Massachusetts Eye and Ear

HARVARD MEDICAL SCHOOL
TEACHING HOSPITAL

Targeting SWI/SNF/Hippo pathway in Squamous Cancers

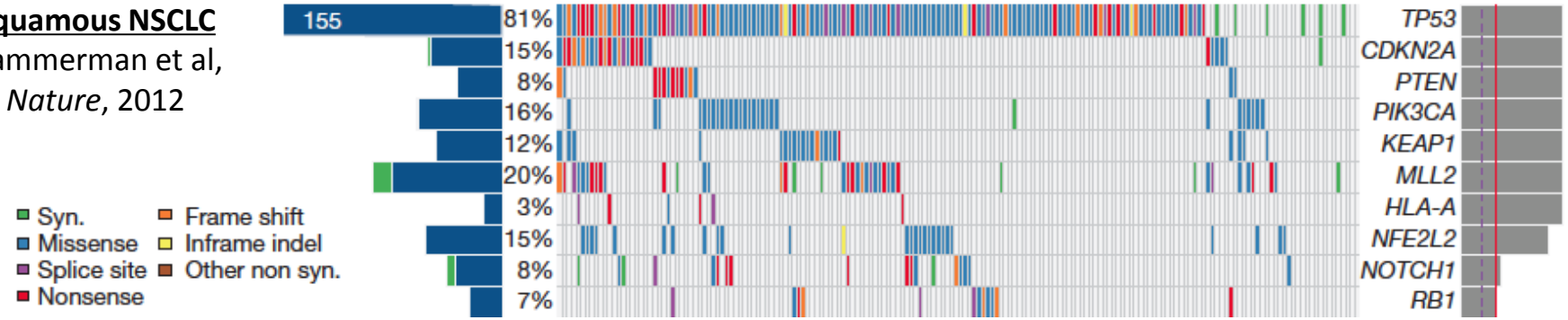
Srinivas Vinod Saladi Ph.D.

Chabner Colloquium, 2019

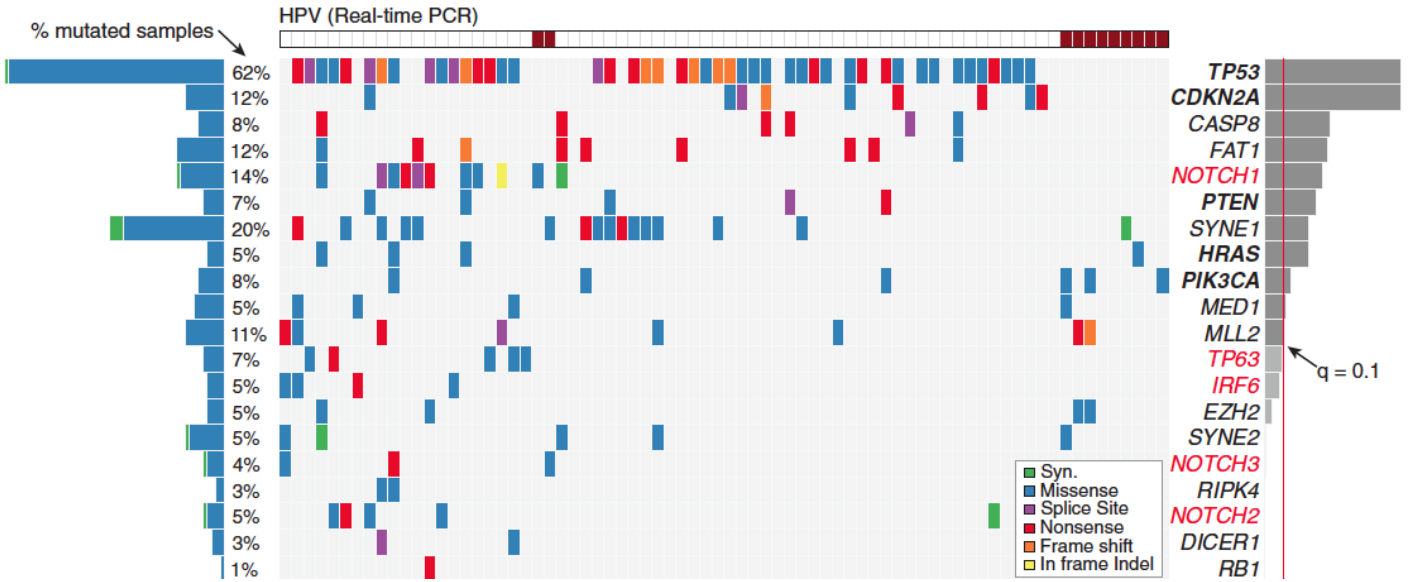
MASSACHUSETTS GENERAL HOSPITAL
CANCER CENTER

Squamous cell carcinoma genomic landscape: largely tumor suppressors

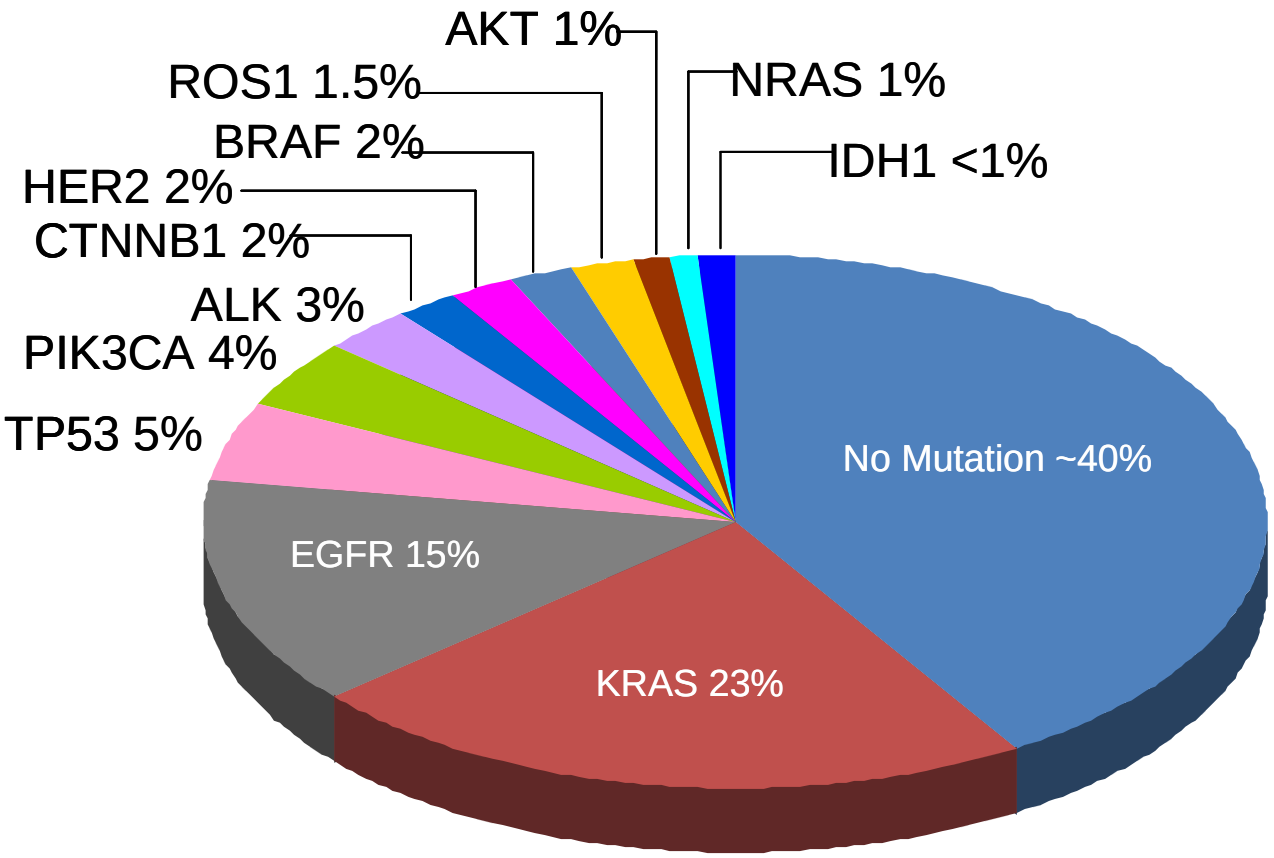
Squamous NSCLC
Hammerman et al,
Nature, 2012



Head & Neck SCC
Stransky et al,
Science, 2011



Lung adenocarcinoma genotypes: many potential therapeutic opportunities



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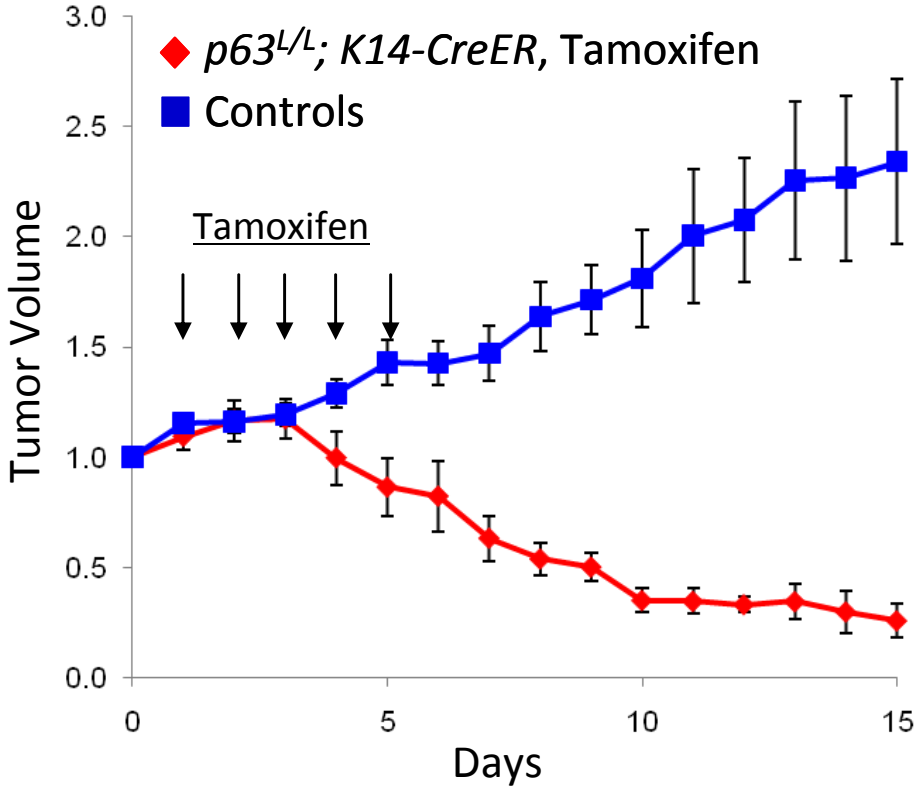
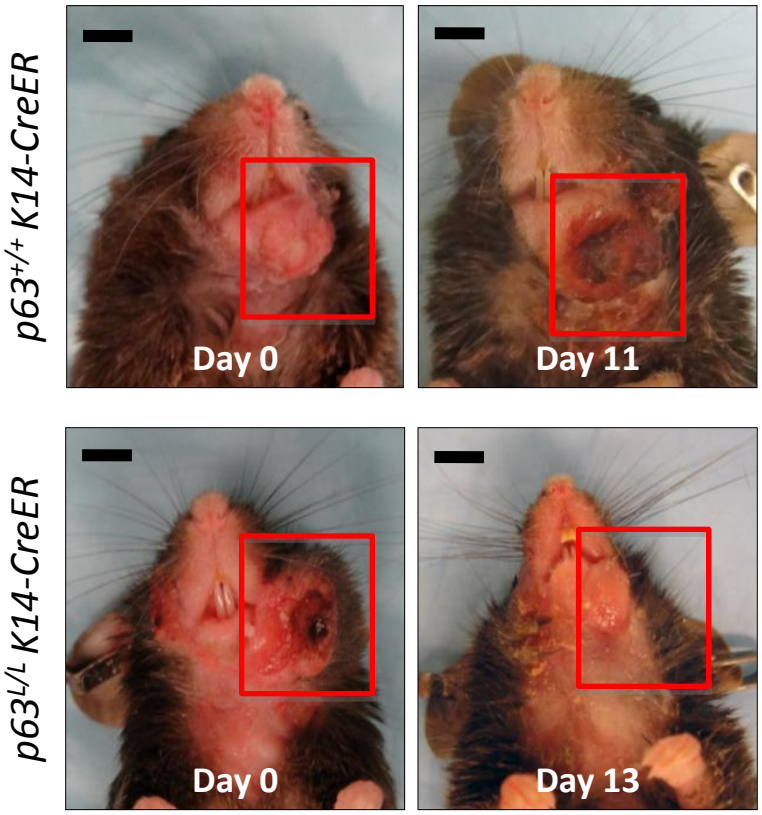
CANCER CENTER

Excision of p63 in SCC *in vivo* results in rapid tumor regression



Oral SCC

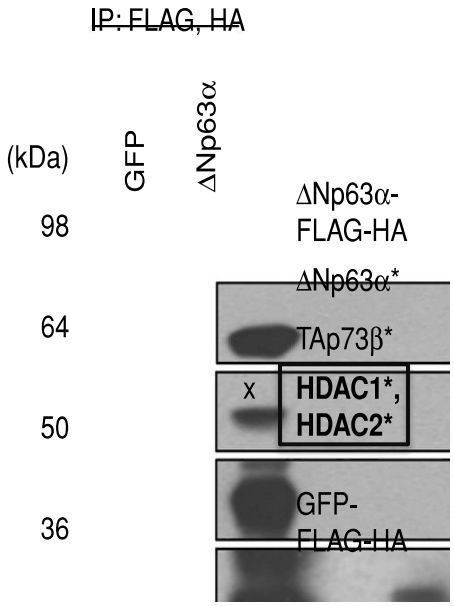


100mg/kg tamoxifen daily
days 1-5

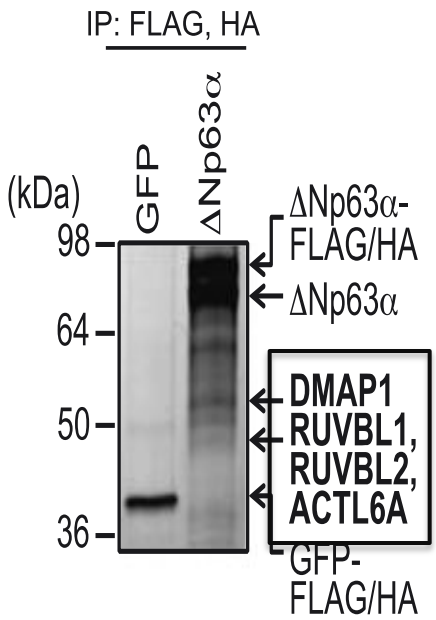
p63 regulates genes by association with distinct epigenetic cofactors/complexes

Cistrome analysis
p63 ChIP-Seq (JHU029)

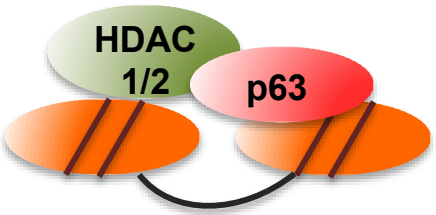
Factors
TP53
TCFP2L1
NRF1
TFCP2
SMARCC1
FOS
E2F1
BCL11A
FOSL2
SMARCA4
FOSL1
BACH2
SMARCB1
TRIM28
RUNX1



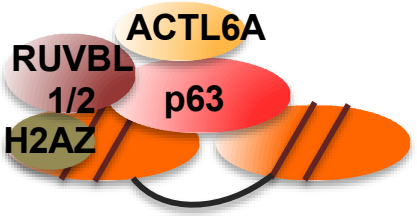
(Ramsey, MR, He L, Forster N, et al, *Cancer Research,* 2011)



(Corrie LG, Ramsey MR, et al, *Genes & Dev*, 2012)



PUMA, NOXA



SAMD9L

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p63 ChIP-Seq (JHU029)

