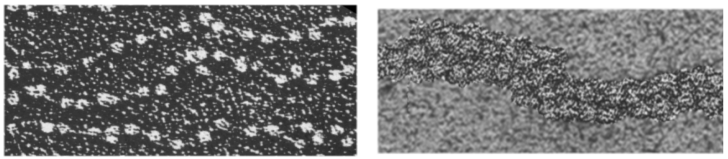
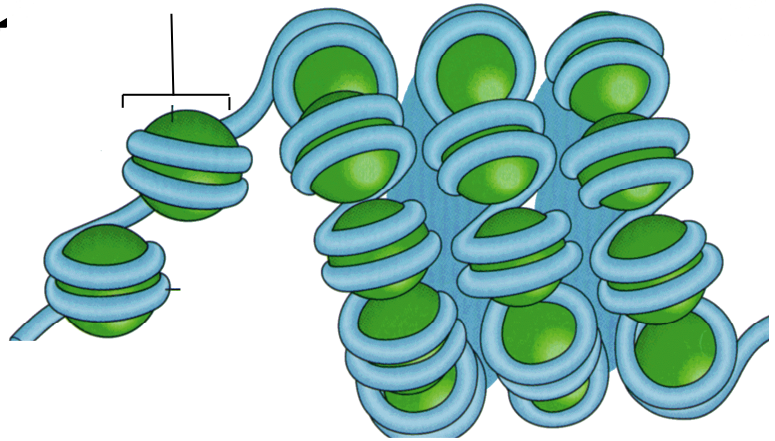
‘Epigenetic Regulation In Cancer’

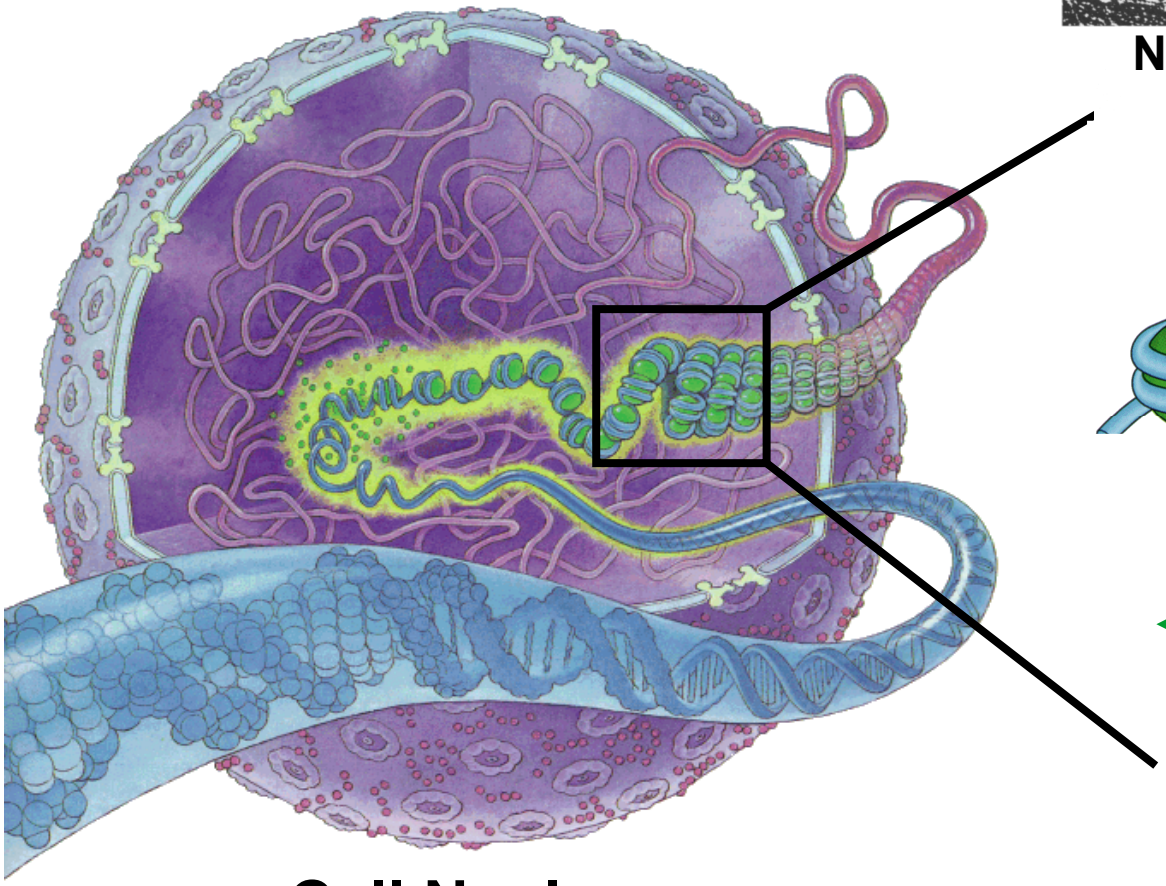
Dr Mark Dawson
University of Cambridge

Chromatin



Nucleosome





Cell Nucleus

'DNA compaction'

