National Human Genome Research Institute

The Forefront of Genomics

FBXW7 mutations in serous endometrial cancers cause increased levels of potentially druggable proteins and *in vitro* sensitivity to SI-2 and dinaciclib

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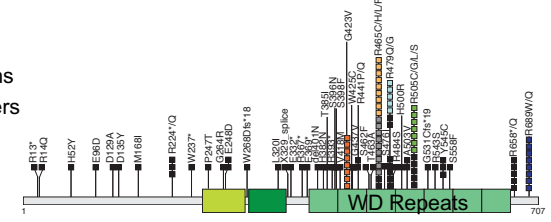
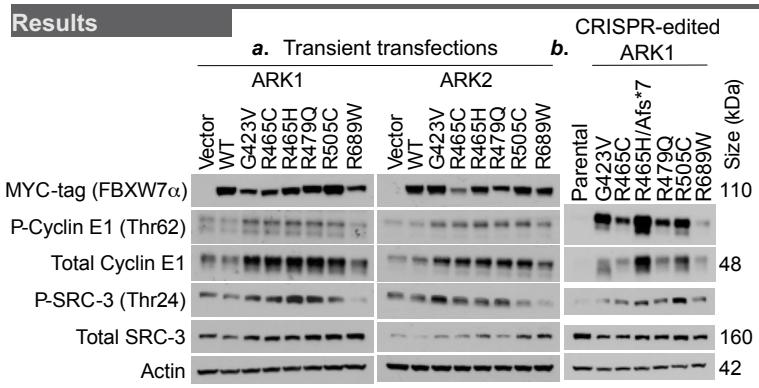
Background

- *FBXW7* is a haploinsufficient tumor suppressor gene and component of a ubiquitin ligase complex that regulates the proteasome-mediated degradation of oncogenic proteins
- Somatic *FBXW7* mutations have been reported in 17-30% of serous endometrial cancers (ECs), 11-28% of uterine carcinosarcomas, and 7-25% of clear cell ECs; recurrent mutations cluster within the substrate binding domain (WD Repeats)
- Prior to this work, the effects of *FBXW7* mutations in EC were unknown

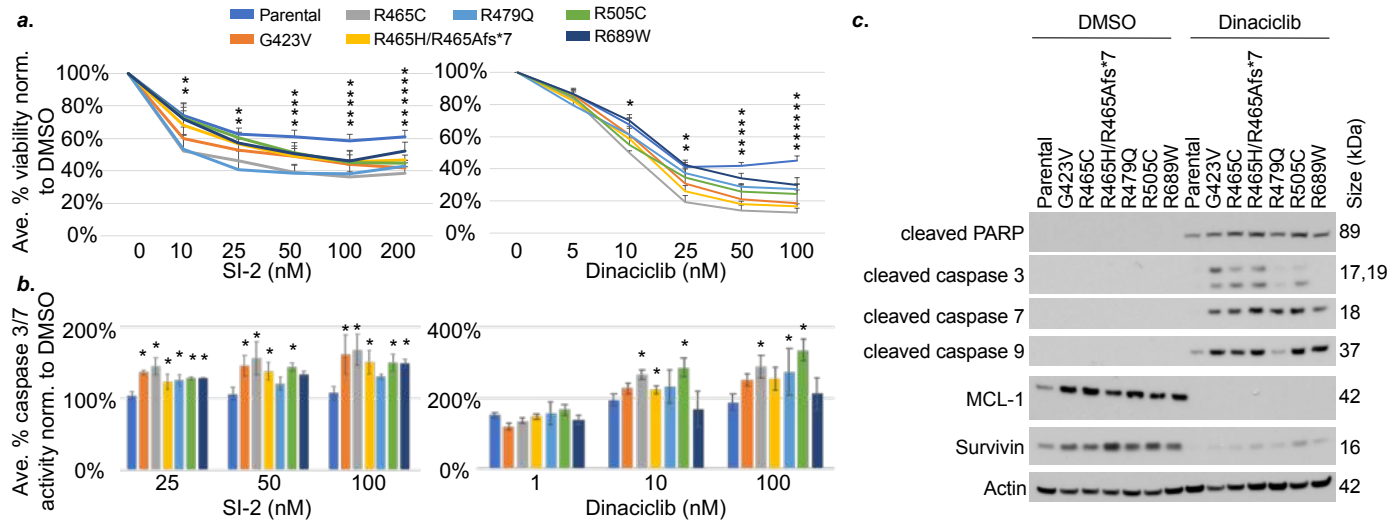


Results

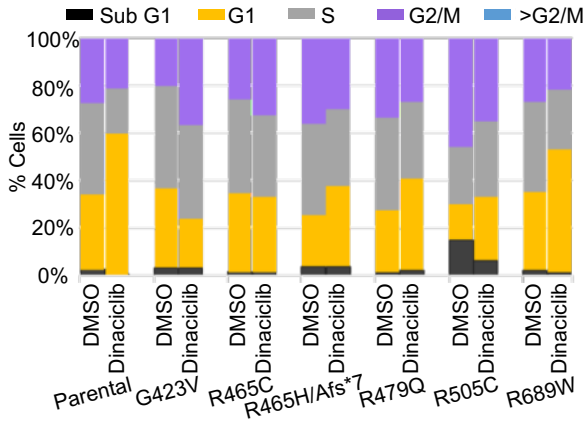


FBXW7* mutations led to increased Cyclin E1 and SRC-3 protein levels in EC cells.** Western blot results of whole cell lysates of (a.***) ARK1 and ARK2 serous EC cells collected 24hrs after transfection with 2.5µg of MYC-tagged vector control, wildtype (WT) or mutant *FBXW7α* expression constructs and (***b.***) Parental ARK1 and CRISPR-edited cells collected 24hrs after plating. Blots are representative of triplicate experiments.

Not shown: *FBXW7* mutations also led to increases in phosphorylated levels of c-MYC, Rictor, GSK3, P70S6, and AKT proteins. Urick ME and Bell DW. *Molecular Carcinogenesis* 2018; https://doi.org/10.1002/mc.22867.



Compared to parental ARK1, CRISPR-edited *FBXW7*-mutant cells exhibited decreased viability and increased apoptosis in response to SI-2 (SRC inhibitor) and dinaciclib (CDK inhibitor; CDK2 binds Cyclin E1). (***a.***) Viability assay results 24hr (SI-2) or 48hr (dinaciclib) after treatments. (***b.***) Caspase 3/7 activity measured 24hr after treatment. Error bars= standard deviation of triplicate experiments; stars indicate significance ($p<0.05$) calculated using a one-way ANOVA and Dunnett's multiple comparisons test. (***c.***) Western blot results 24hr after treatment.



Dinaciclib treatment caused changes in cell cycle that differed between parental ARK1 and CRISPR-edited *FBXW7*-mutant cells. Flow cytometry results 24hr after treatment with 100nM dinaciclib or DMSO; representative of triplicate experiments.

Conclusion

- Recurrent somatic *FBXW7* mutations caused increased phosphorylated protein levels of Cyclin E1, SRC3, c-MYC, Rictor, GSK3, P70S6, and AKT in serous endometrial cancer cells and increased sensitivity to SI-2 (a SRC inhibitor) and dinaciclib (a CDK inhibitor; CDK2 binds Cyclin E1) in ARK1 cells.
- These findings represent the first direct biochemical evidence that *FBXW7* mutations have functional relevance in serous endometrial cancer cells.

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