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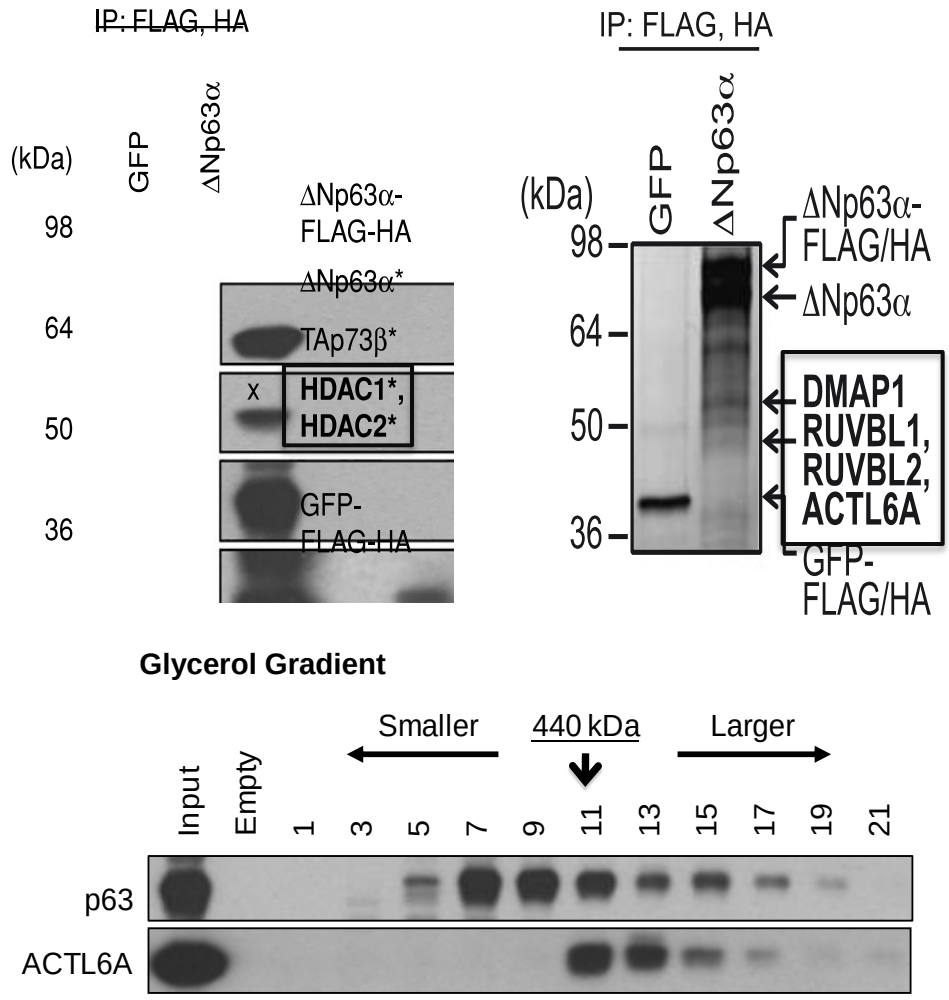
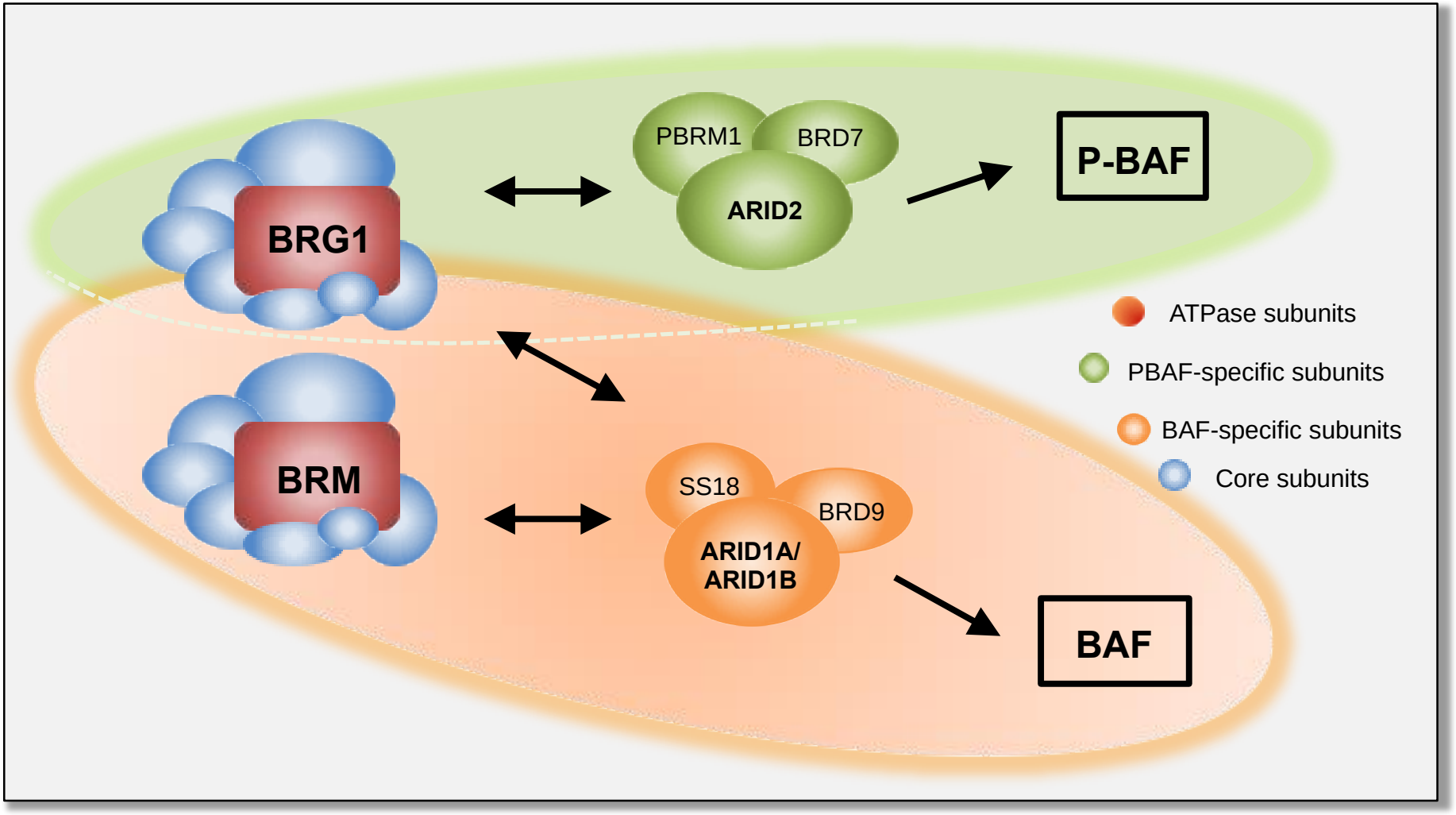
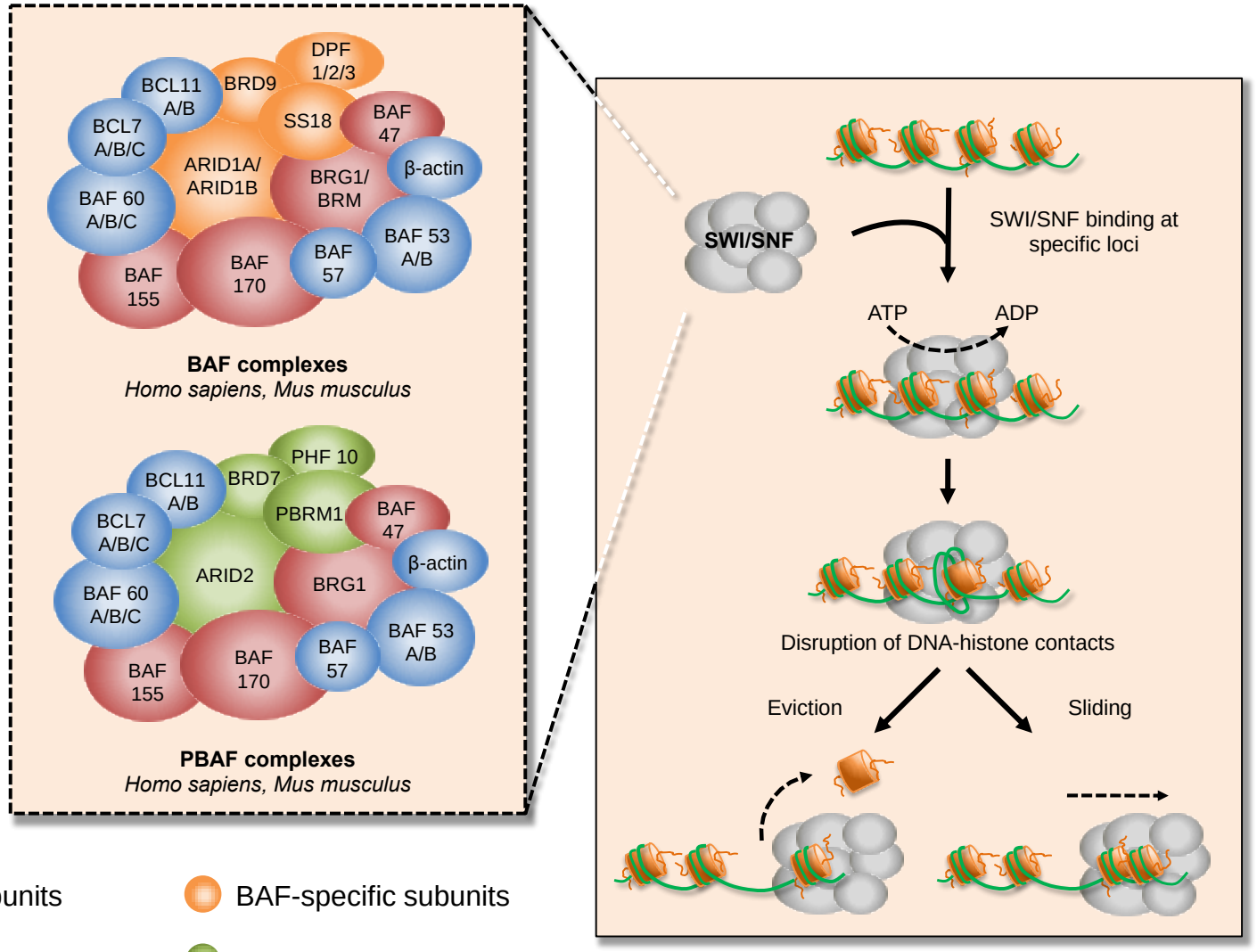


SWI/SNF-ATP – dependent chromatin remodeling enzymes are highly heterogeneous



SWI/SNF complexes control nucleosome position



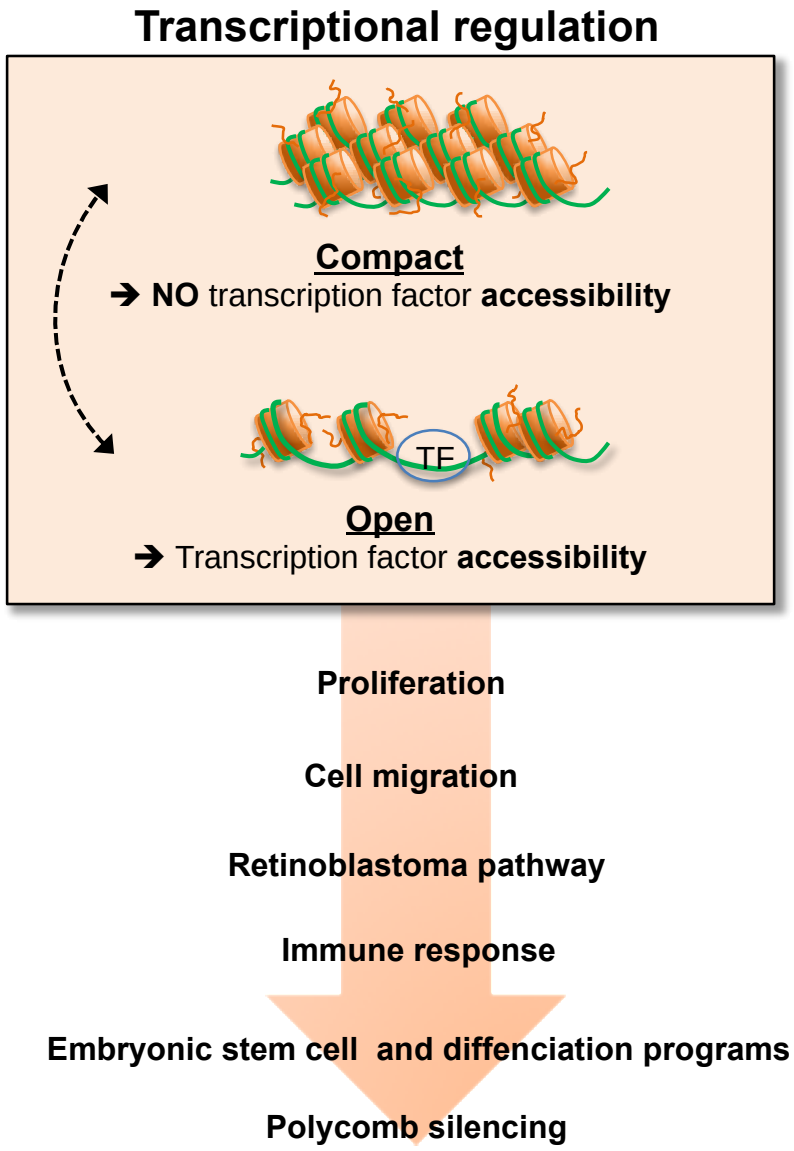
Core subunits

BAF-specific subunits

BAF & PBAF subunits

PBAF-specific subunits

SWI/SNF complex mediated Transcriptional Regulation



Chromatin (in highly condensed form)

SWI/SNF (ATP dependant)

Unwinding of Chromatin

Binding of Transcription Factors

Transcription of Genes

Functions of SWI/SNF

SWI/SNF enzymes play a key role in transcriptional regulation, replication, DNA repair, and cell cycle control.

SWI/SNF enzymes are critical for cellular differentiation and embryonic development

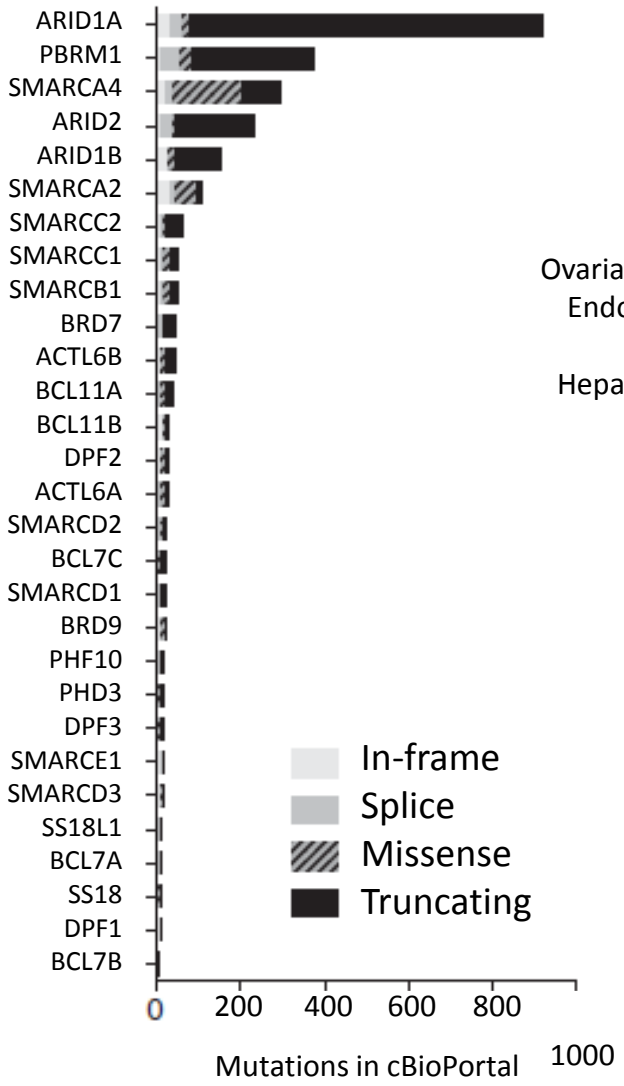
SWI/SNF subunits are mutated or deleted in many forms of cancer and one of the subunits BAF47 may act as tumor suppressor.

Gastric carcinomas with lymph node metastasis over-express BRG1 and are depleted of BRM expression.

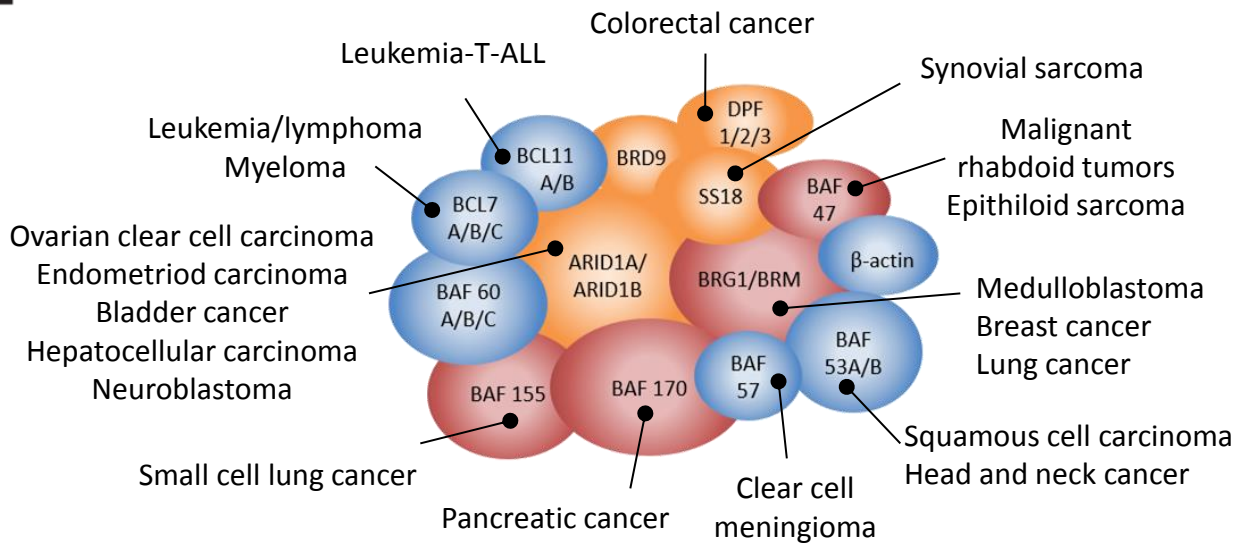
BRG1 expression is high in invasive forms of prostate cancer.

