Alteration in Androgen Receptor Expression by the 3' Untranslated Region

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Background. Prostate cancer is the 2nd most common cancer in men and affects nearly 240,000 males annually in the United States with about 29,700 deaths each year. Androgen blocking is the mainstay of therapy but is associated with significant adverse effects. Therefore, understanding the molecular mechanisms of regulation of androgen receptor (AR) will permit the development of novel, targeted therapies but the mechanisms of AR regulation are poorly understood. AR mRNA contains a very long 3' untranslated region (UTR) of 6.kbp and it is a target for many miRNAs and RNA binding proteins. Aim. To understand the mechanism of regulation of AR gene expression through its 3'UTR. Hypothesis. It was hypothesized that the 3'UTR in the AR gene transcriptionally regulates the expression of AR. Methods. All studies were performed in PC-3 cells grown to differentiation. Amplification of constructs was done using PCR, protein assays using immunoblots, and cloning using appropriate restriction enzymes and transfection using a lipofectamine protocol. Results. Transcriptional regulation of AR was examined by expression of a plasmid containing AR gene with and without the 3'UTR into PC-3 cells. AR 3' UTR constructs were amplified, sequence confirmed and cloned between SpeI and SacI sites in pMIR-REPORT luciferase vector using the cloning protocol. The expression of the AR protein at different conditions was quantified by Immunoblotting assays. Transfection of the 3'UTR significantly decreases AR gene expression. This confirms that 3'UTR poses a regulatory effect on AR gene. Conclusion. An in vitro model for determining the mechanisms of AR regulation in prostate cancer was established. Full length 3'UTR is necessary for transcriptional regulation of AR.