'open' chromatin

'condensed' chromatin

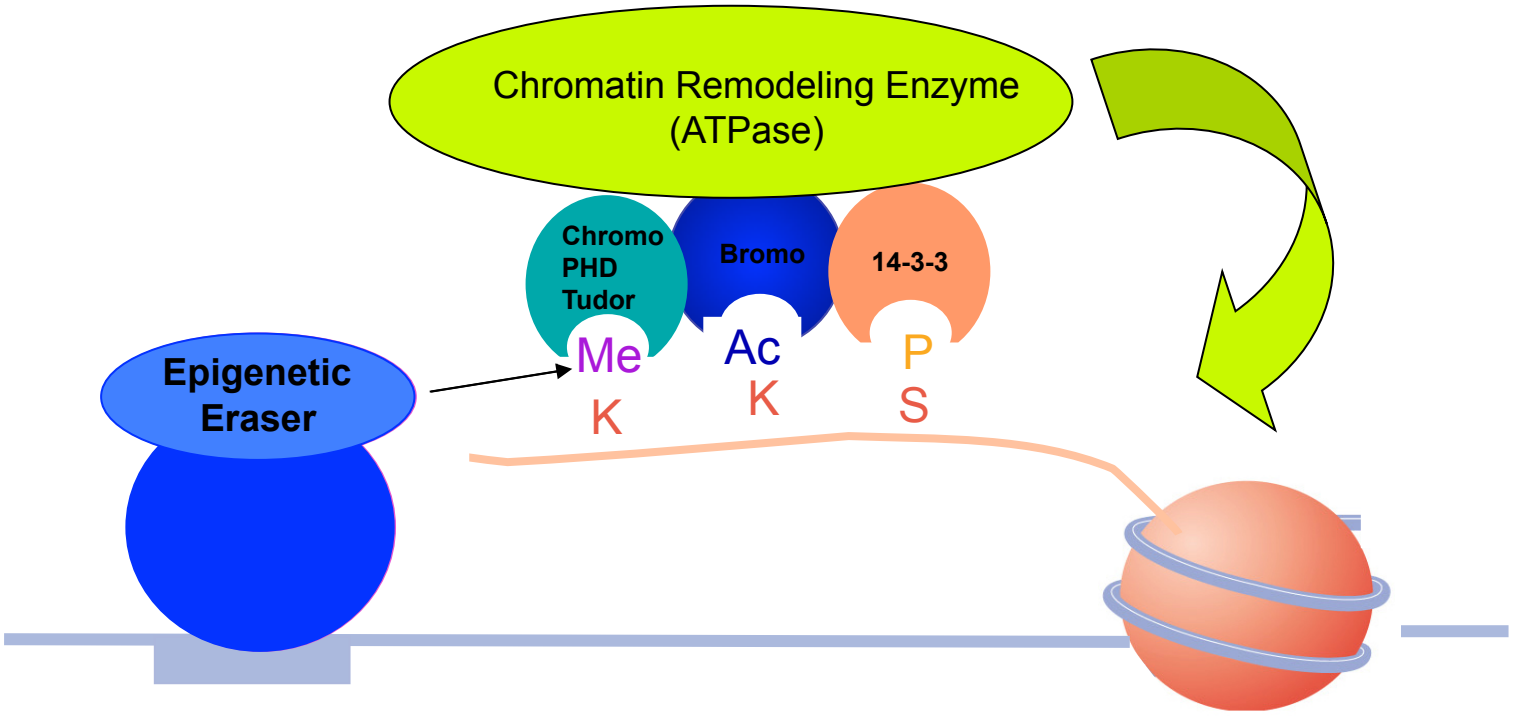
Active genes

Euchromatin

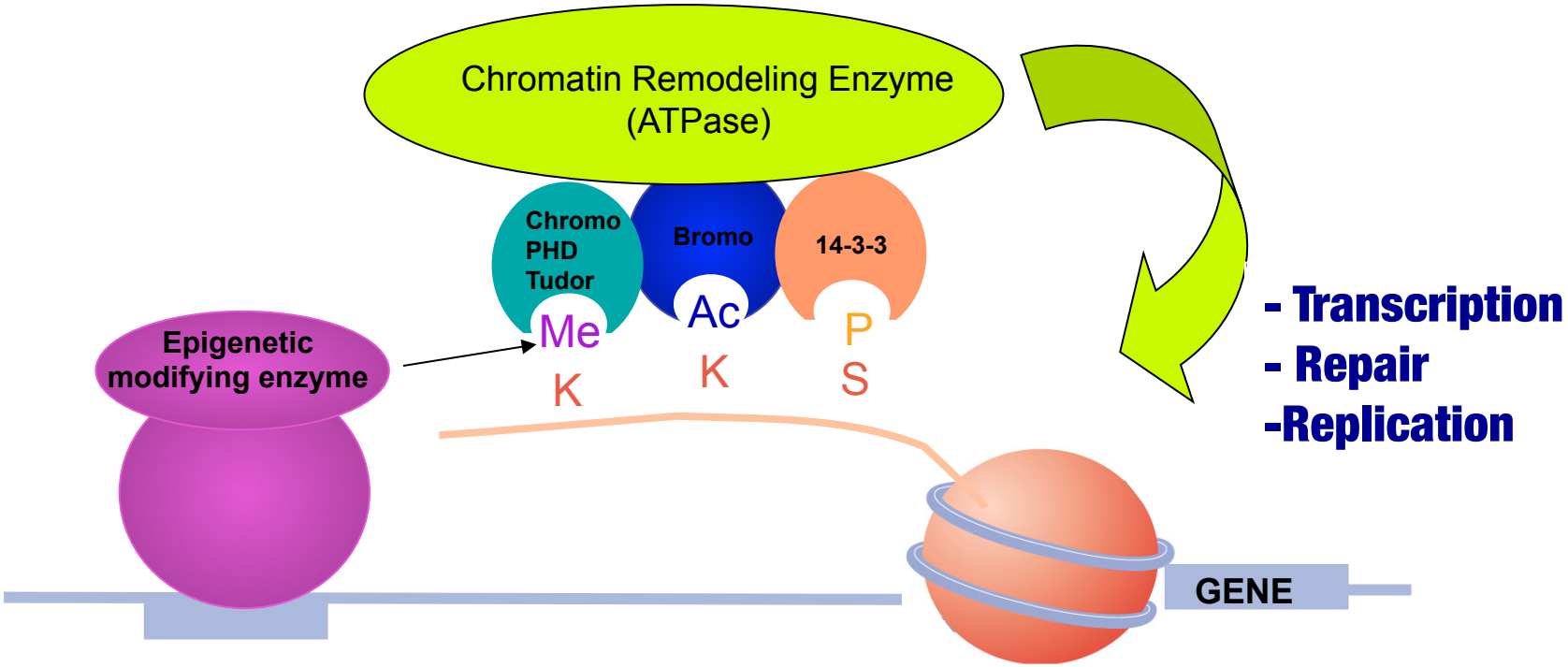
Repressed genes

Heterochromatin

Epigenetic writers, readers & erasers



Chromatin writers, readers & erasers



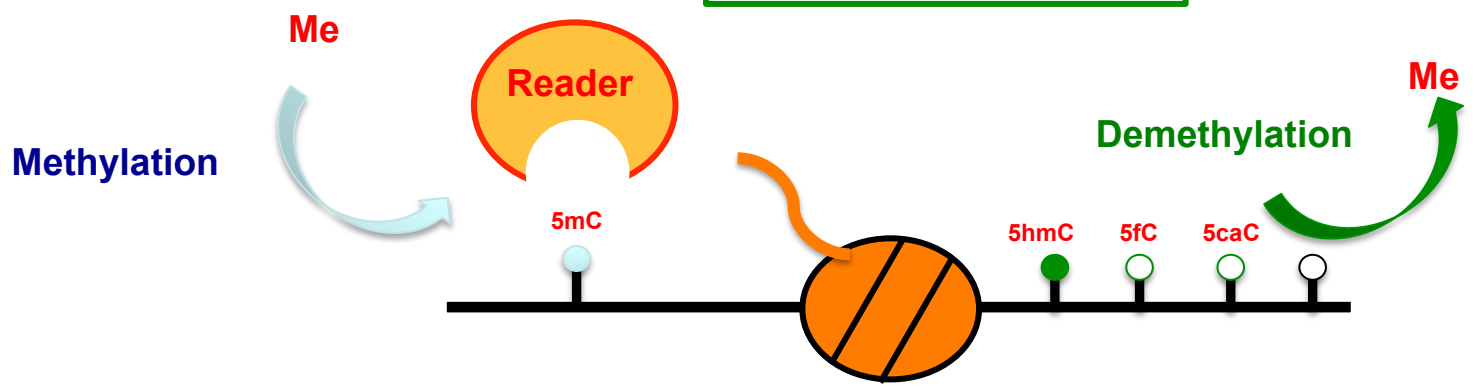
Cancer mutations: DNA methylation

Methyltransferase

ENZYME	MUTATION	TUMOUR
DNMT3A^	M, F, N, S	AML

Hydroxymethylation and derivatives

ENZYME	MUTATION	TUMOUR
TET1	T	AML
TET2	M, N, F	AML, MPD, MDS, CMML



^ = PWWP domain

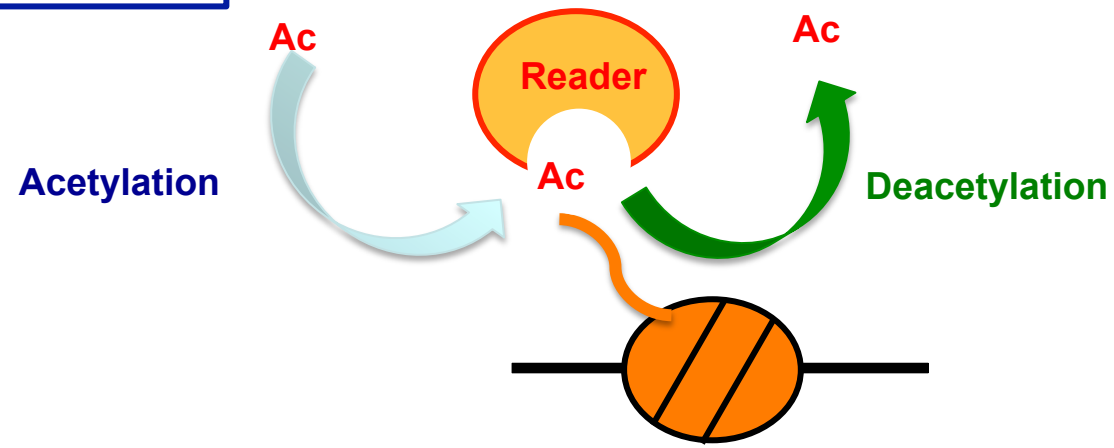
Cancer mutations: Histone Acetylation

Acetyltransferases

ENZYME	MUTATION	TUMOUR
KAT3A (CBP)*	T, N, F, M	AML, ALL, DLBCL, B-NHL,TCC
KAT3B (p300)*	T, N, F, M	AML, ALL, DLBCL, TCC, Colorectal, Breast, Pancreatic
KAT6A (MOZ)+	T	AML, MDS
KAT6B (MORF)	T	AML, Uterine leiomyoma