SWI/SNF complex altered in multiple cancers

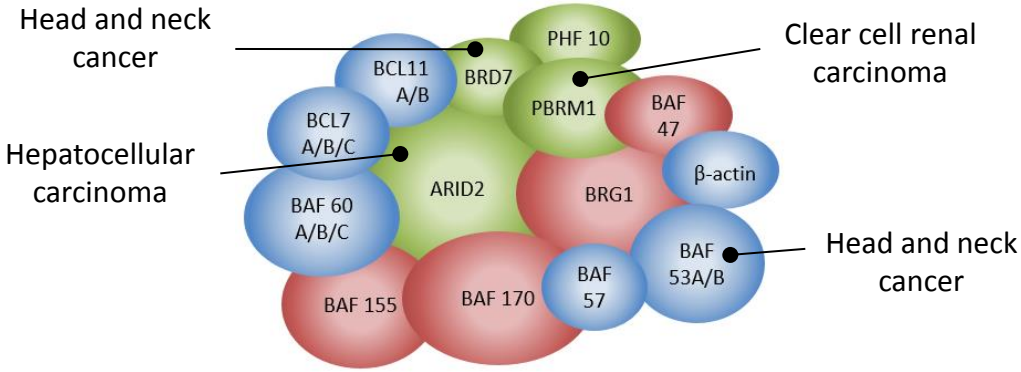
BAF/PBAF subunits



mSWI/SNF complex (BAF)

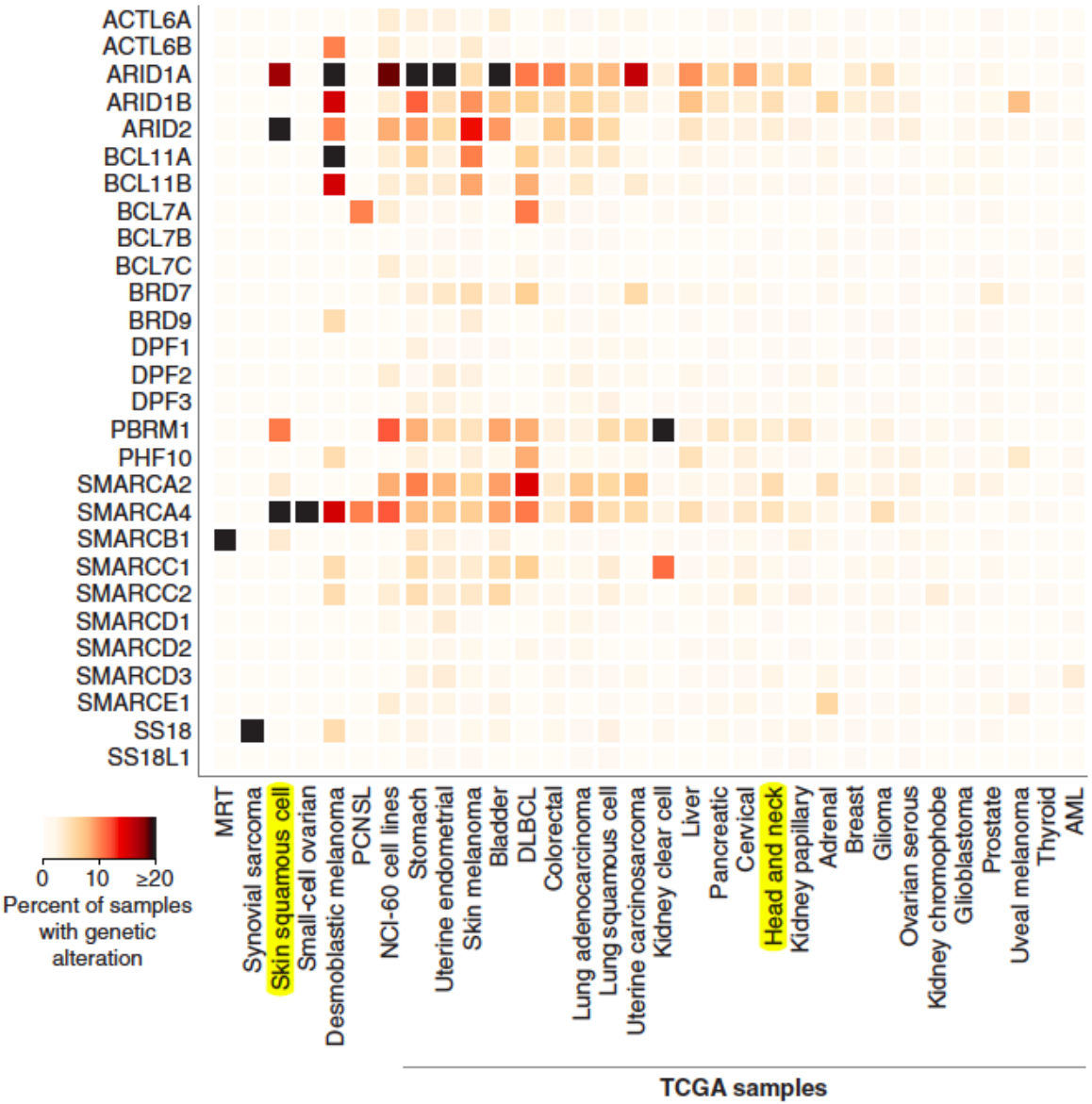


Polybromo-containing BAF (PBAF)



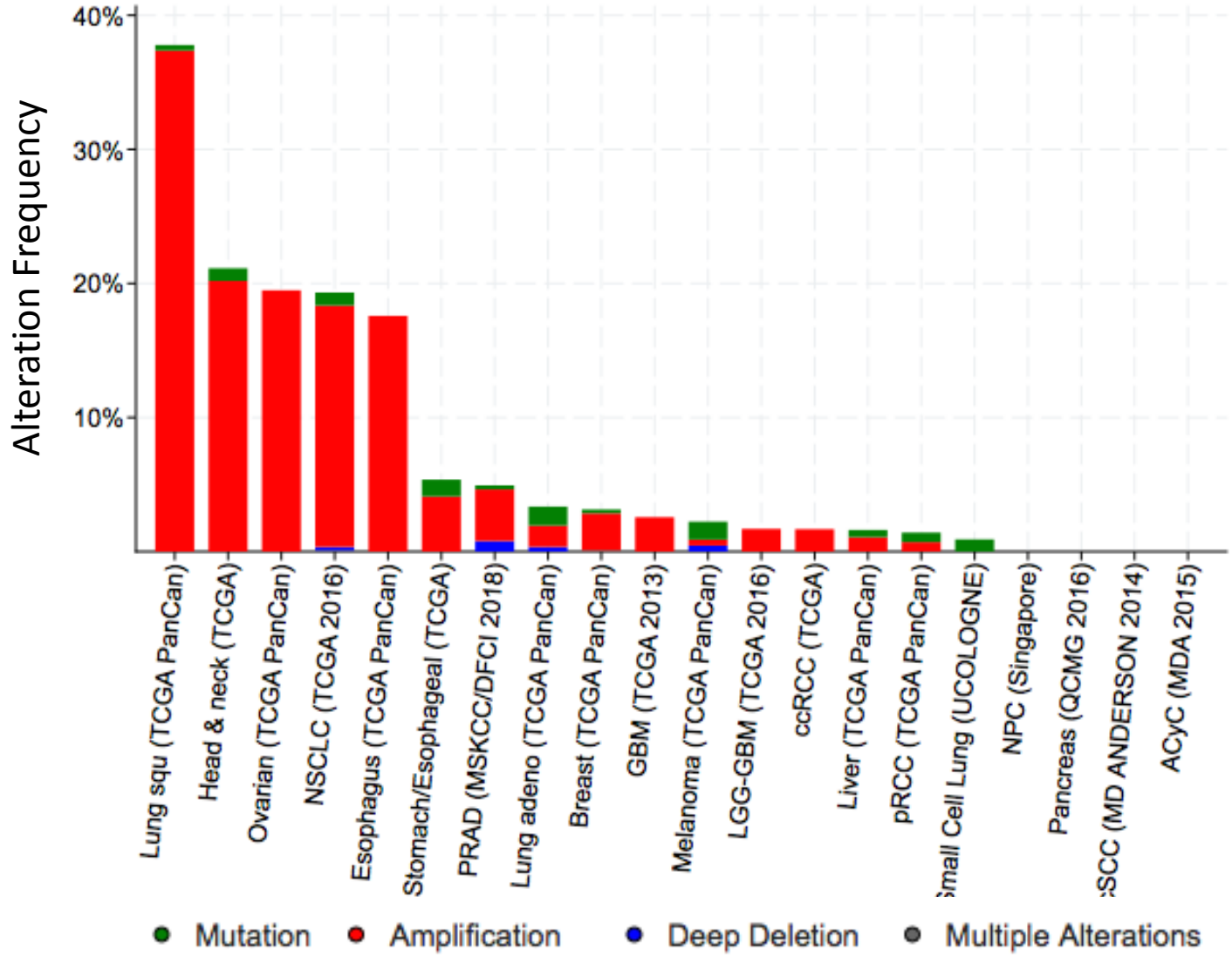
Kadoch et al. Human Cancer 2015
Kadoch et al. Biochemistry 2016

SWI/SNF complex widely considered as tumor suppressor



Hodges & Crabtree, 2016

ACTL6A (Baf53a) subunit of SWI/SNF complex is amplified in multiple cancers



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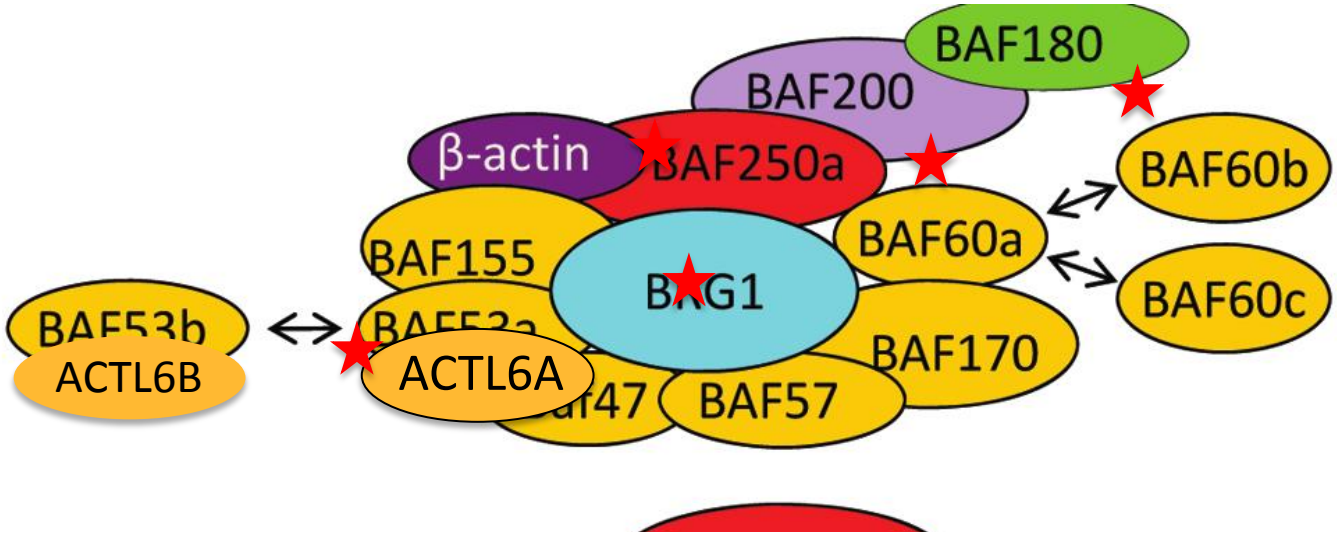
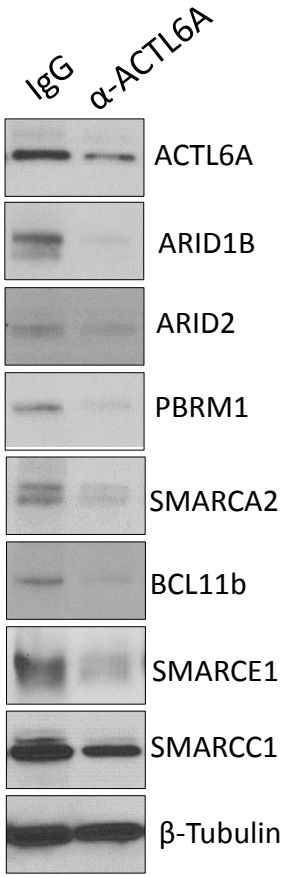


Immunodepletion

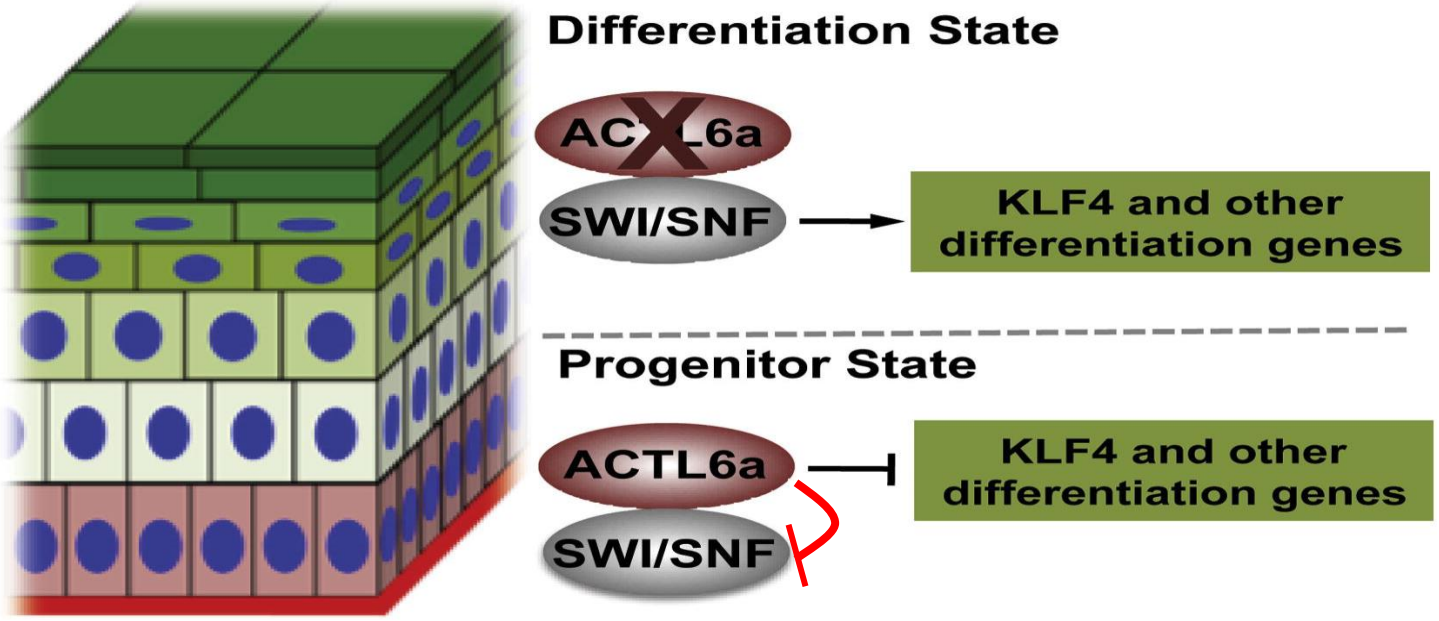


SWI/SNF complex subunits

ACTL6A (Baf53a): Progenitor State
ACTL6B (Baf53b): Differentiated State

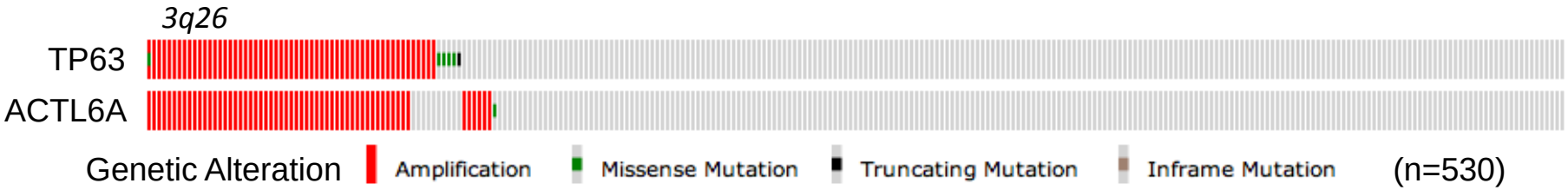
(Saladi SV, et al, *Epigenetics,* 2011)

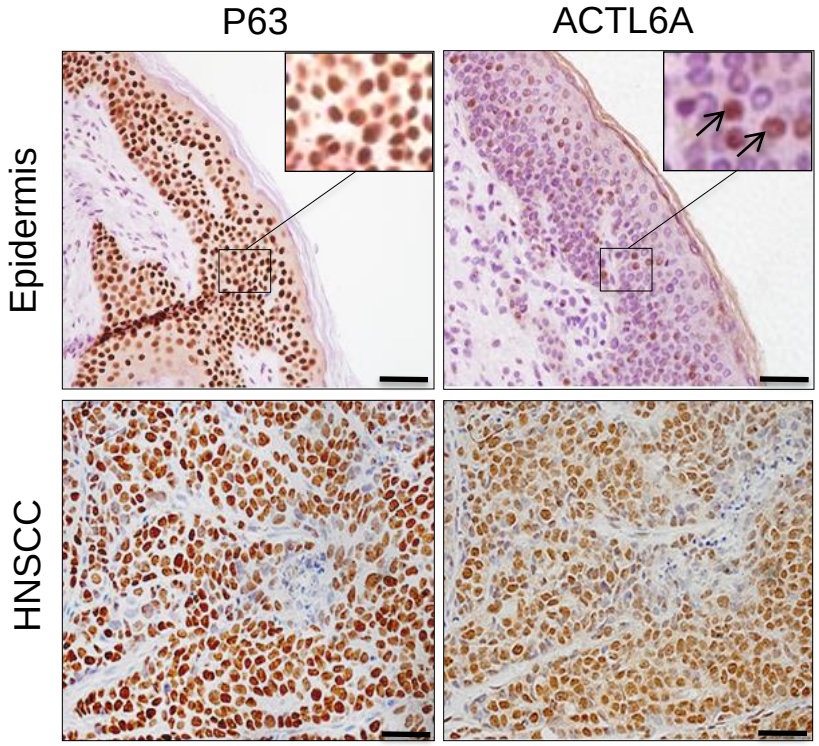
ACTL6A is required for maintaining the progenitor state in epithelium



(Bao X et al., *Cell Stem Cell* ,2013)

