New Therapy in Prostate Cancer

JMJD2C controls Prostate Tumour Growth

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Technology

The Jumonji C (JMJC) domain-containing protein JMJD2C is the first histone tridemethylase regulating androgen receptor function. Knockdown of JMJD2C inhibits androgen-induced removal of trimethyl H3K9, transcriptional activation and tumor cell proliferation. Importantly, JMJD2C colocalizes with androgen receptor and LSD1 in normal prostate and in prostate carcinomas. JMJD2C and LSD1 interact and both demethylases cooperatively stimulate androgen receptor-dependent gene transcription. In addition, androgen receptor, JMJD2C and LSD1 assemble on chromatin to remove methyl groups from mono, di and trimethylated H3K9. Thus, our data suggest that specific gene regulation requires the assembly and coordinate action of demethylases with distinct substrate specificities. Lysine specific Demethylase 1 (LSD1) co-localises with AR in normal human prostate and in prostate tumor. LSD1 interacts with AR and stimulates AR-dependent transcription. We identify pargyline as an inhibitor of LSD1 that blocks AR-dependent transcription. Furthermore LSD1 knockdown by RNAI abrogates androgen induced transcriptional activation and cell proliferation.

Innovation

- better treatment of prostate tumor

Application

Modulation of JMJD2C activity is tissues where AR has a pivotal physiological role i.e.:
- Treatment of prostate tumor
- Control of fertility
- Treatment of Alzheimer’s disease
- Treatment of Parkinson’s disease

Market Potential

Prostate cancer represents the most frequent malignant disease in men worldwide and the second leading cause of death from malignant tumors.

Proof of Concept

Please contact us for further information.

Patent Status

- European Patent Application Pending
- Filed (PRD): January 26th, 2007
- International Patent Application (PCT)
Co-operative demethylation by JMJD2C and LSD1 promotes androgen receptor-dependent gene expression

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Abstract

Posttranslational modifications of histones such as methylation regulate chromatin structure and gene expression. Methylation of histones had been considered to be permanent until recently when lysine-specific demethylase 1 (LSD1), the first histone demethylase, was discovered. LSD1 interacts with the androgen receptor (AR) and promotes androgen-dependent transcription of target genes by ligand-induced demethylation of mono- and dimethylated histone H3 at lysine 9 (H3K9). In contrast, trimethylated histone H3 is not demethylated by LSD1 suggesting that androgen-dependent demethylation of trimethyl H3K9 is controlled by as yet unknown histone demethylases. Here we identify the Jumonji C (JmJC) domain-containing protein JMJD2C as the first histone demethylase regulating AR function. JMJD2C interacts with AR and promotes AR-mediated transcription of target genes in vivo and in stimulation of AR-dependent transcription. Conversely, knockdown of JMJD2C in prostate cancer cells inhibits androgen-induced removal of trimethyl H3K9, transcriptional activation, and tumor cell proliferation. Importantly, JMJD2C not only co-localizes with AR but also with LSD1 in normal prostate and prostate carcinoma. In addition, AR, JMJD2C, and LSD1 assemble on chromatin to remove methyl groups from mono-, di-, and trimethylated H3K9 and both demethylases co-operatively stimulate AR-dependent transcription. Furthermore, ligand-dependent assembly of AR with JMJD2C and LSD1 on AR target genes results in demethylation of trimethyl H3K9. LSD1 promotes androgen-dependent transcription, and tumor cell proliferation. In contrast, knockdown of LSD1 abolishes AR localization in the nucleus and reduces AR binding to target genes. LSD1 binds to AR in a histone H3 (H3K9) methylated state.

Conclusion:

1) JMJD2C and LSD1 co-operatively regulate AR transcriptional activity
2) JMJD2C demethylates trimethyl histone H3 at Lysine 9 and thereby generates the substrate for LSD1
3) JMJD2C controls androgen-dependent proliferation of prostate cancer cells
4) JMJD2C is a potential target to block prostate tumour growth

Figure 1: JMJD2C co-localizes with and interacts with both, AR and LSD1.

Figure 2: JMJD2C interacts with chromatin and demethylates H3K9. LNCaP cells were incubated with or without the AR agonist R1881 (a, b), and transfected with stealth RNAi (b). CHP or Re-CHP was performed with the indicated antibodies. The precipitated chromatin was amplified by PCR using primers flanking the promoter region (ARE I+II) and an internal control (rIgG). Western bands were detected with a-AR, a-JMJD2C, or a-LSD1 antibodies as indicated. a, c, d, GST pull-down assays were performed with labeled JMJD2C and the bacterially expressed GST-AR or GST-LSD1 fusion proteins. GST, and GST-LSD1 proteins were used as control. (R1881- induced expression (%)

Figure 3: JMJD2C knockdown blocks AR-induced transcriptional activity. CV1 (a, b, d), or LNCaP (c) cells were transfected with AR-dependent reporters in the presence or absence of R1881. CV1 cells were co-transfected with AR expression plasmid (a, b); JMJD2C but not the other JMJD2 family members JMJD2A, JMJD2B, or JMJD2C (b) controls AR-induced transcriptional activity on different natural AR-regulated promoters and cell lines. Limited amounts of JMJD2C, JMJD2C R196A, or LSD1 (b) were tested for co-operative stimulation of AR-dependent reporter activity (d). Bars represent mean ±SD (n=5).

Figure 4: JMJD2C knockdown blocks AR-induced transcriptional activity and tumor cell proliferation. In LNCaP cells, miRNA-mediated JMJD2C knockdown reduces expression of the endogenous PSA gene (a, left panel), AR-dependent reporter activity (b), and R1881-induced cell proliferation (c). Knockdown of JMJD2C was verified by Western blot analysis (a, right panel) using a-JMJD2C or a-AR antibodies. Bars represent mean ±SD (n=4).