Readers

READER	MUTATION	TUMOUR
BROMODOMAIN		
BRD3	T	Midline Carcinoma
BRD4	T	Midline Carcinoma
TRIM33+	T	Papillary Thyroid
PBRM1	N, F, M, S, D	Renal, Breast



* = Bromodomain
+ = PHD Finger

Cancer mutations: Histone Methylation

Methyltransferases

ENZYME	MUTATION	TUMOUR
MLL1*+	T, PTD	AML, ALL, TCC
MLL2+	N, F, M	Medullobtastoma, Renal, DLBCL, FL
MLL3+	N	Medullobtastoma, TCC
SETD2	N, F, S, M	Renal
NSD1+^	T	AML
NSD2+^	T	Multiple myeloma
NSD3^	T	AML
EZH2	M	DLBCL, MPD,MDS

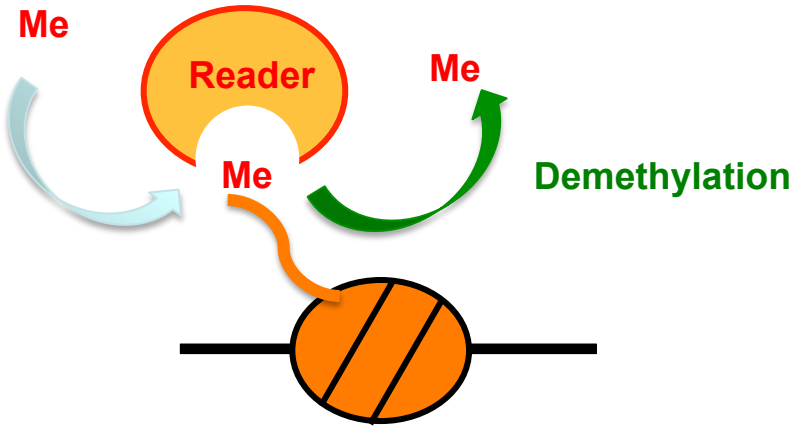
Readers

READER	MUTATION	TUMOUR
PHD		
TRIM33*	T	Papillary Thyroid
ING1	M, D	Melanoma, Breast
ING4	D	HNC
PWWP		
MSH6	M, N, F, S	Colorectal

Demethylases

ENZYME	MUTATION	TUMOUR
KDM5A (JARID1A)+	T	AML
KDM5C (JARID1C)+	N, F, S	Renal
KDM6A (UTX)	D, N, F, S	AML, TCC, Renal, Oesophageal, Multiple myeloma

Methylation



* = Bromodomain
+ = PHD Finger
^ = PWWP domain

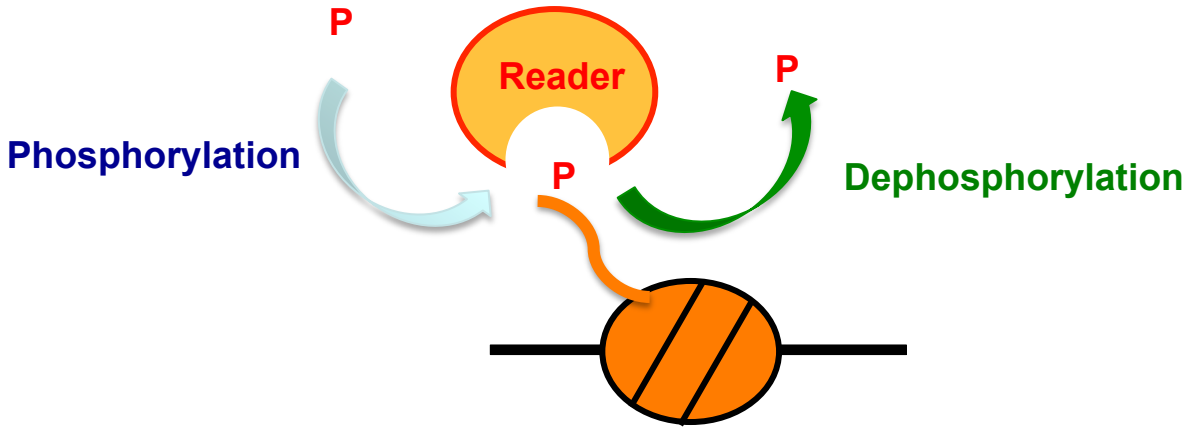
Cancer mutations: Histone phosphorylation

Kinase

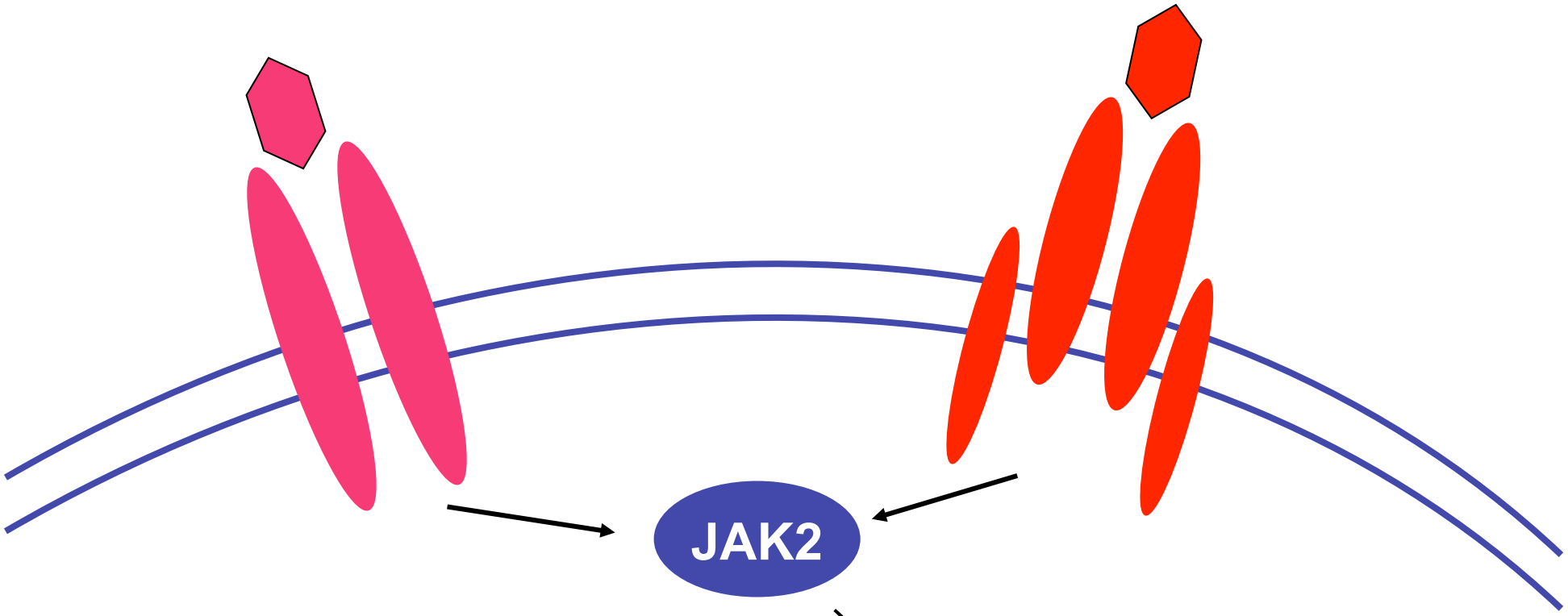
ENZYME	MUTATION	TUMOUR
JAK2	T, M	AML, ALL, MPD, CML
PIM1	T	NHL