ACTL6A is co-amplified with p63 and confers poor prognosis in HNSCC





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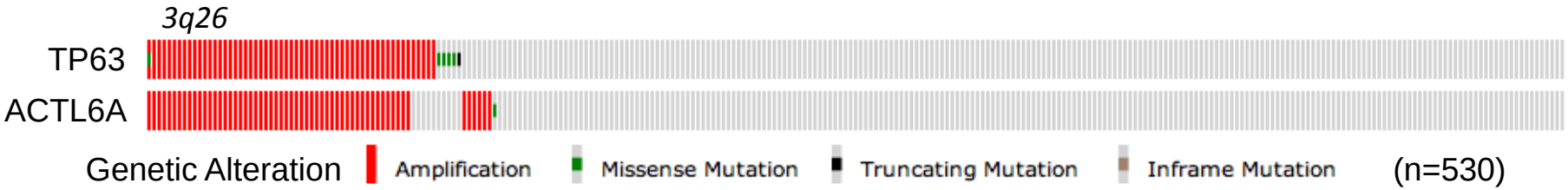
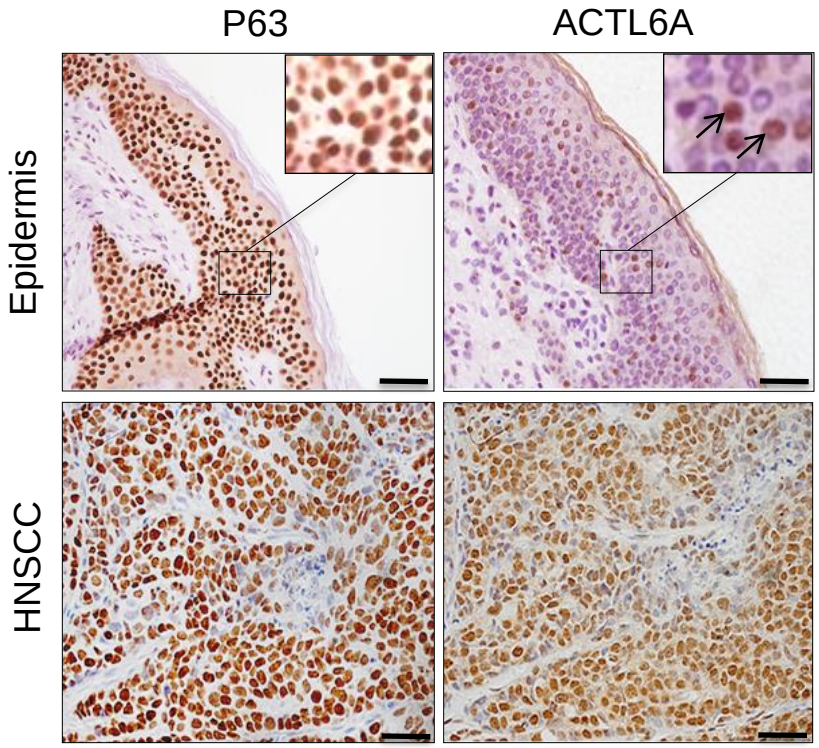
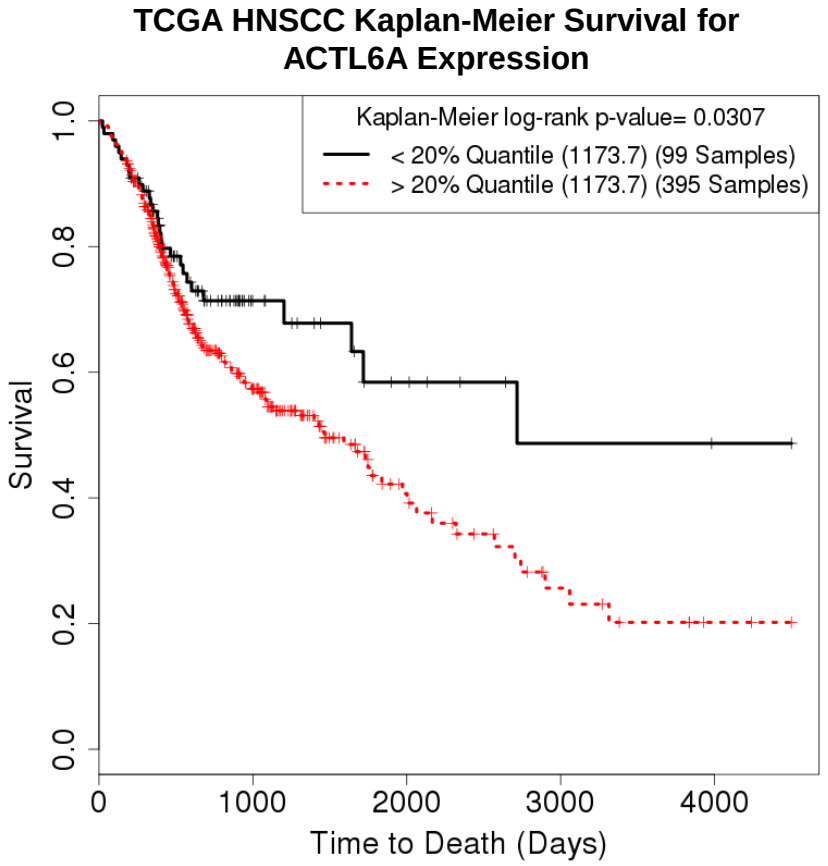
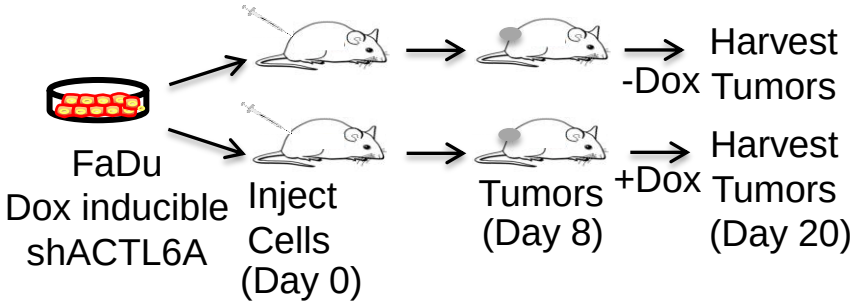
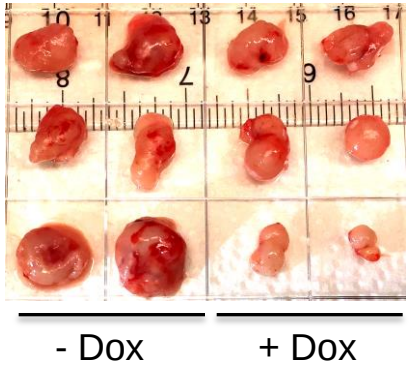


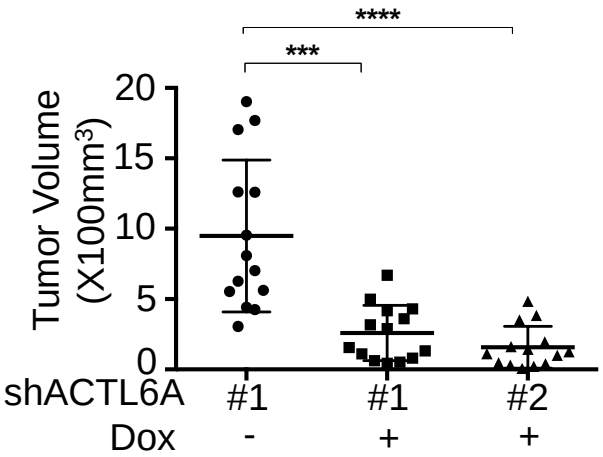




ACTL6A is required for tumor progression *in vivo*







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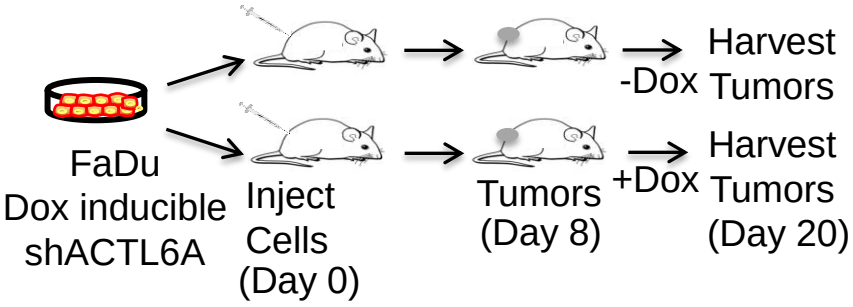
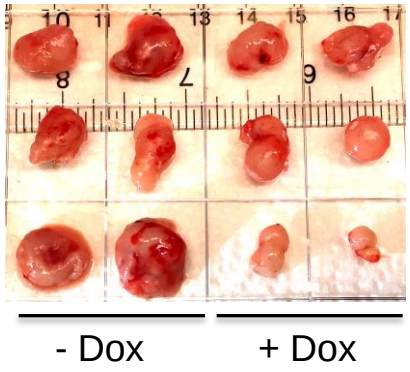
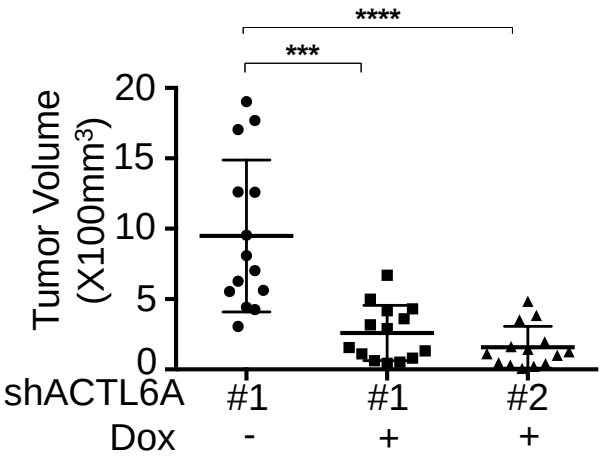
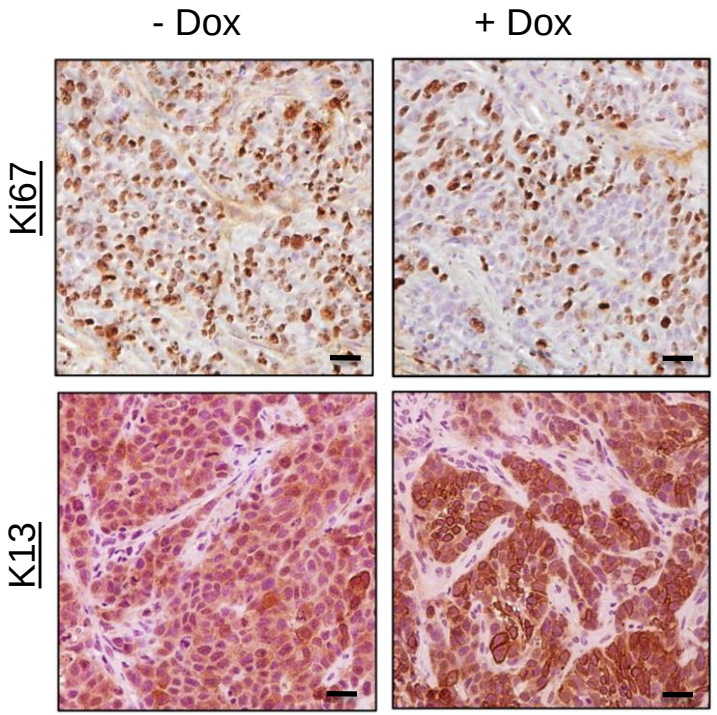
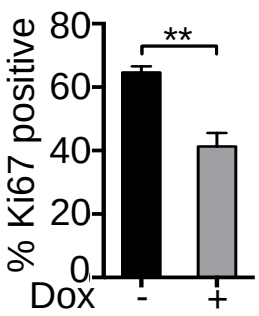
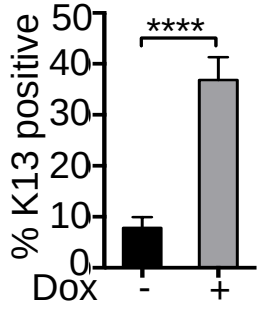




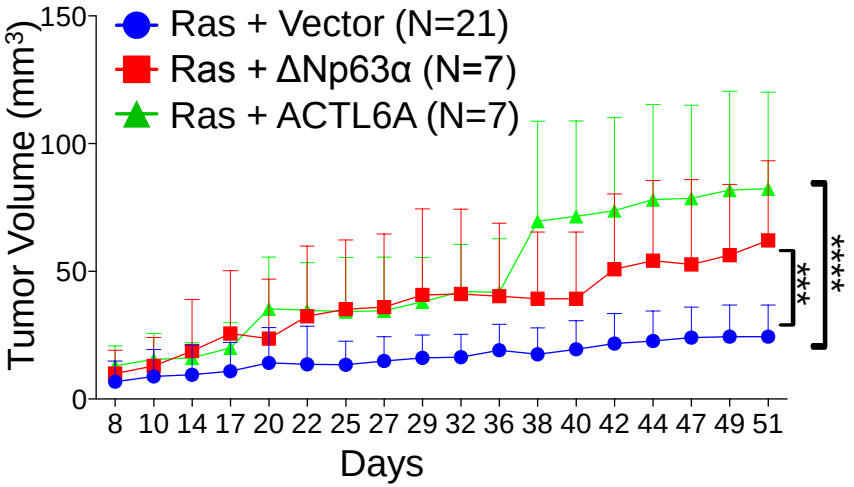








ACTL6A promotes transformation of mouse keratinocytes via activation of YAP



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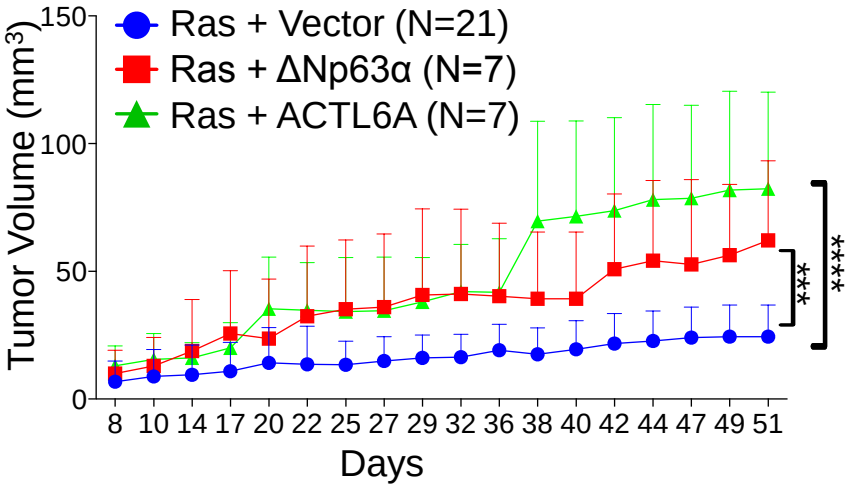
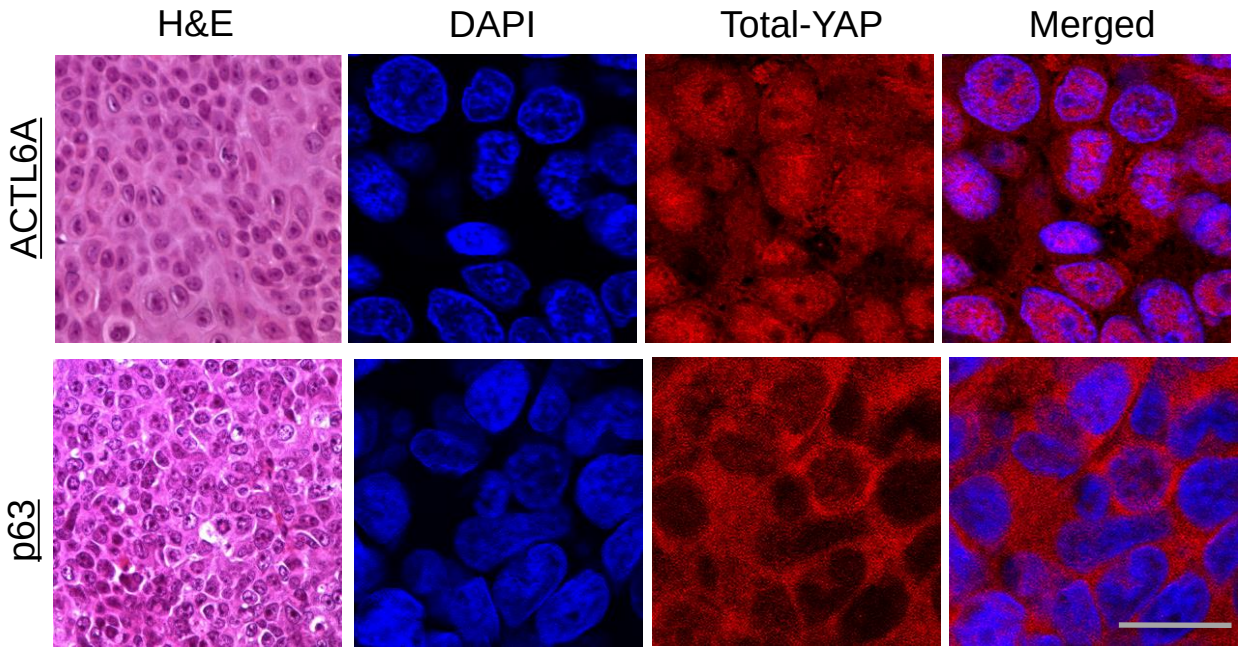
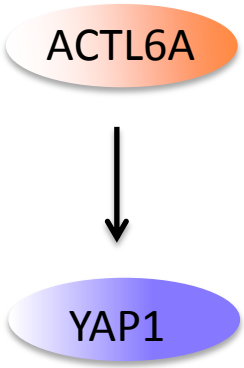
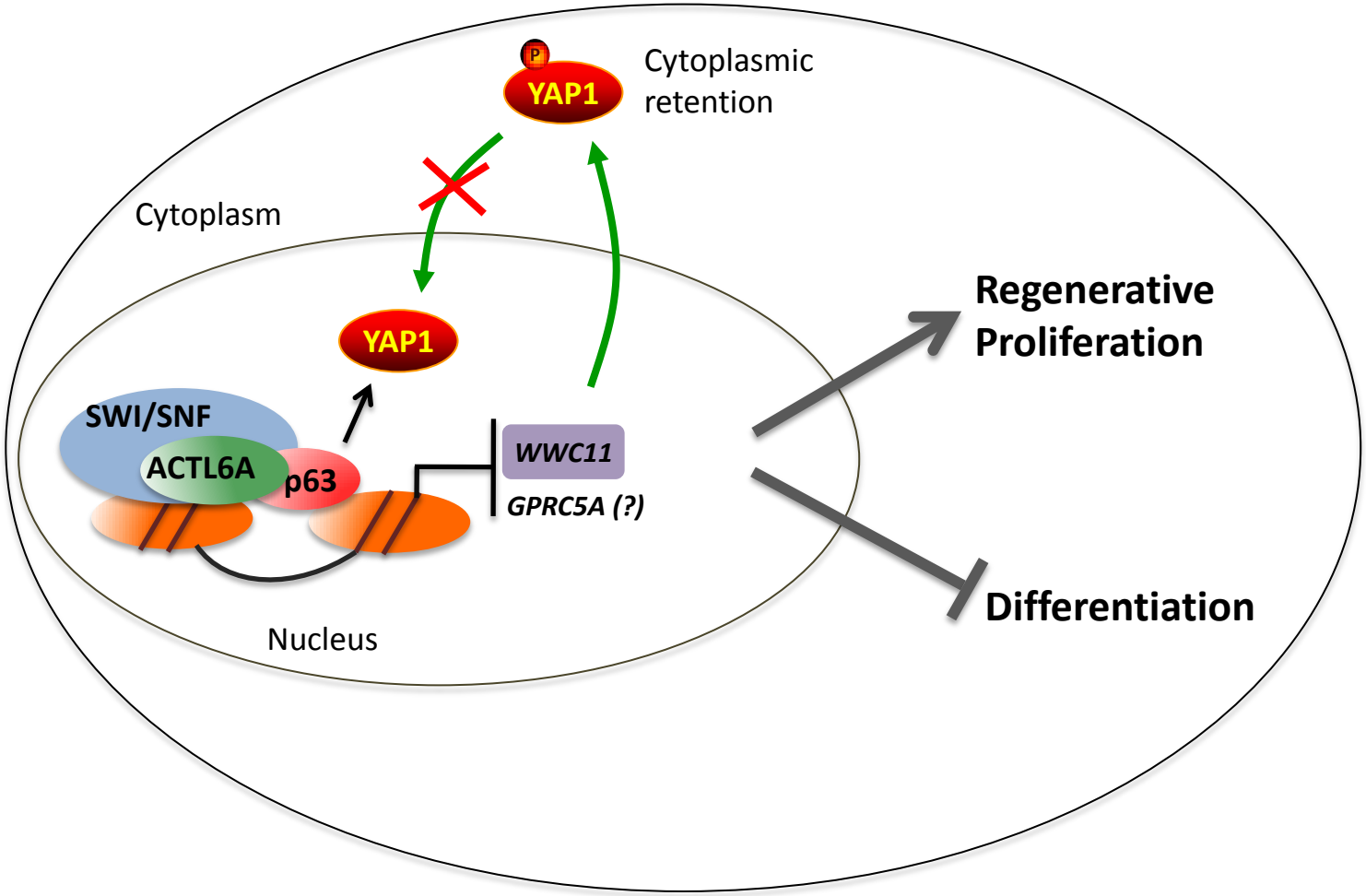




