantly, the truncated AR-V7 (75kDa) splice variant, which can activate AR-regulated gene expression (about 600 genes). Similarly, the recent discovery of PCGEM1, a prostate specific androgen regulated gene, provides a new target for development of novel therapeutics.

Finally, the biological roles of these lncRNAs were investigated in stable cell lines...
of CWR22Rv1 harboring dox-induced shRNA against PCGEM1 or PRNCR1. In addition to reduced expression of canonical AR target genes a reduction of cell growth and inhibition of tumor growth in a xenograft model (CRPC) was demonstrated.

Taken together, novel findings from this report, along with previous studies of PCGEM1, AR and PRNCR1, reveal biological importance of prostate associated lncRNAs (PCGEM1 and PRNCR1) in full length and truncated AR-dependent gene activation in PCa (Figure 1). As PCGEM1 and PRNCR1 strongly enhance AR activity in PCa, they may be explored as potential new therapeutic targets in CRPC.

REFERENCES

1 Hieronymus H, Sawyers CL. Traversing the genomic landscape of prostate cancer from diagnosis to death. Nat Genet 2012; 44: 613–4.
2 Beltran H, Rubin MA. New strategies in prostate cancer: translating genomics into the clinic. Clin Cancer Res 2013; 19: 517–23.
3 Dobi A, Sreenath T, Srivastava S. Androgen dependent oncogenic activation of ETS transcription factors by recurrent gene fusions in prostate cancer: biological and clinical implications. In: Wang Z, editor. Androgen-responsive genes in prostate cancer. Springer: New York; 2013: p307–28.
4 Nagano T, Fraser P. No-nonsense functions for long noncoding RNAs. Cell 2011; 145: 178–81.
5 Cheetham SW, Gruhl F, Mattick JS, Dinger ME. Long noncoding RNAs and the genetics of cancer. Br J Cancer 2013; 108: 2419-25.
6 Lanz RB, McKenna NJ, Onate SA, Albrecht U, Wong J, et al. A steroid receptor coactivator, SRA, functions as an RNA and is present in an SRC-1 complex. Cell 1999; 97: 17-27.
7 Bussemakers MJ, van Bokhoven A, Verhaegh GW, Smit FP, Karthaus HF, et al. DD3: a new prostate-specific gene, highly overexpressed in prostate cancer. Cancer Res 1999; 59: 5975–9.
8 Srikantan V, Zou Z, Petrovics G, Xu L, Augustus M, et al. PCGEM1, a prostate-specific gene, is overexpressed in prostate cancer. Proc Natl Acad Sci U S A 2000; 97: 12216–21.
9 Prensner JR, Iyer MK, Balbín OA, Dhanasekaran SM, Cao Q, et al. Transcriptome sequencing across a prostate cancer cohort identifies PCAF-1 an unannotated lncRNA implicated in disease progression. Nat Biotechnol 2011; 29: 742–9.
10 Chung S, Nakagawa H, Uemura M, Piao L, Ashikawa K, et al. Association of a novel long non-coding RNA in Bq24 with prostate cancer susceptibility. Cancer Sci 2011; 102: 245–52.
11 Day JR, Jost M, Reynolds MA, Groskopf J, Rittenhouse H. PCA3: from basic molecular science to the clinical lab. Cancer Lett 2011; 301: 1–6.
12 Crawford ED, Rege KD, Trabulsi EJ, Qian J, Dregnovska KP, et al. Diagnostic performance of PCA3 to detect prostate cancer in men with increased prostate specific antigen: a prospective study of 1,962 cases. J Urol 2012; 188: 1726–31.
13 Yang L, Lin C, Jin C, Yang JC, Tanasa B, et al. lncRNA-dependent mechanisms of androgen-receptor-regulated gene activation programs. Nature 2013; 500: 598–602.
14 Petrovics G, Zhang W, Makarem M, Street JP, Connelly R, et al. Elevated expression of PCGEM1, a prostate-specific gene with cell growth-promoting function, is associated with high-risk prostate cancer patients. Oncogene 2004; 23: 605–11.
15 Xu X, Ravindranath L, Tran N, Petrovics G, Srivastava S. Regulation of apoptosis by a prostate-specific and prostate cancer-associated noncoding gene, PCGEM1. DNA Cell Biol 2006; 25: 135–41.

How to cite this article: Petrovics G, Srivastava S. Long non-coding RNA mediated activation of androgen receptor in prostate cancer. Asian J Androl 28 March 2014. doi: 10.4103/1008-682X.126398. [Epub ahead of print]