Readers

READER	MUTATION	TUMOUR
BRCT		
BRCA1	D, M, N, F, S	Ovarian, Breast, Prostate



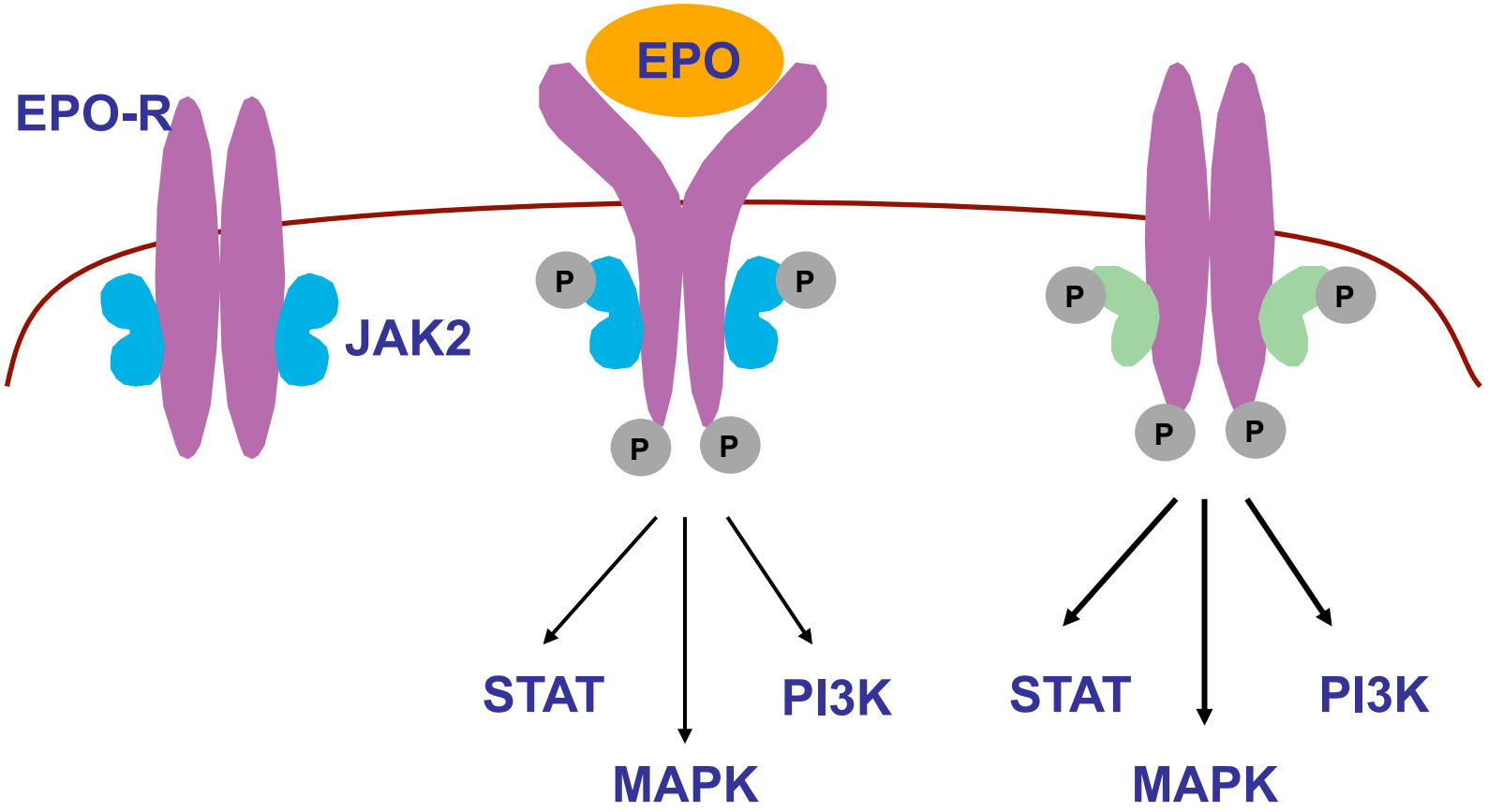
Identification of a unique mutation in JAK2



James et al Nature 2005
Baxter et al Lancet 2005
Levine et al Cancer Cell 2005
Kralovics et al NEJM 2005

Proliferation
Differentiation
Survival

Consequence of the JAK2 mutations



Campbell & Green; NEJM 2006

A JAK - chromatin link

JAK signaling globally counteracts heterochromatic gene silencing

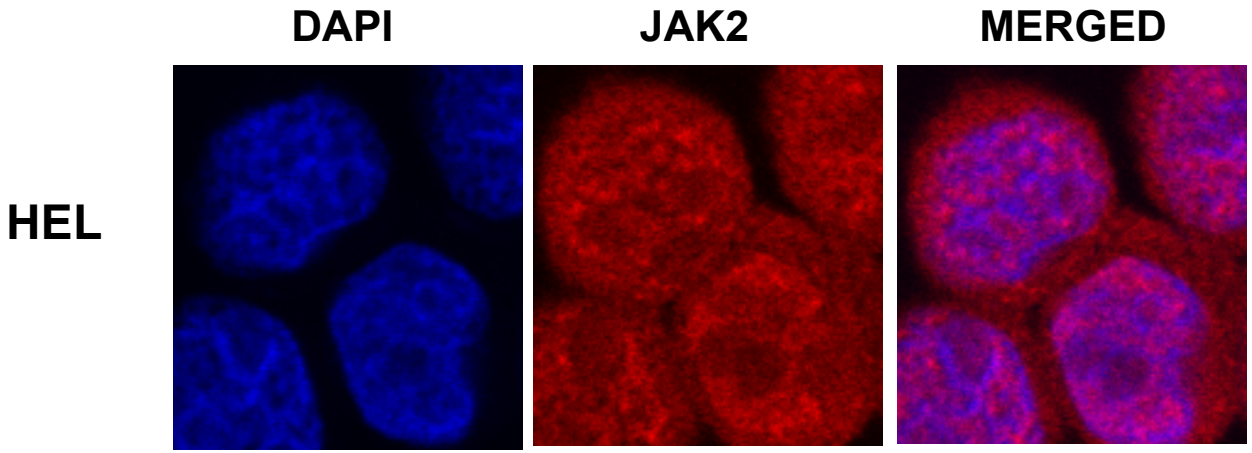
Song Shi, Healani C Calhoun, Fan Xia, Jinghong Li, Long Le & Willis X Li