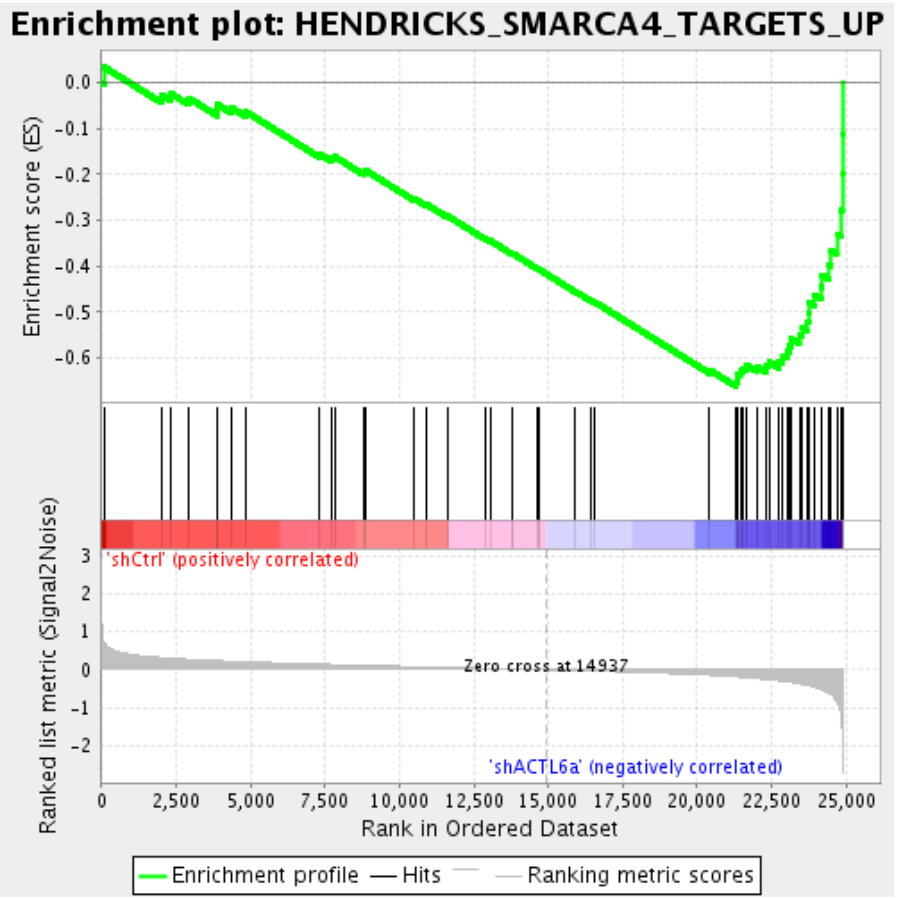


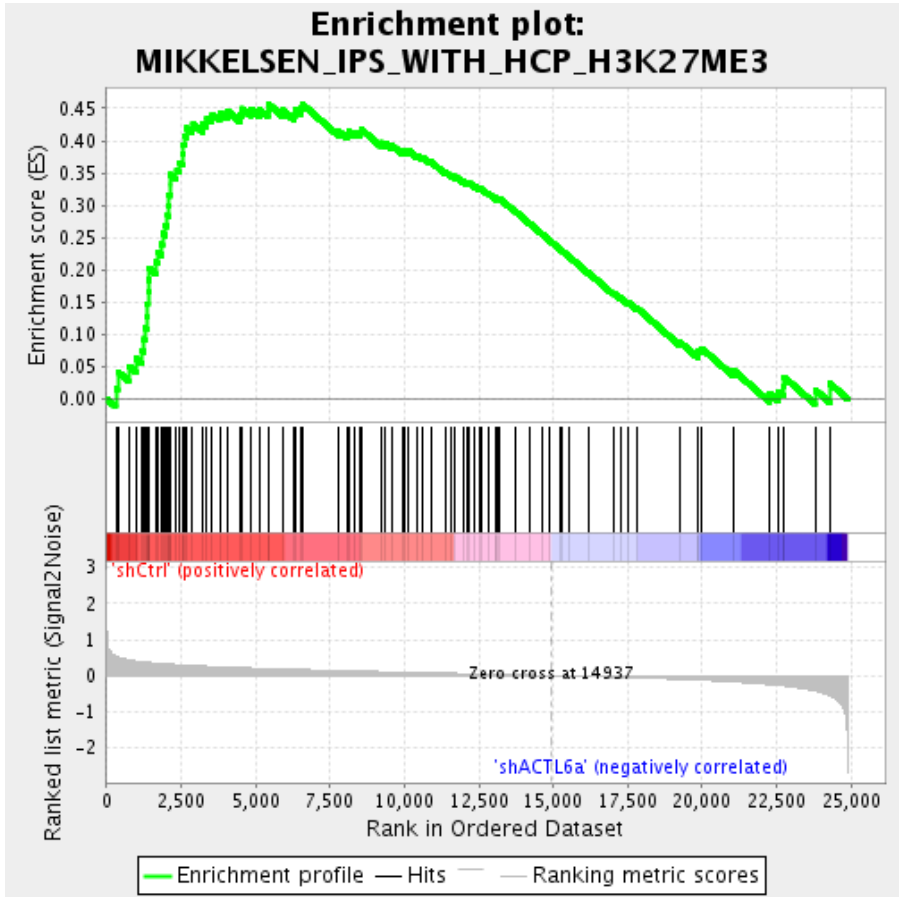
ACTL6A promotes Tumorigenesis in HNSCC



Cancer Cell, 2017

ACTL6A inhibits SMARCA4 (BRG1) and correlates to high H3K27me3 (EZH2) at its target genes





SWI/SNF antagonistic to PRC2 complex

Cancer Cell. 2010 Oct 19;18(4):316-28. doi: 10.1016/j.ccr.2010.09.006.

Epigenetic antagonism between polycomb and SWI/SNF complexes during oncogenic transformation.

Wilson BG[1], Wang X, Shen X, McKenna ES, Lemieux ME, Cho YJ, Koellhoffer EC, Pomeroy SL, Orkin SH, Roberts CW.

Author information

MCB, 2008

SWI/SNF Mediates Polycomb Eviction and Epigenetic Reprogramming of the *INK4b-ARF-INK4a* Locus

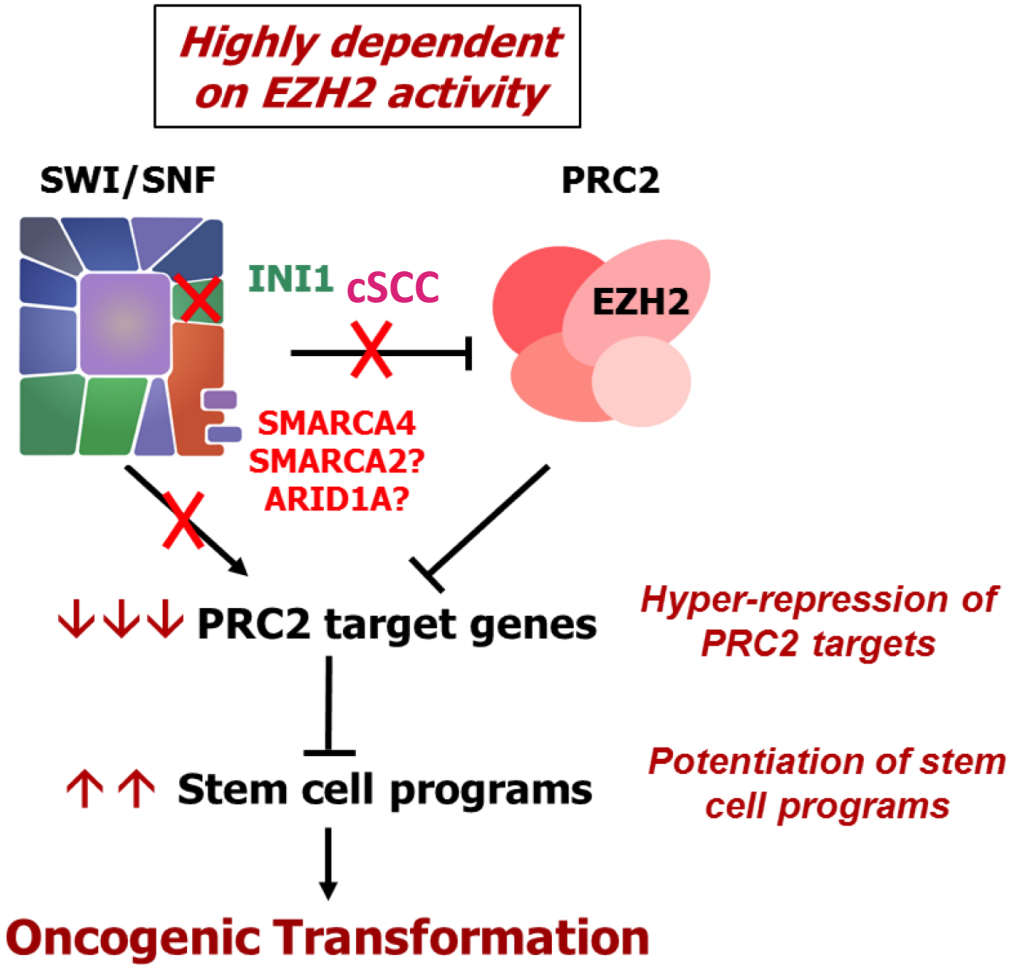
Sima Kheradmand Kia, Marcin M. Gorski, Stavros Giannakopoulos and C. Peter Verrijzer*

ARID1B is a specific vulnerability in **ARID1A**-mutant cancers.

Helming KC, Wang X, Wilson BG, Vazquez F, Haswell JR, Manchester HE, Kim Y, Kryukov GV, Ghandi M, Aguirre AJ, Jagani Z, Wang Z, Garraway LA, Hahn WC, Roberts CW.

Nat Med. 2014 Mar;20(3):251-4. doi: 10.1038/nm.3480. Epub 2014 Feb 23.

SWI/SNF antagonizes PRC2 complex

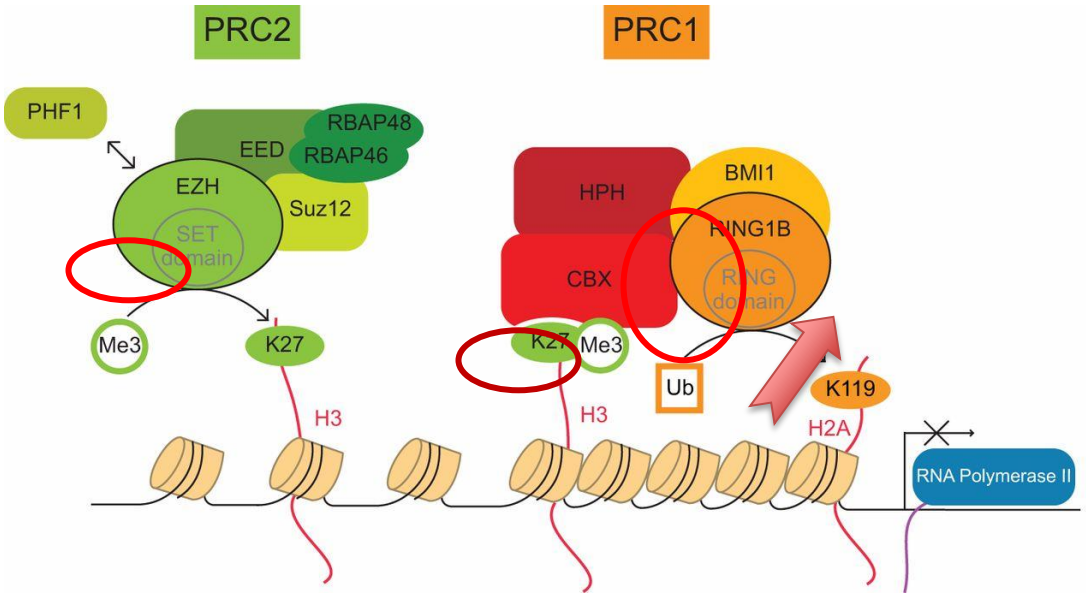


Kia SK,et al, *Mol Cell Bio*, 2008

Wilson B, et al, *Cancer Cell*, 2010

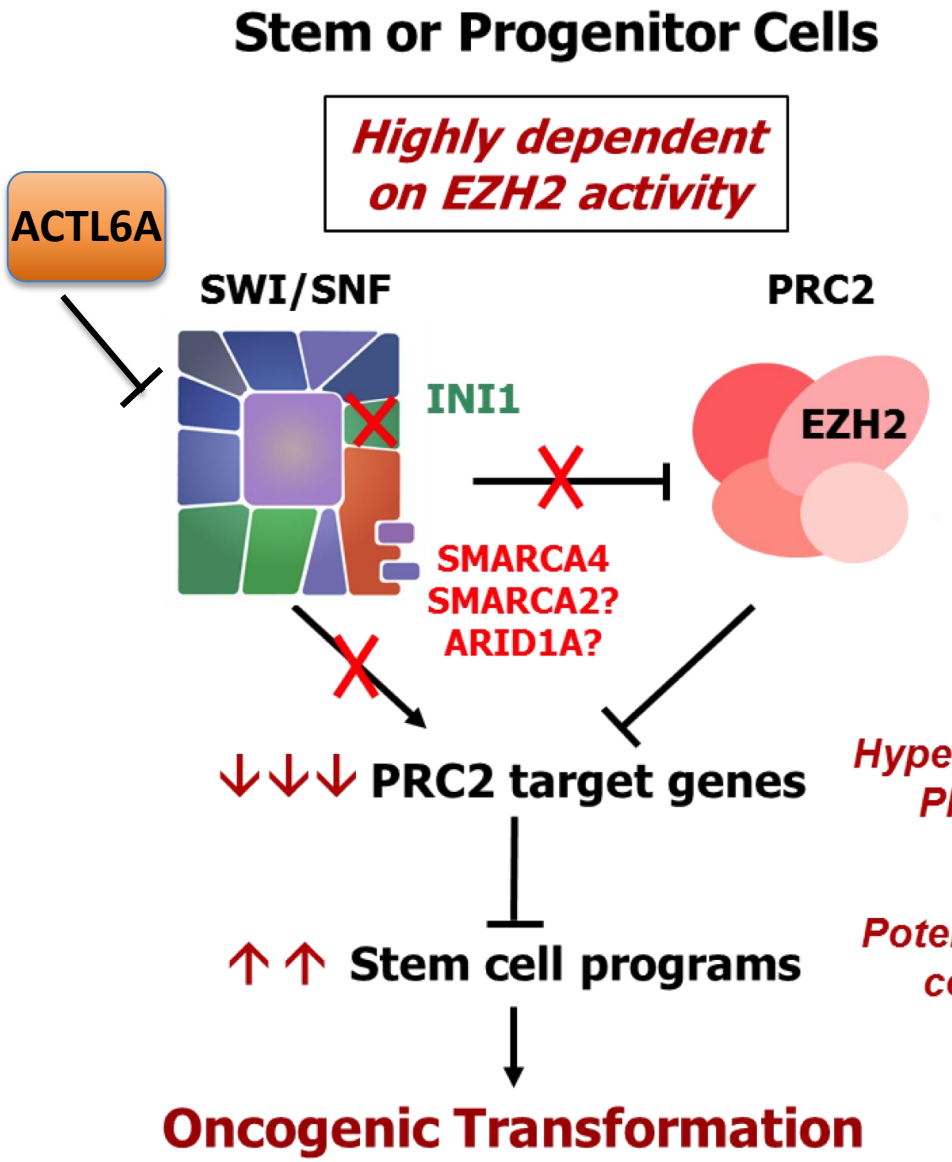
Saladi SV, et al, *Pigment Cell & Mel Res*, 2013

Poly Comb Repressor (PRC) complex



Herviou et al. Oncotarget - 2016

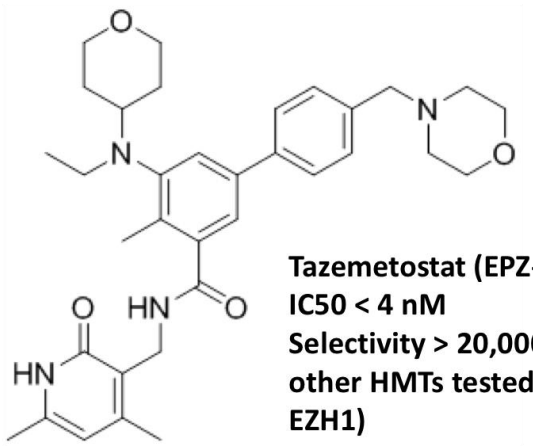
SWI/SNF complex subunits alterations renders cells sensitive to EZH2 inhibitors



