GATA3 REGULATES LIPID CONTENT AND COOPERATES WITH AURORA B KINASE IN THE MITOTIC SPINDLE OF HORMONE-REFRACTORY PROSTATE CANCER CELLS

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Background
GATA3 is an important determinant for cell lineage in prostate development and loss of this transcription factor has been implicated in more aggressive prostate tumor growth. The tumor inhibiting mechanisms of GATA3 remain unclear. A critical protein in mitosis, Aurora B kinase, is upregulated in prostate cancer and its targeted inhibitor can suppress tumor growth. We postulated that GATA3 in prostate cancer directly regulates cell growth via cyclin D1 and controls mitotic spindle assembly by cooperating with aurora B kinase.

Methods
To assess the mitotic spindle, immunofluorescence (IF) stains were performed for DAPI, Aurora B kinase and GATA3. Prostate cancer cell line, C4-2b, was transfected with GATA3 siRNA and cell cycle proteins analyzed. GATA3 heterozygote murine embryos were phenotyped and immunostained for Ki67, adipose triglyceride lipase (ATGL), perilipin 2, pigment epithelium-derived factor (PEDF) and angiogenic factors (VEGF and PEDF). To simulate a lipid-enriched tumor microenvironment, C42b cells were treated with oleic acid (OA) and stained for Oil-Red-O and GATA3.

Results
IF studies revealed that in the early phase of mitosis, GATA3 was dispersed in the nucleus. As mitosis progressed GATA3 condensed and aligned with chromatids. GATA3 co-localized with Aurora B kinase at opposing ends of each daughter nucleus until they were cleaved. In the lipid-rich OA-treated experimental group demonstrated malalignment of the mitotic spindle orientation and mitotic progression. Reducing GATA3 in prostate cancer cells showed a 38% increase in cell cycle regulator, cyclin D1, elevated intracytoplasmic lipid and more than a twofold increase in cell number. $GATA3^{+/-}$ embryos demonstrated an increased number of proliferating cells in many organ systems associated with an increased vascular density. The liver had excessive neutral lipid accumulation and increased binucleation of the hepatocytes.

Conclusions
A role for GATA3 in mitotic spindle assembly was discovered with co-localization of an essential cell division regulator, Aurora B kinase. GATA3 knockdown in tumor cells resulted in an increase in proliferating cells and cyclin D1 and this pro-proliferative phenotype was exacerbated by a lipid stimulus. GATA3 deficiency in mice revealed hepatic steatosis supporting a new function for GATA3 in lipid metabolism. Therapeutic targeting of the lipid-GATA3 axis could be effective at arresting tumor growth and progression.