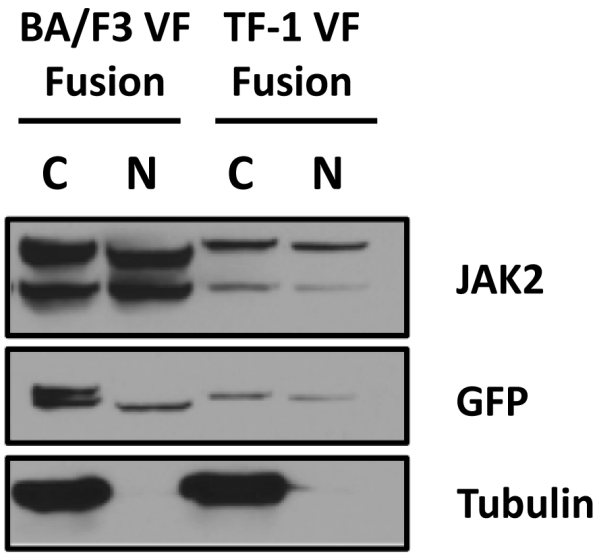
nature genetics

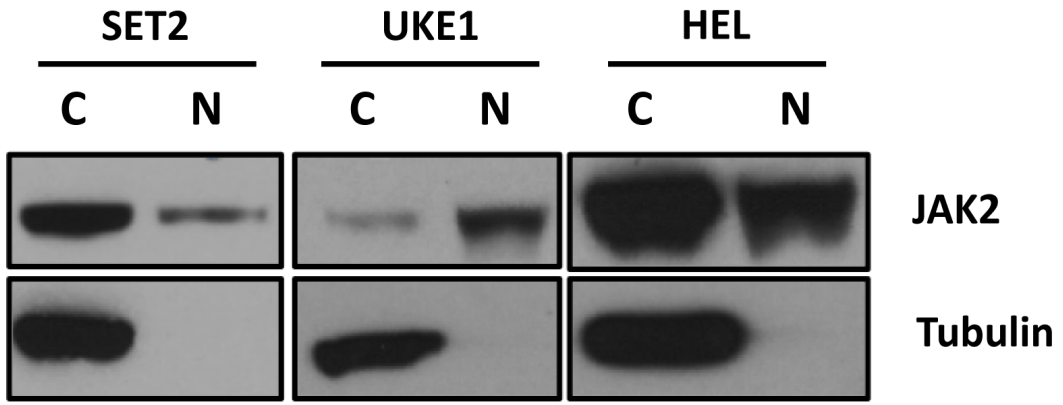
Heterochromatin associated proteins

Mutant JAK →

JAK2 is present in the nucleus of myeloid cells

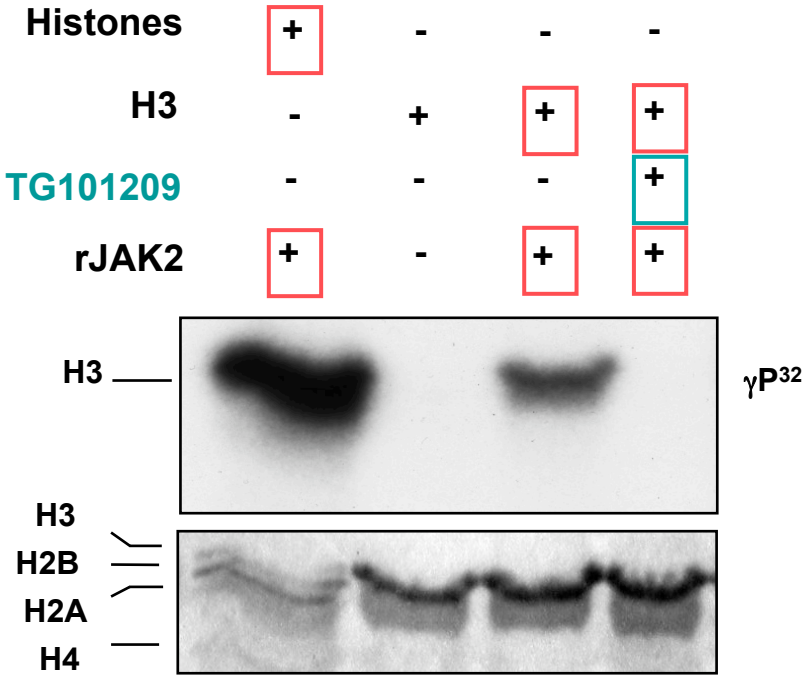


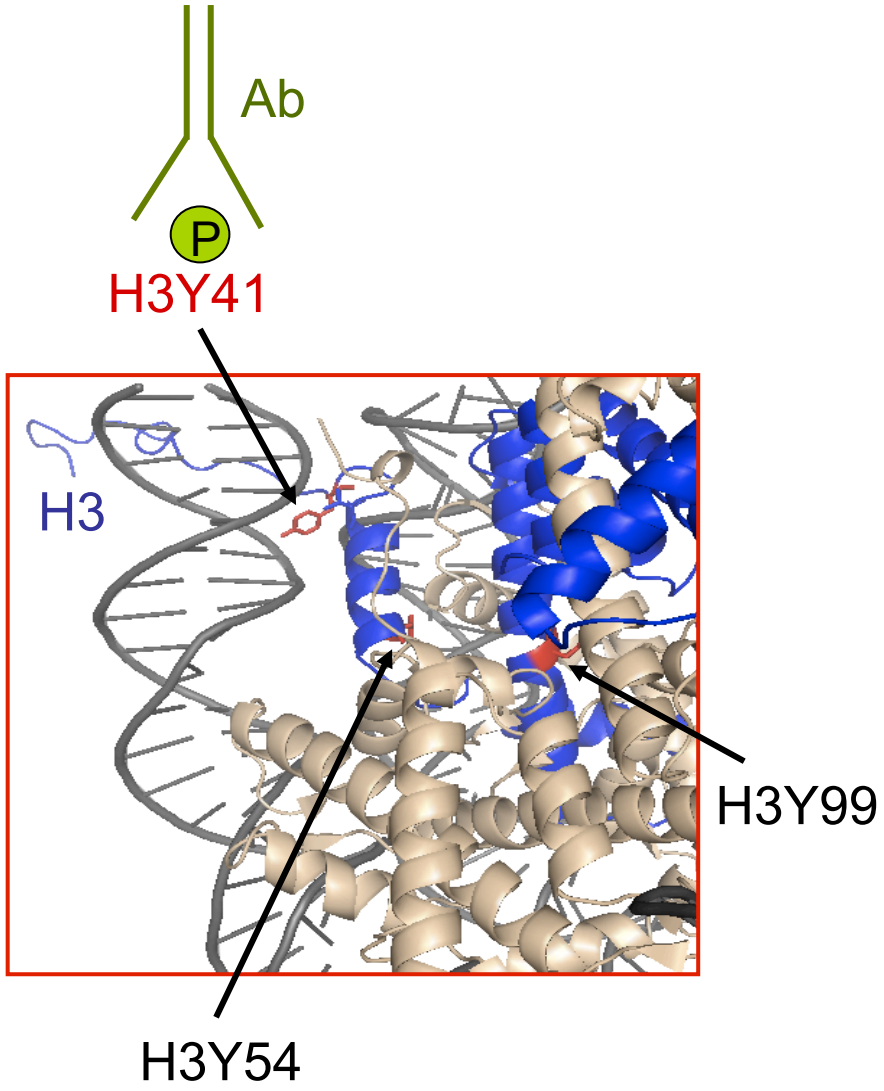