Hyper-repression of PRC2 targets

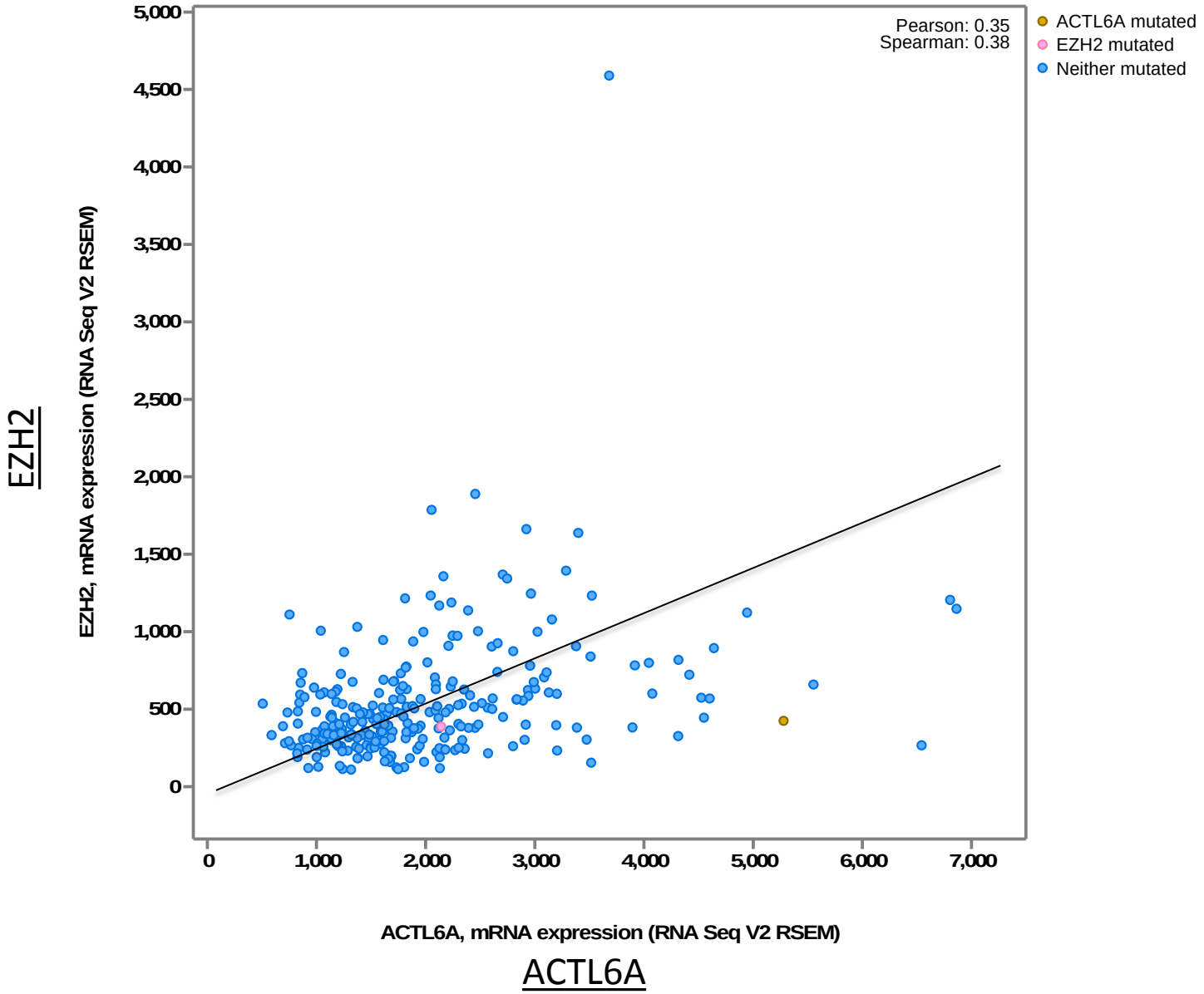
Potentiation of stem cell programs

Clinical Compound



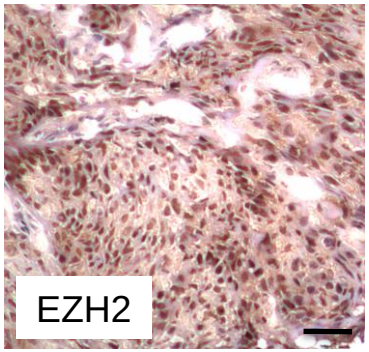
Tazemetostat (EPZ-6438)
IC50 < 4 nM
Selectivity > 20,000-fold over all other HMTs tested (100-fold for EZH1)

ACTL6A expression correlates strongly with EZH2 expression

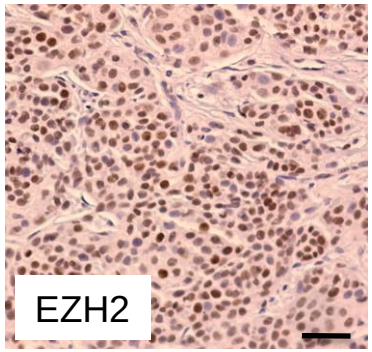


EZH2 in HNSCC patients

PDX (frozen)



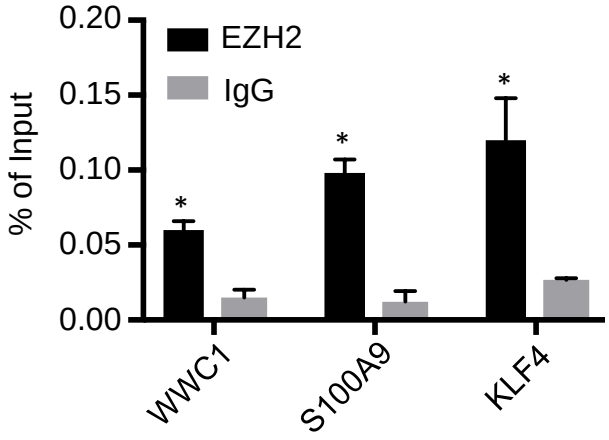
HNSCC (FFPE)

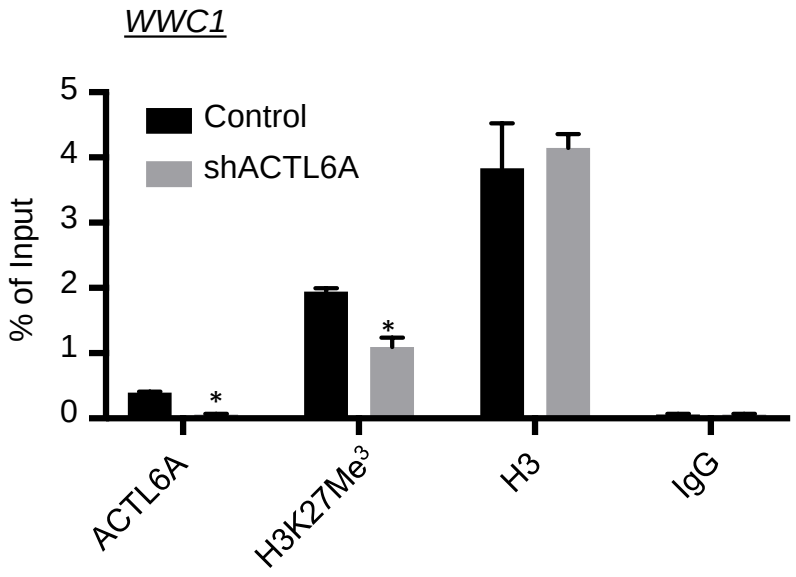


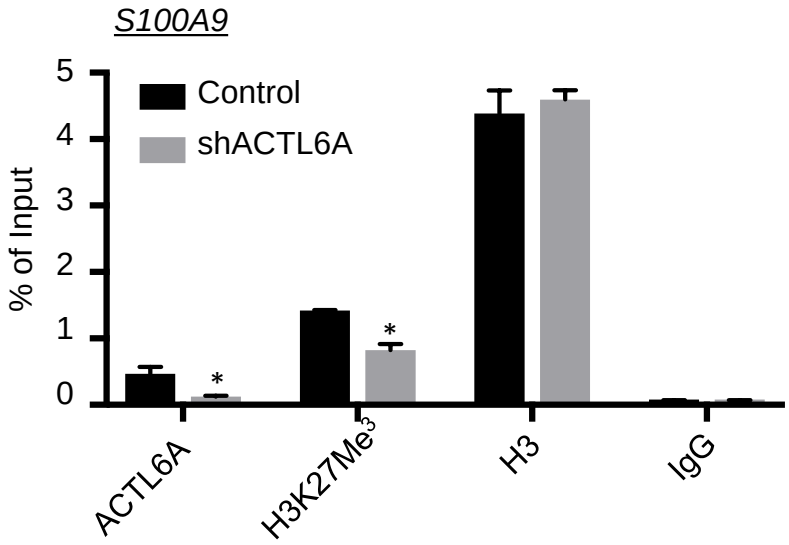
Scale bar represents 50µM

(PDX samples from Sara Pai)

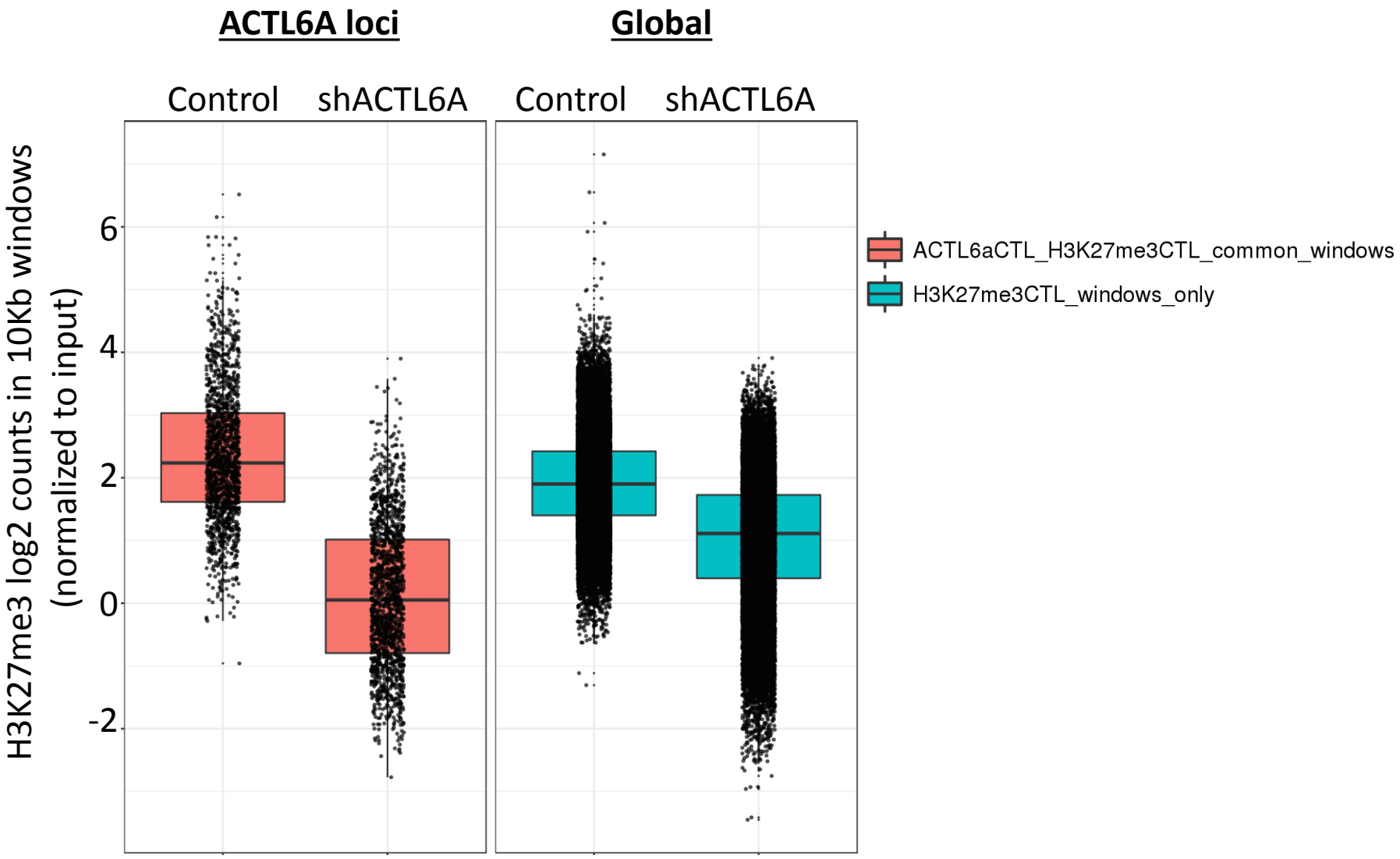
ACTL6A cooperates with EZH2 to repress differentiation programs



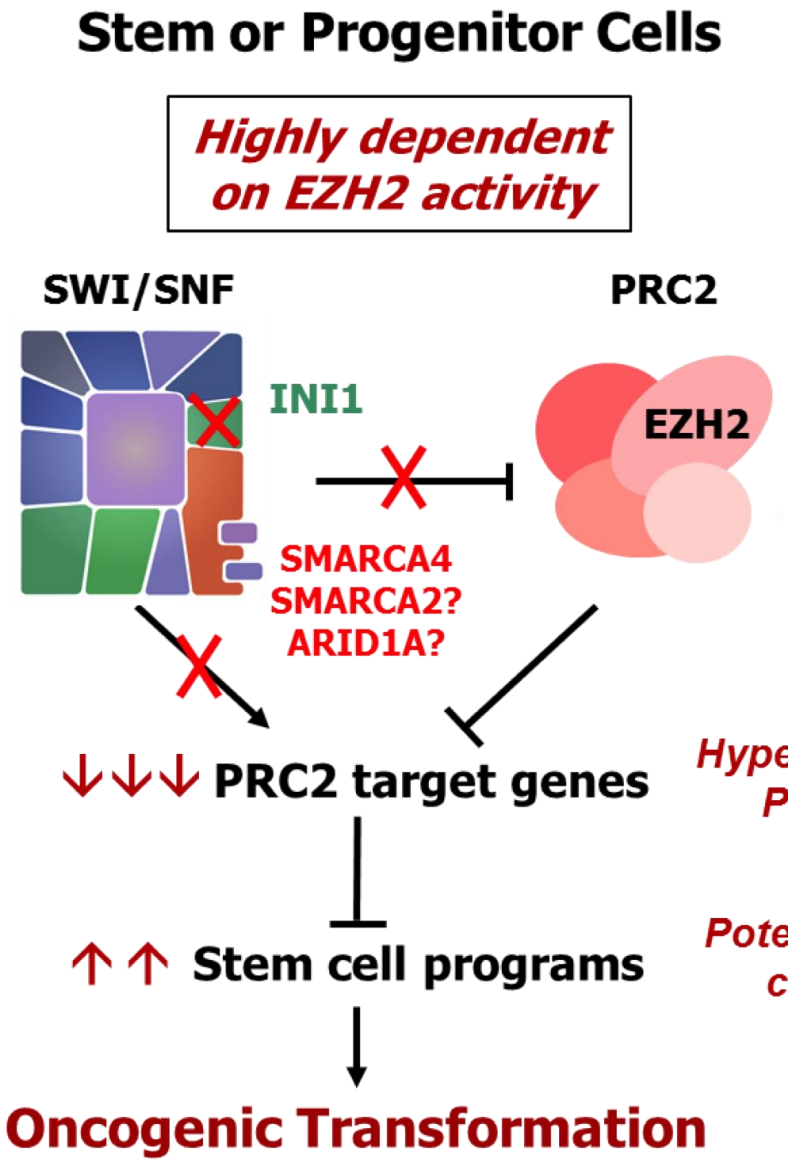




ACTL6A cooperates with EZH2 and promotes H3K27me3 globally



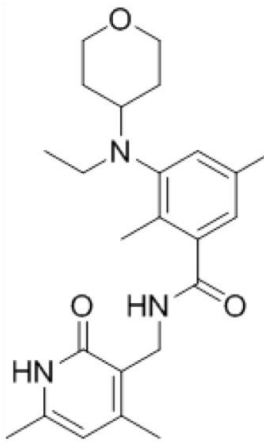
SWI/SNF complex subunits alterations renders cells sensitive to EZH2 inhibitors



Hyper-repression of PRC2 targets

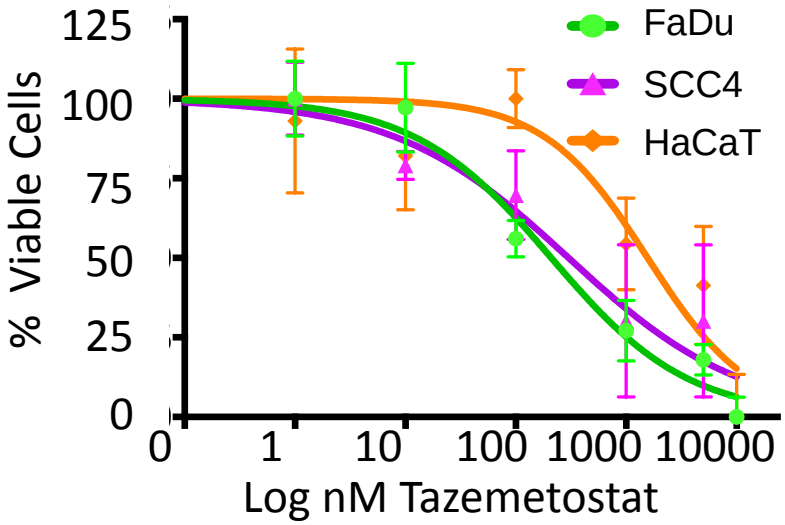
Potentiation of stem cell programs

Clinical Compound

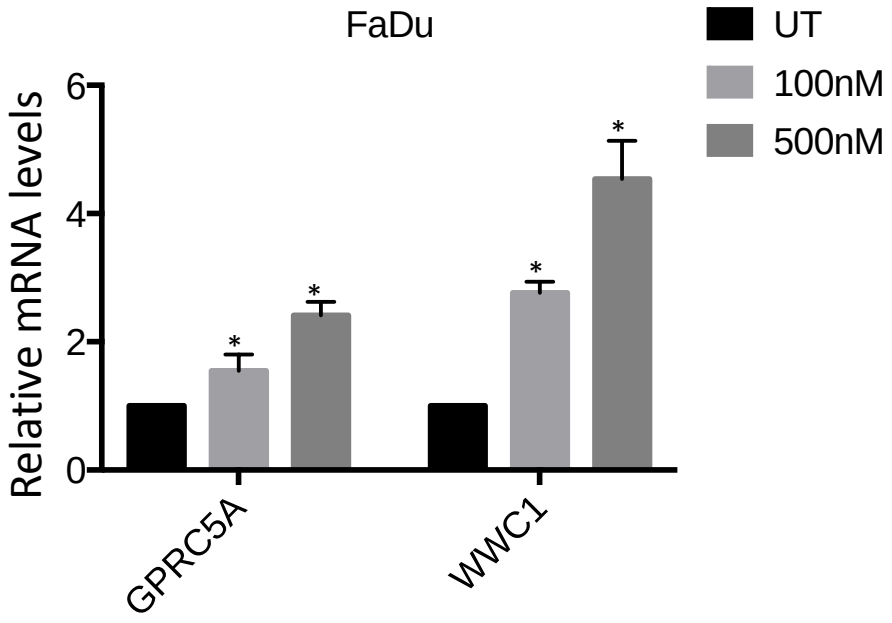


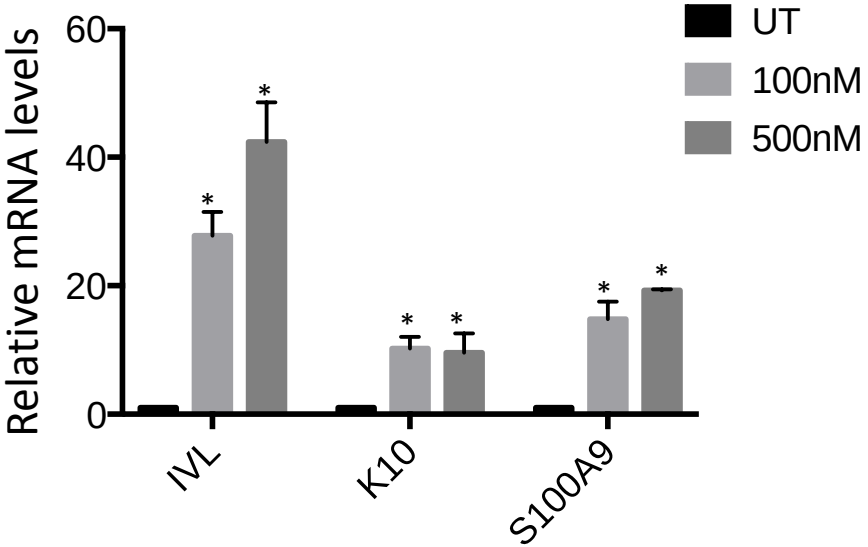
Tazemetostat (EPZ-6438)
IC50 < 4 nM
Selectivity > 20,000-fold over all other HMTs tested (100-fold for EZH1)