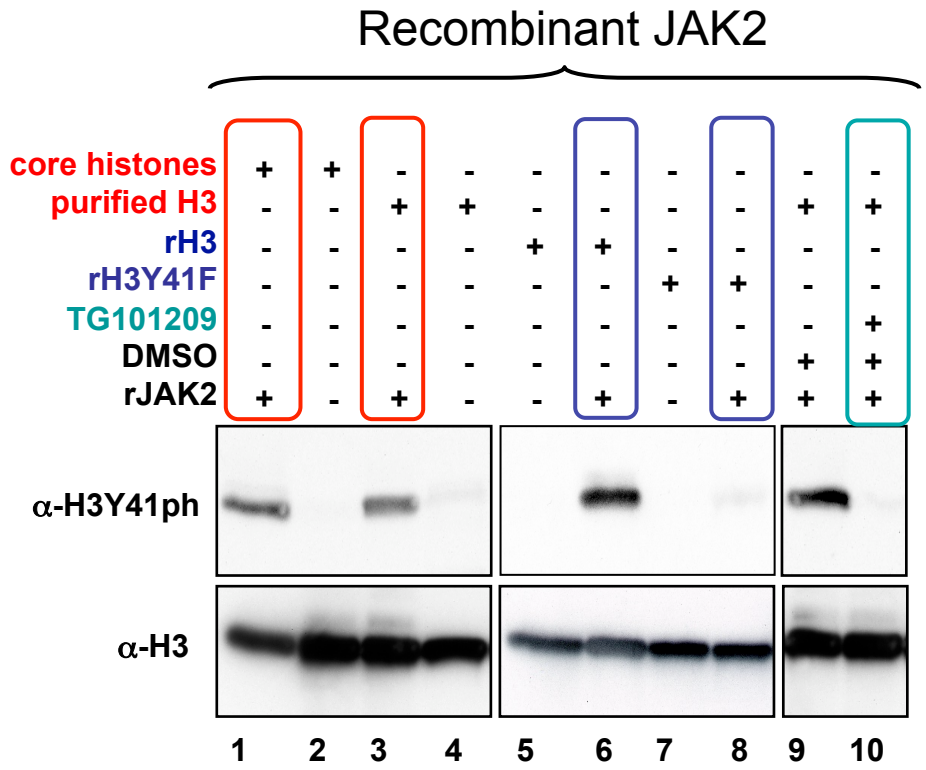
Dawson et al - Nature 2009
Dawson et al – Blood 2011

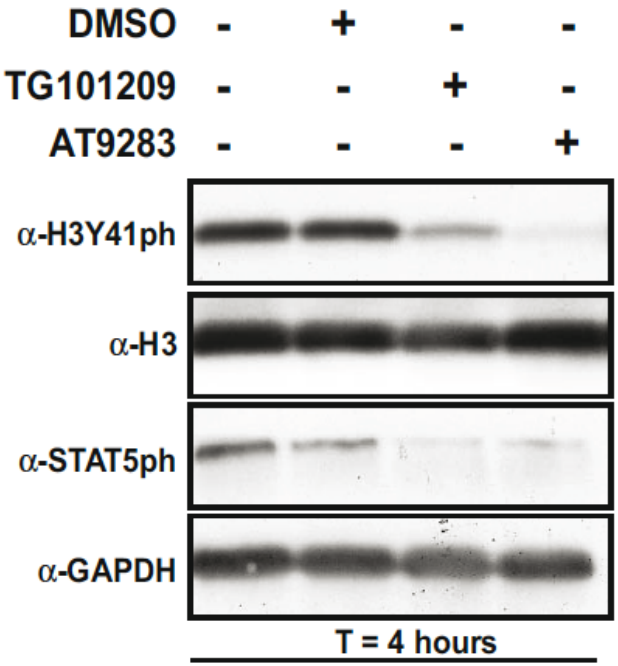
JAK2 phosphorylates histone H3



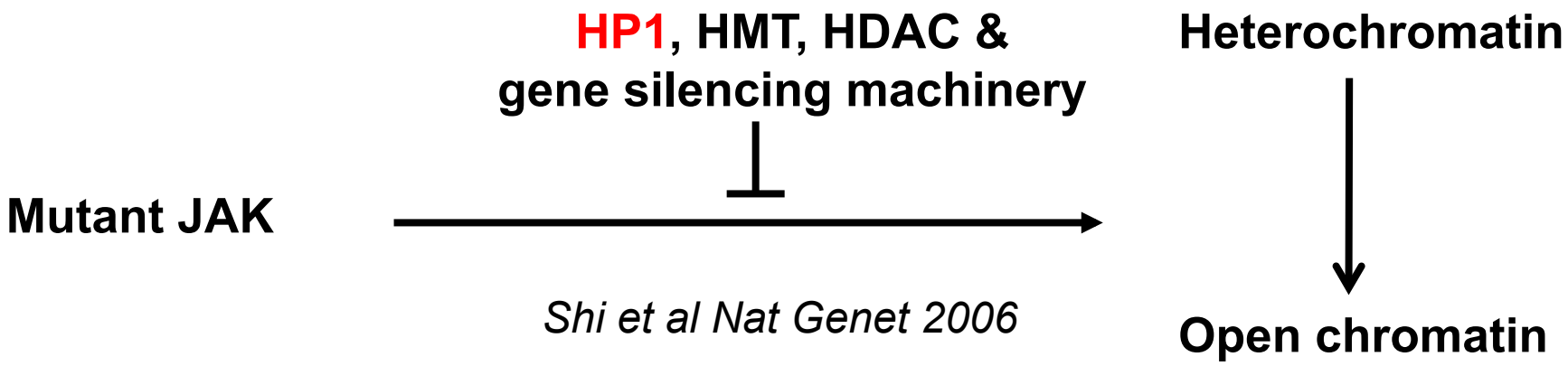


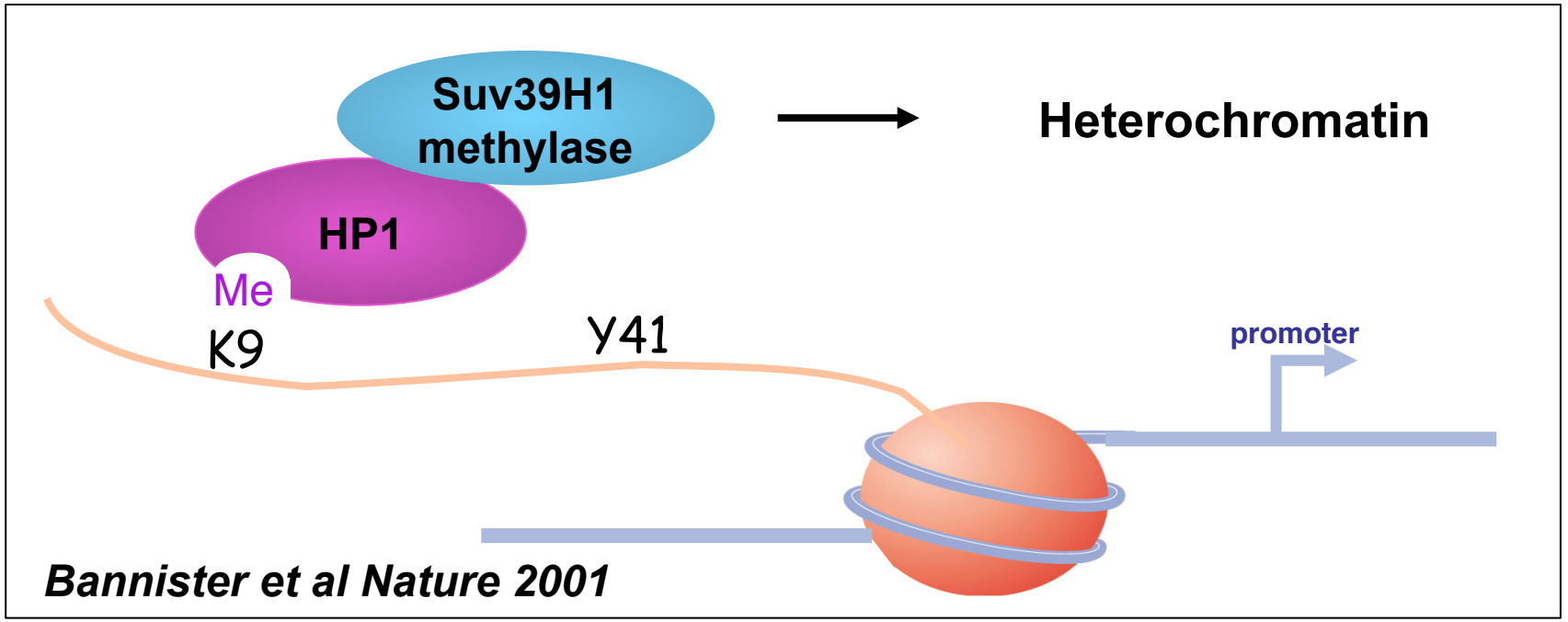
JAK2 phosphorylates H3Y41 directly in vitro & in vivo