EZH2 regulates differentiation program in SCC

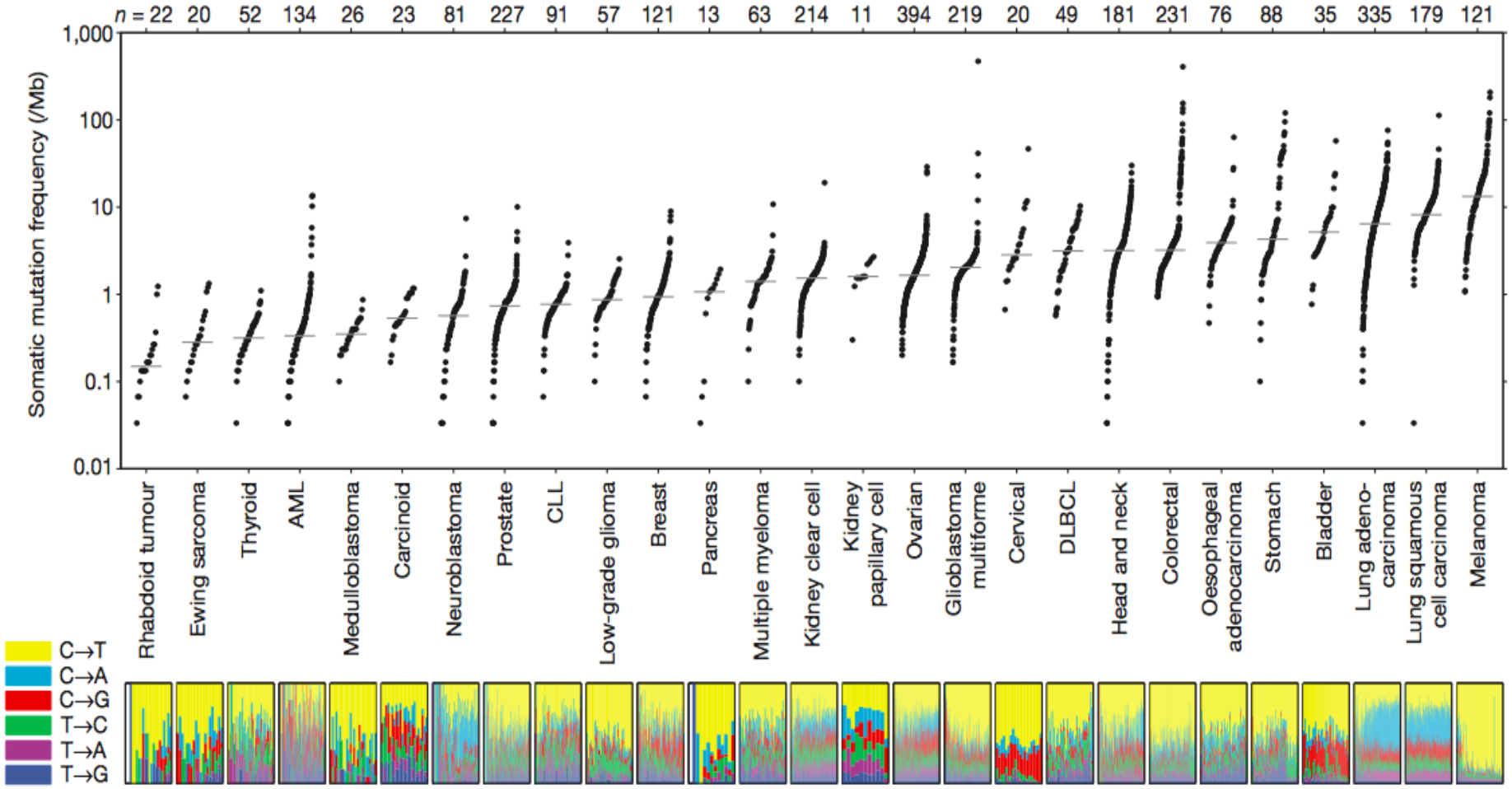


HaCaT to FaDu:p= 0.043
HaCaT to SCC4: p=0.013



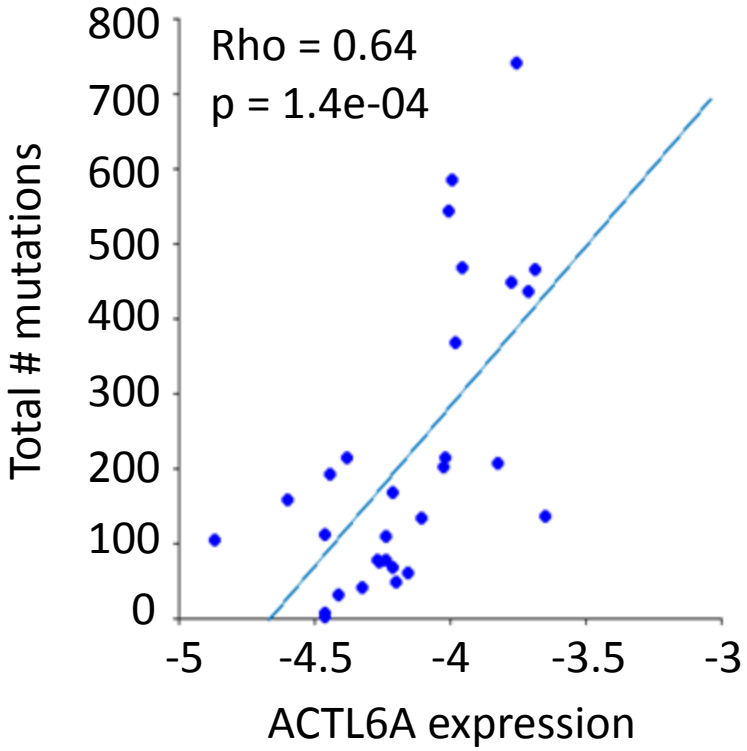


High mutation load in HNSCC



Lawrence M, et al Nature, 2013

ACTL6A correlates to high mutation load in HNSCC

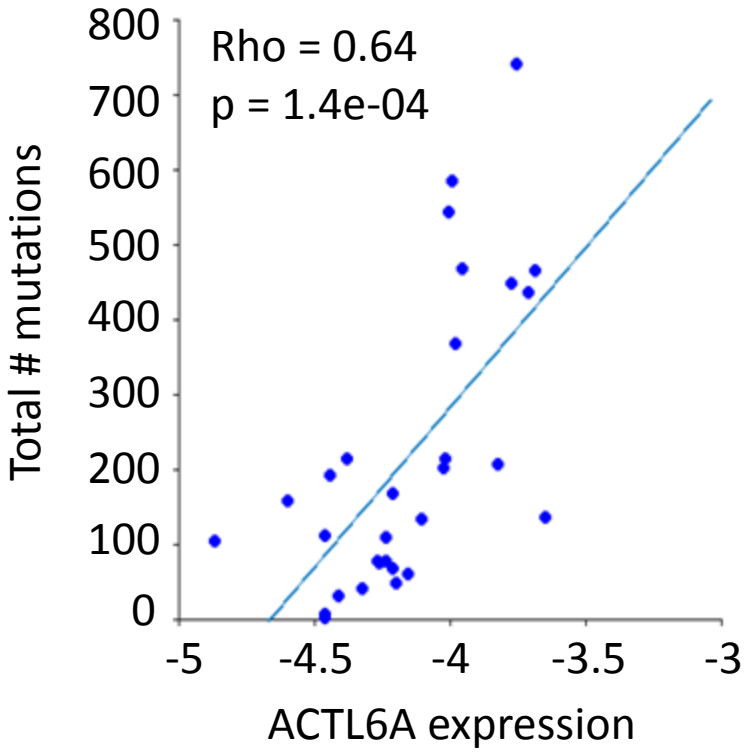


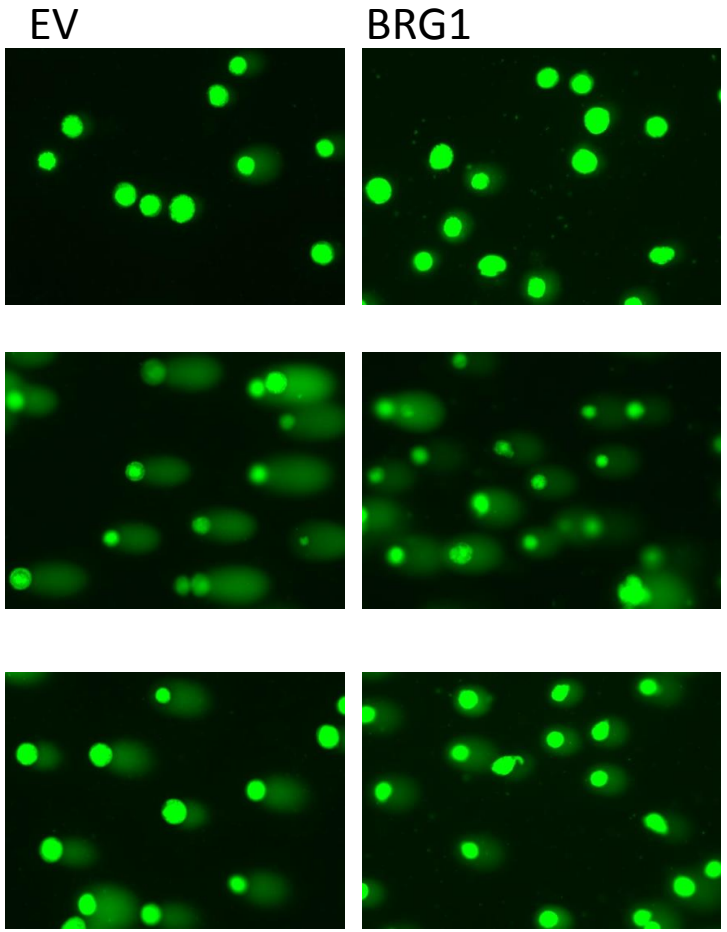
More Mutation Load

↓

Better response to Immune check point inhibitors

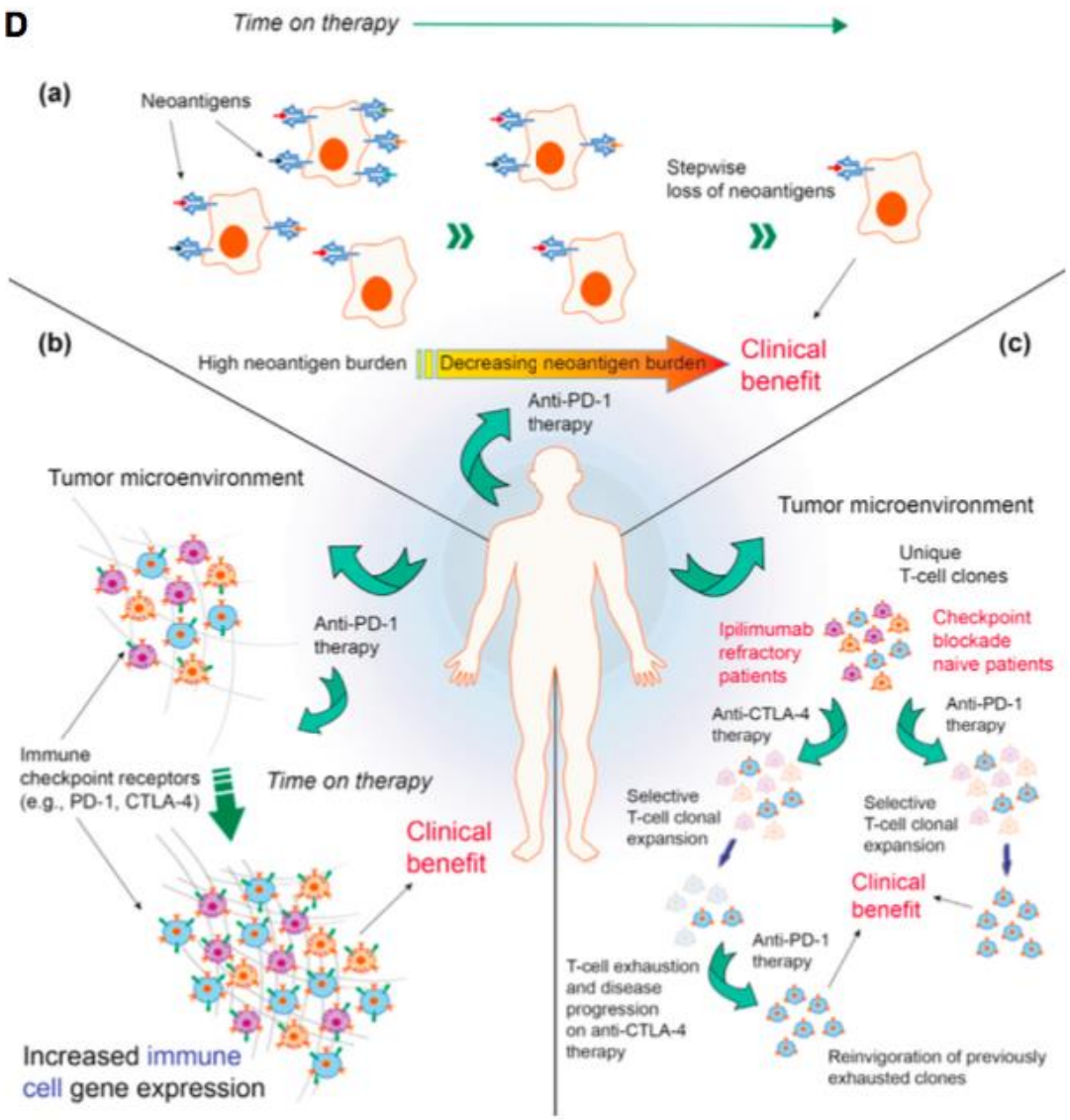
ACTL6A correlates to high mutation load in HNSCC





Comet Assay

High Mutation Load Predicts Better Clinical Outcome to Immune Check Point Inhibitors



Riaz N, Cell, 2017

High Mutation Load Predicts Better Clinical Outcome to Immune Check Point Inhibitors

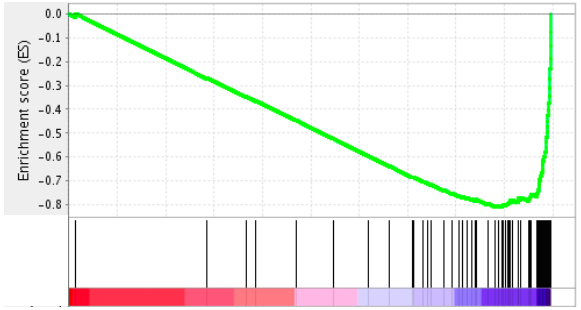
Only 15-20% respond to Immune Check Point Inhibitors in HNSCC