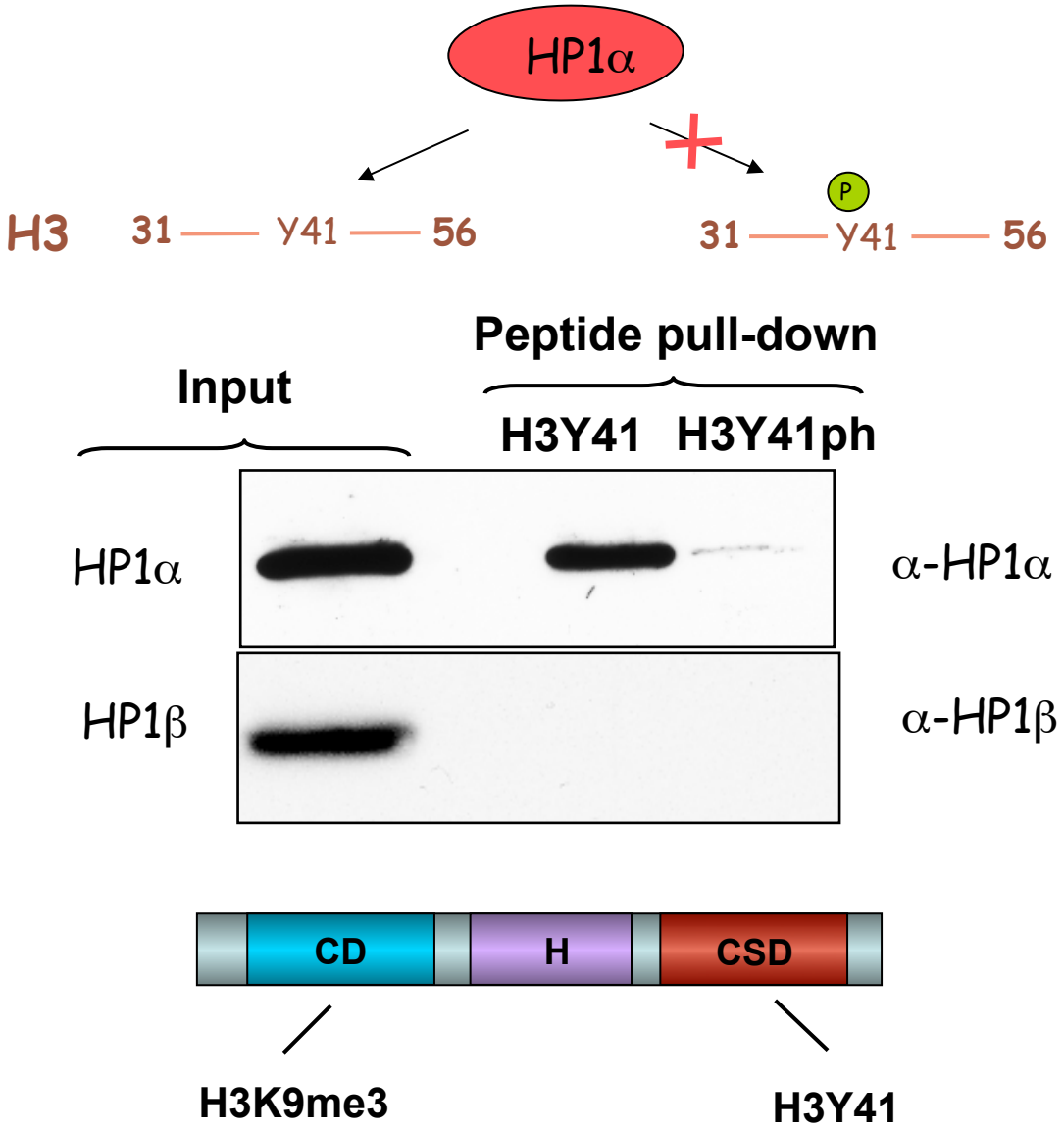


What is the function of H3Y41ph ?