Ferris R, et al , *NEJM*, 2016

ACTL6A mediated immune modulation in HNSCC

HNSCC Cells

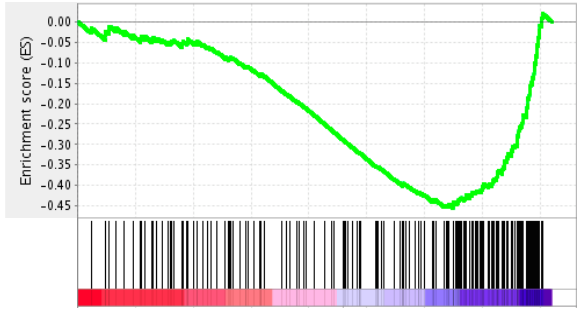
BROWNE_INTERFERON_RESPONSIVE_GENES



Control/shACTL6A; HIGH → LOW

HNSCC Primary Tumors

PD1_LIGATION_VS_CTRL_IN_ACT_TCELL_LINE_UP

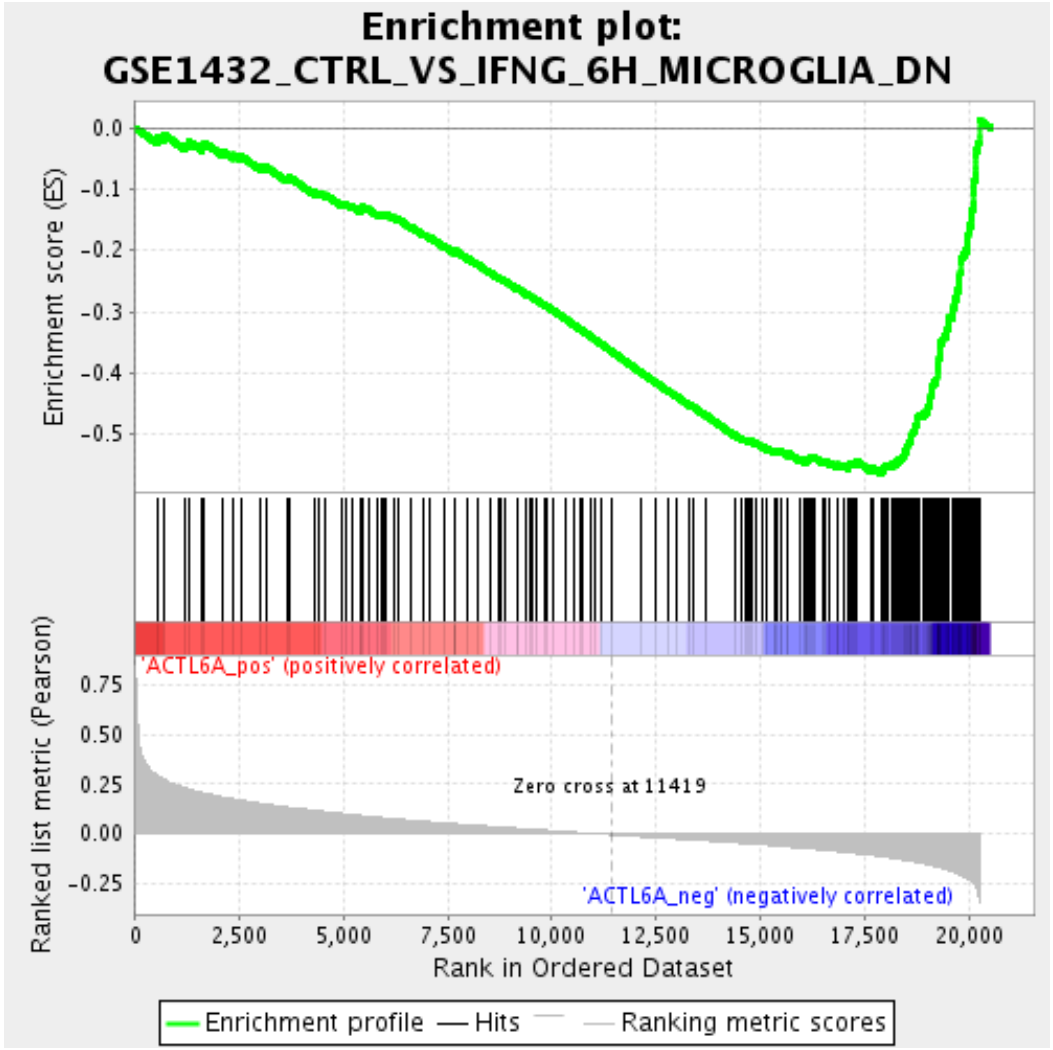


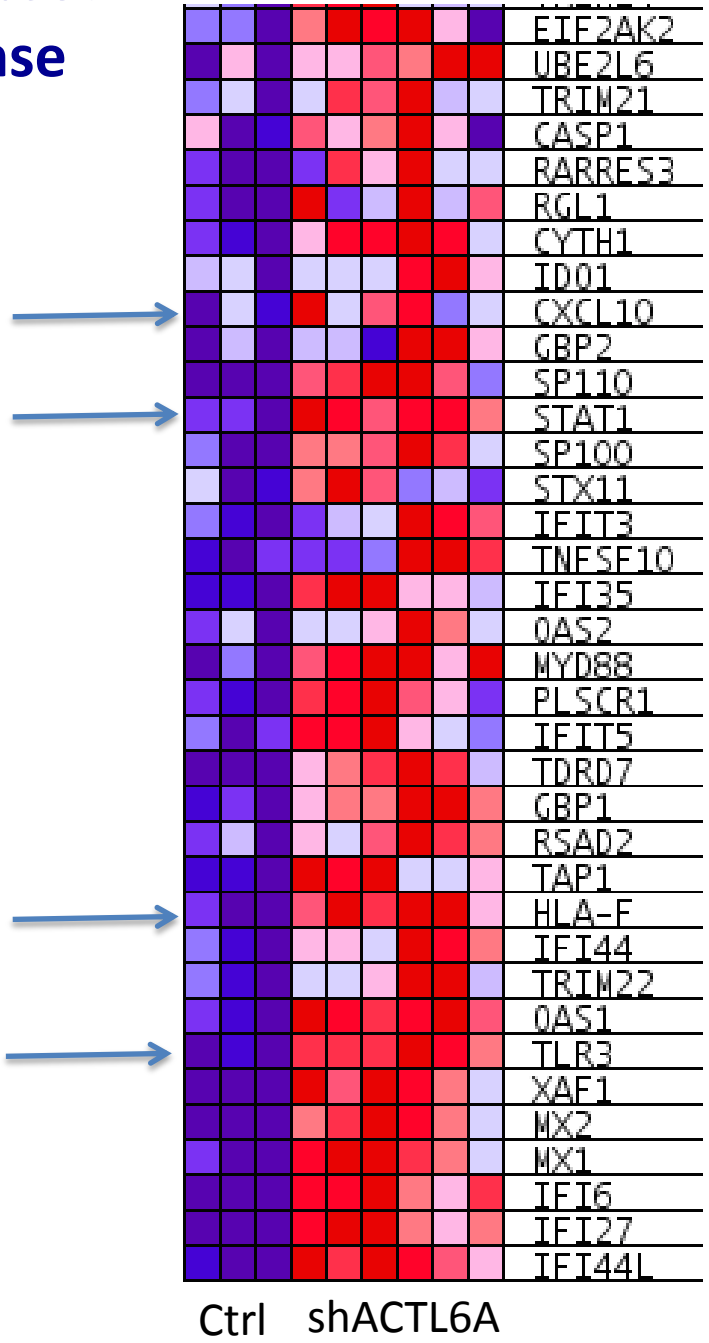
Correlation with ACTL6A; HIGH → LOW

Repressed Programs

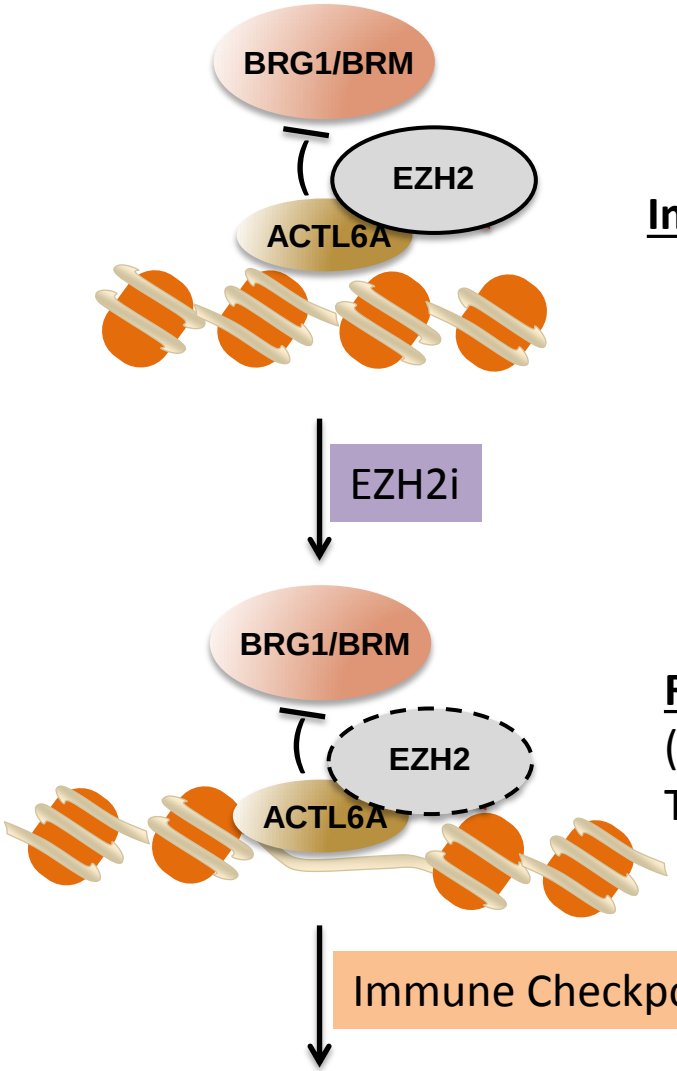
Predicts PD1/PD-L1 response in HNSCC
HLA-DRA
CXCL-10
STAT1/2
PDL-2
Cytotoxic T Cell Factors
Granzyme A,B
IL-2R, 9R, 21R
ICOS co-stimulator
HLA Components
HLA-B, C, E
HLA-DRA, B6, B1; -DPA1, B1
TNF Family Members
Fas Ligand
Lymphotoxin Alpha, Beta

ACTL6A suppresses the InterferonG gene subset thereby evading the tumor immune response





Predictive Treatment Model for HNSCC patients: Epigenetic therapy coupled with Immunotherapy

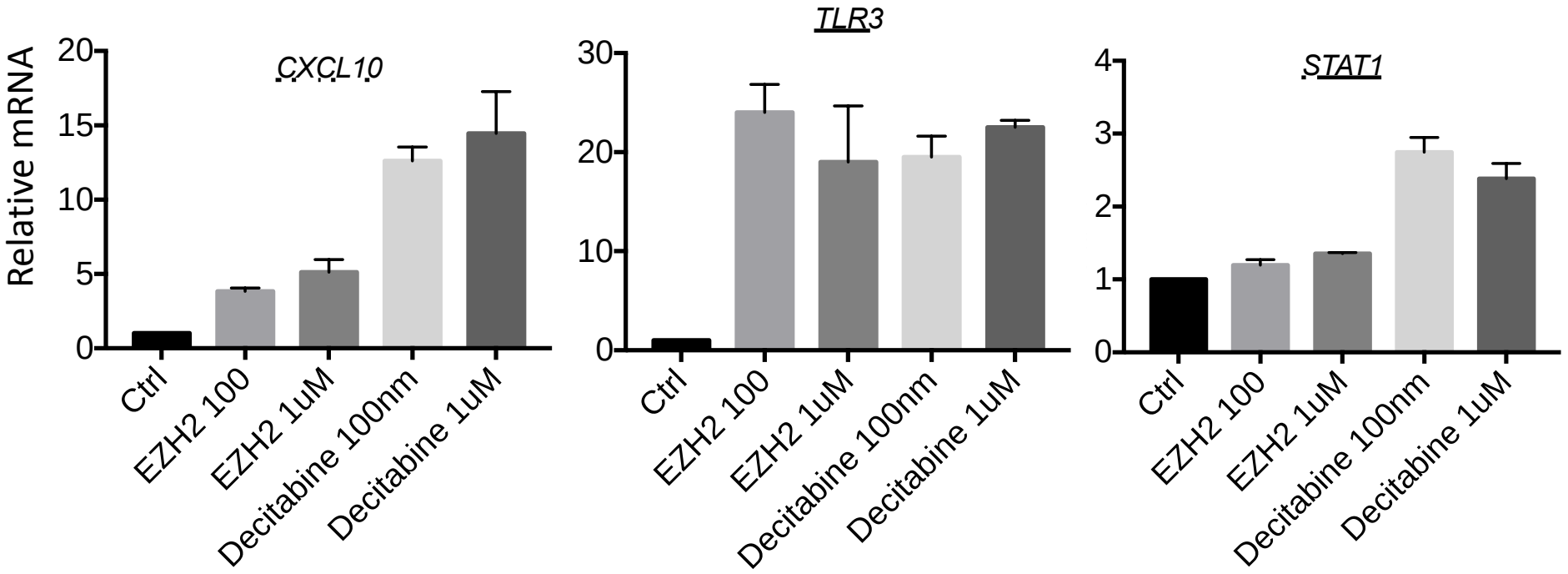


Immunologically Cold Tumors

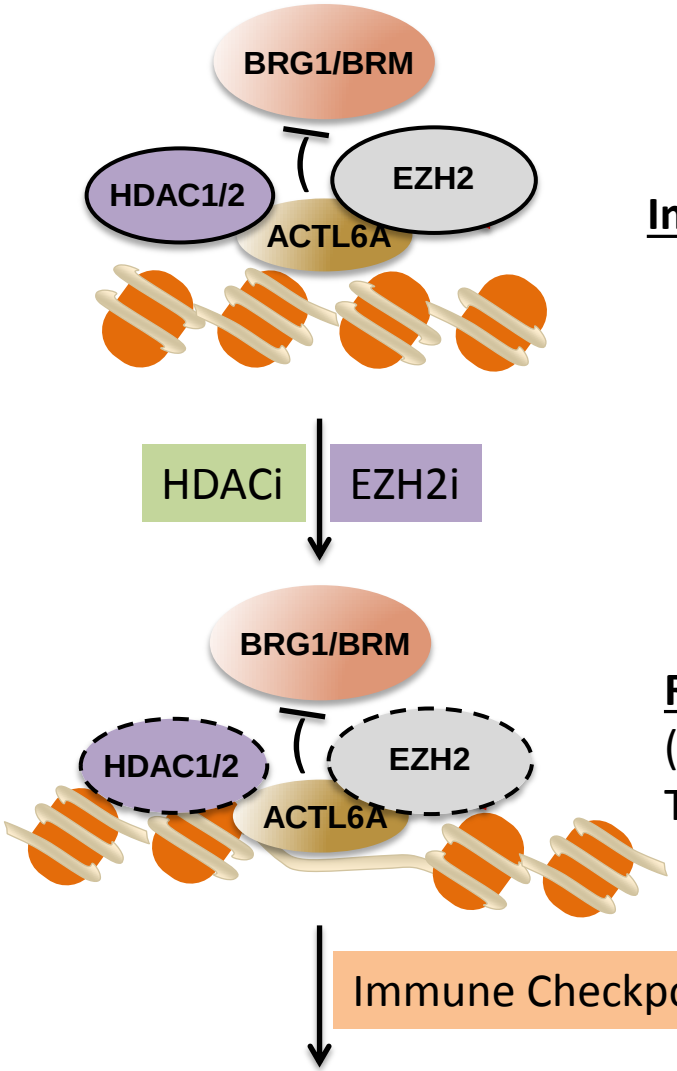
Reactivation of Immune program
(Interferon gamma responsive genes,
T Cell responsive genes)

(Saladi SV, et al., *Manuscript in prep)*

EZH2 and DNMT inhibitors reactivate Immune program



Predictive Treatment Model for HNSCC patients: Epigenetic therapy coupled with Immunotherapy



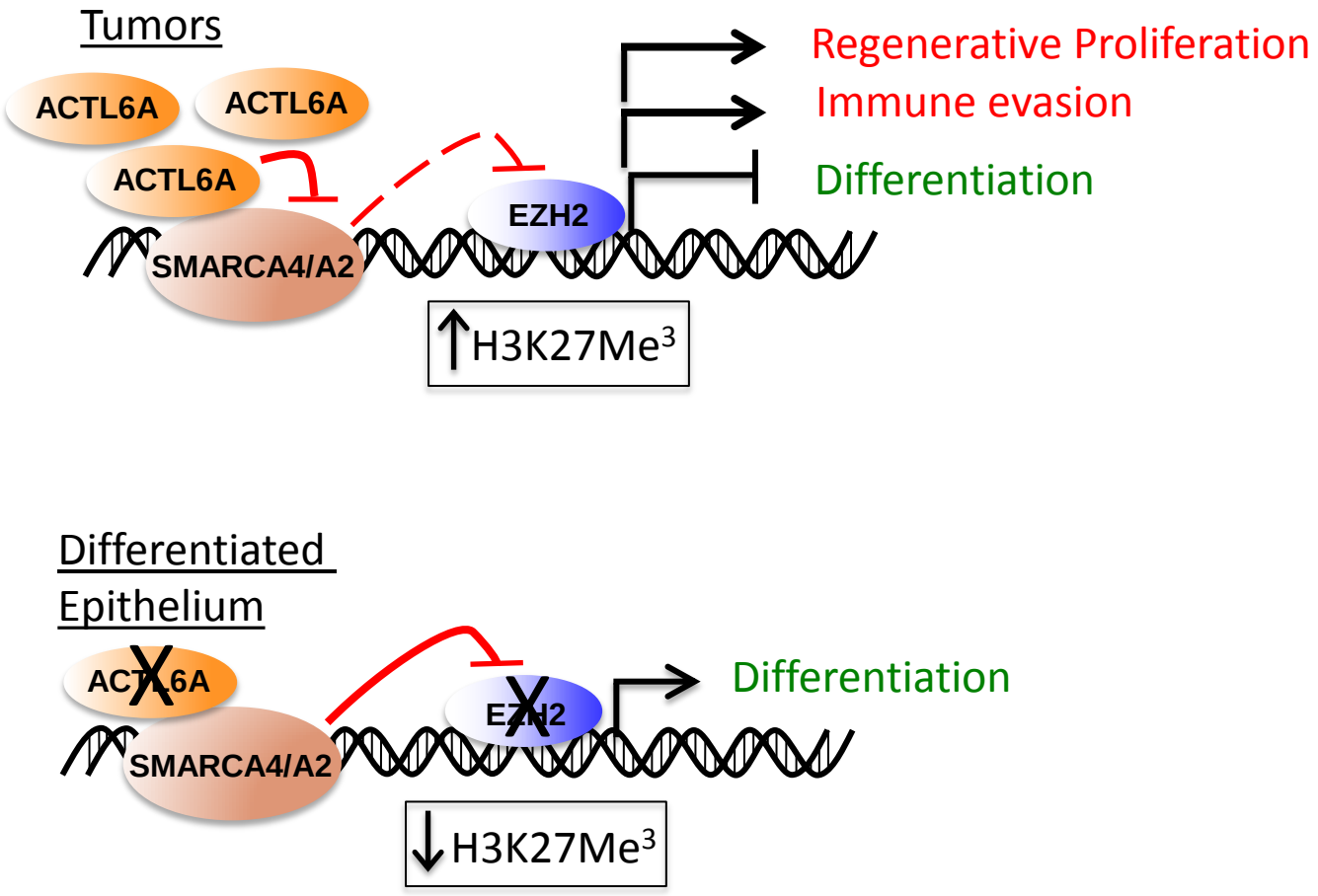
Immunologically Cold Tumors

Reactivation of Immune program
(Interferon gamma responsive genes, T Cell responsive genes)

Better Clinical response

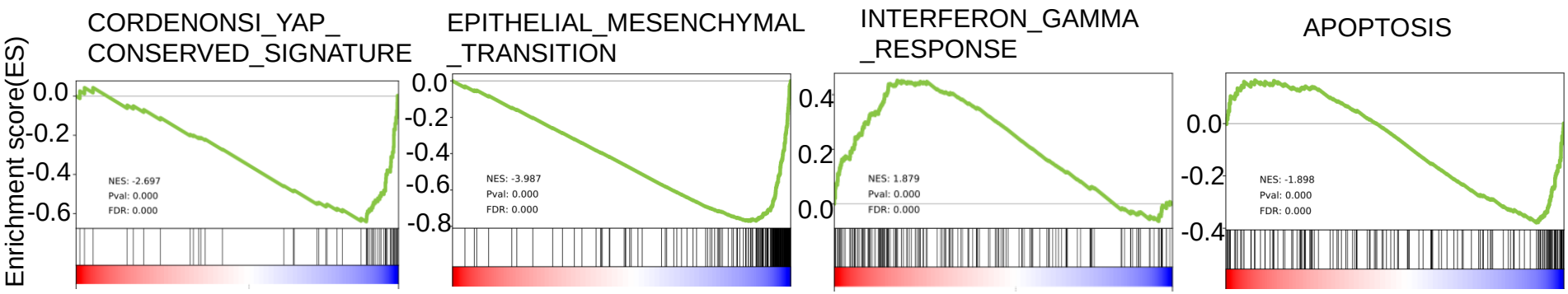
(Saladi SV, et al., *Manuscript in prep)*

ACTL6A, subunit of SWI/SNF complex is a driver oncogene in HNSCC



ACTL6A/YAP-dependent regenerative program in squamous carcinomas

GSEA analysis with BRD4 inhibition(JQ1 treatment) in all cancer cell line



Acknowledgements

Saladi Lab

Nana Chen
Rozenn Riou
Nicholas Abt
Pavan Kannan
Muna Alhusban

MEEI

Mark Varvares
Kevin Emerick
Derrick Lin

MGH CRM

Jayaraj Rajagopal
Brian Lin

MGH Cancer Center

Michael Lawrence
Leif Ellisen
Genevieve Boland
Bradley Bernstein
Soldano Ferrone
Robert Morris

Dana Farber

Martin Sattler
David Frank

Funding

Massachusetts Eye and Ear

NORMAN KNIGHT CHARITABLE FOUNDATION

MRF MELANOMA RESEARCH FOUNDATION