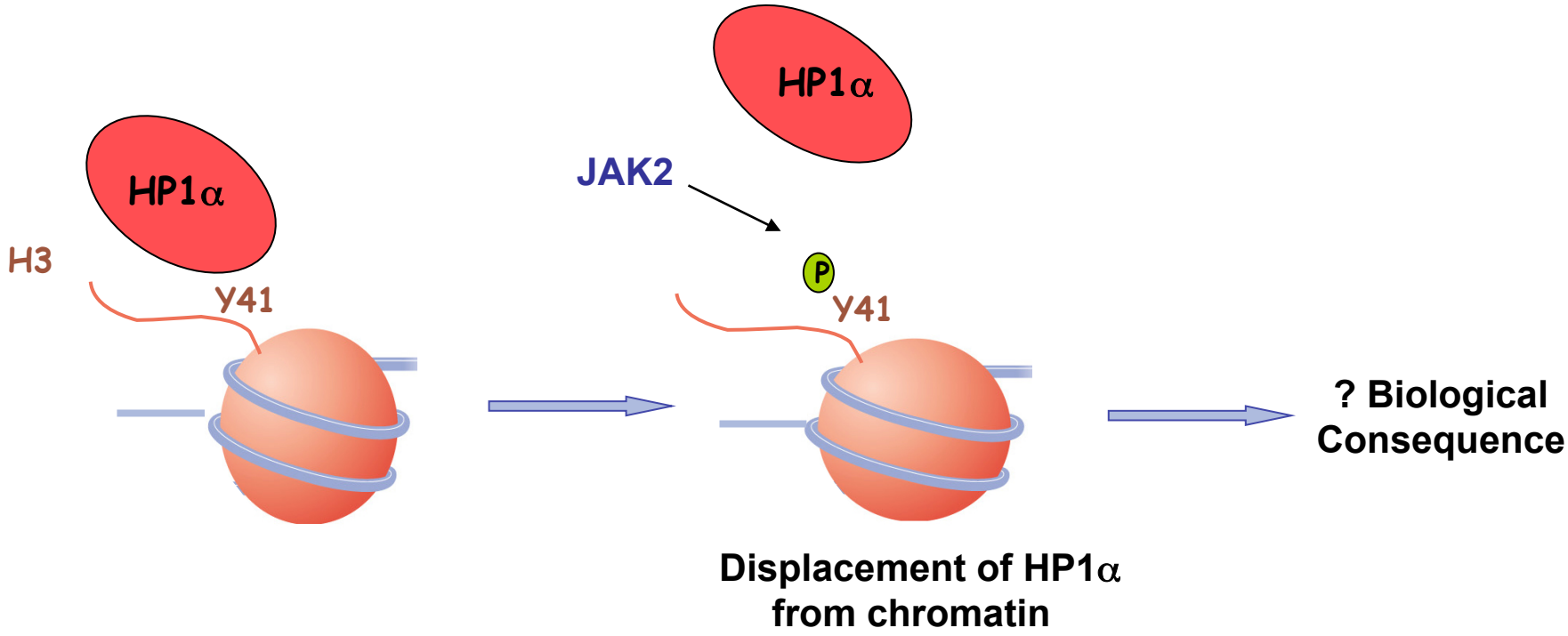




HP1α binds H3 and is disrupted by H3Y41ph



JAK2 signaling to chromatin



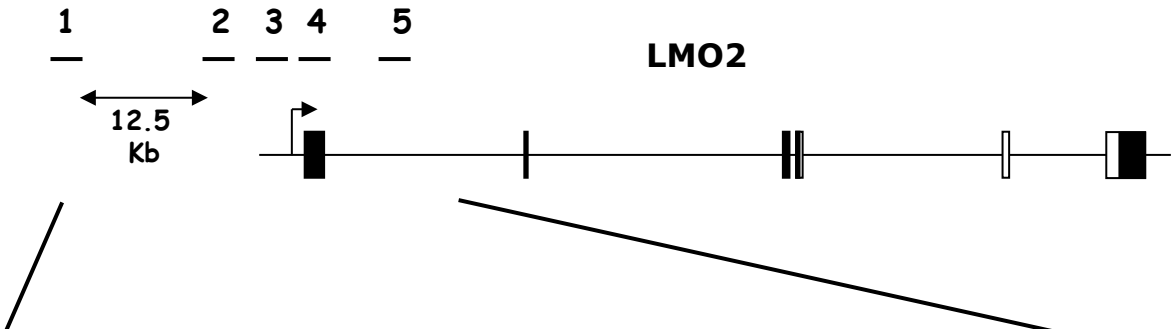
The LMO2 oncogene is an unexpected target of JAK2 signaling

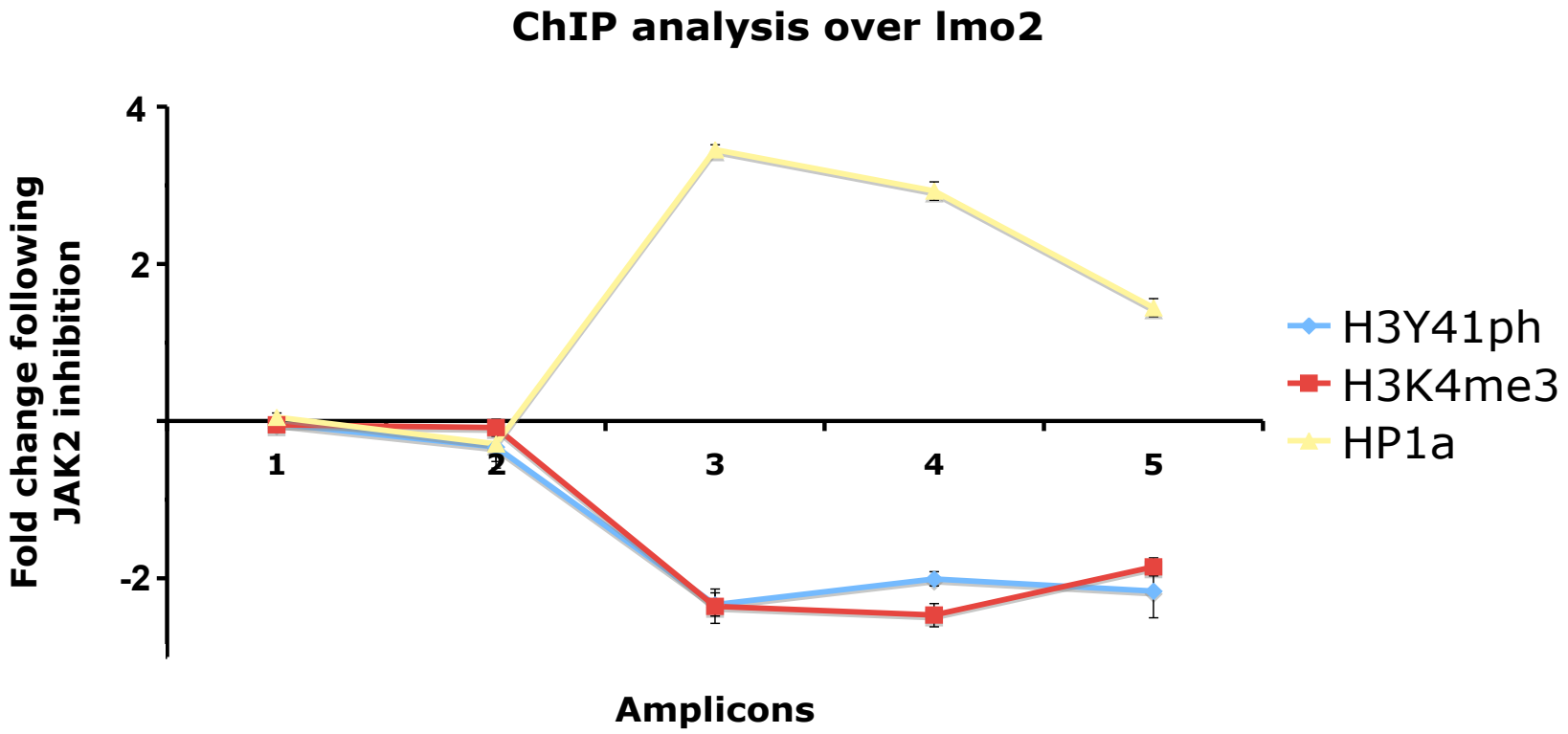
Most Down-regulated Genes of 18,164 tiled transcripts

Gene ID	LOG2 Fold Change	# STAT5 Sites
STS-1	-2.12	1
ID1	-2.11	1
IGFBP5	-1.93	2
FLJ11795	-1.92	4
PIM1	-1.87	1
HSPA5	-1.70	3
LOC317671	-1.65	1
DARC	-1.59	1
PIM2	-1.58	2
HSPC111	-1.56	0
TUBAL3	-1.51	1
PLVAP	-1.49	1
BCL2L1	-1.46	3
GDF3	-1.46	1
RAB3IL1	-1.45	3
HMBS	-1.43	0
SDF2L1	-1.41	1
SLCO4A1	-1.41	0
NME1	-1.39	0
KCNH2	-1.39	0
PCOLCE2	-1.35	1
HBBP1	-1.33	1
ISG20L1	-1.33	0
PSKH2	-1.32	0
LOC201164	-1.31	0
LMO2	-1.30	0
NOLA1	-1.29	1
GPR56	-1.29	1
C1ORF33	-1.28	0
EGR1	-1.27	1
FLJ43339	-1.27	3
C1ORF186	-1.25	2
RRS1	-1.24	1
CCDC58	-1.24	0
RGS19	-1.23	2
XTP3TPA	-1.22	0
TMC6	-1.21	1
SLA2	-1.18	0
KCNN4	-1.18	1
AGTRL1	-1.17	0

- Integrated genome wide expression array using HEL +/- JAK2 inhibitor
- computational detection of STAT5 sites
- 9 of top 30 genes down-regulated by inhibitor lacked STAT5 sites
- Includes Lmo2
  - essential for haematopoiesis
  - known leukaemia oncogene

JAK2 signaling directly to chromatin alters the expression of LMO2



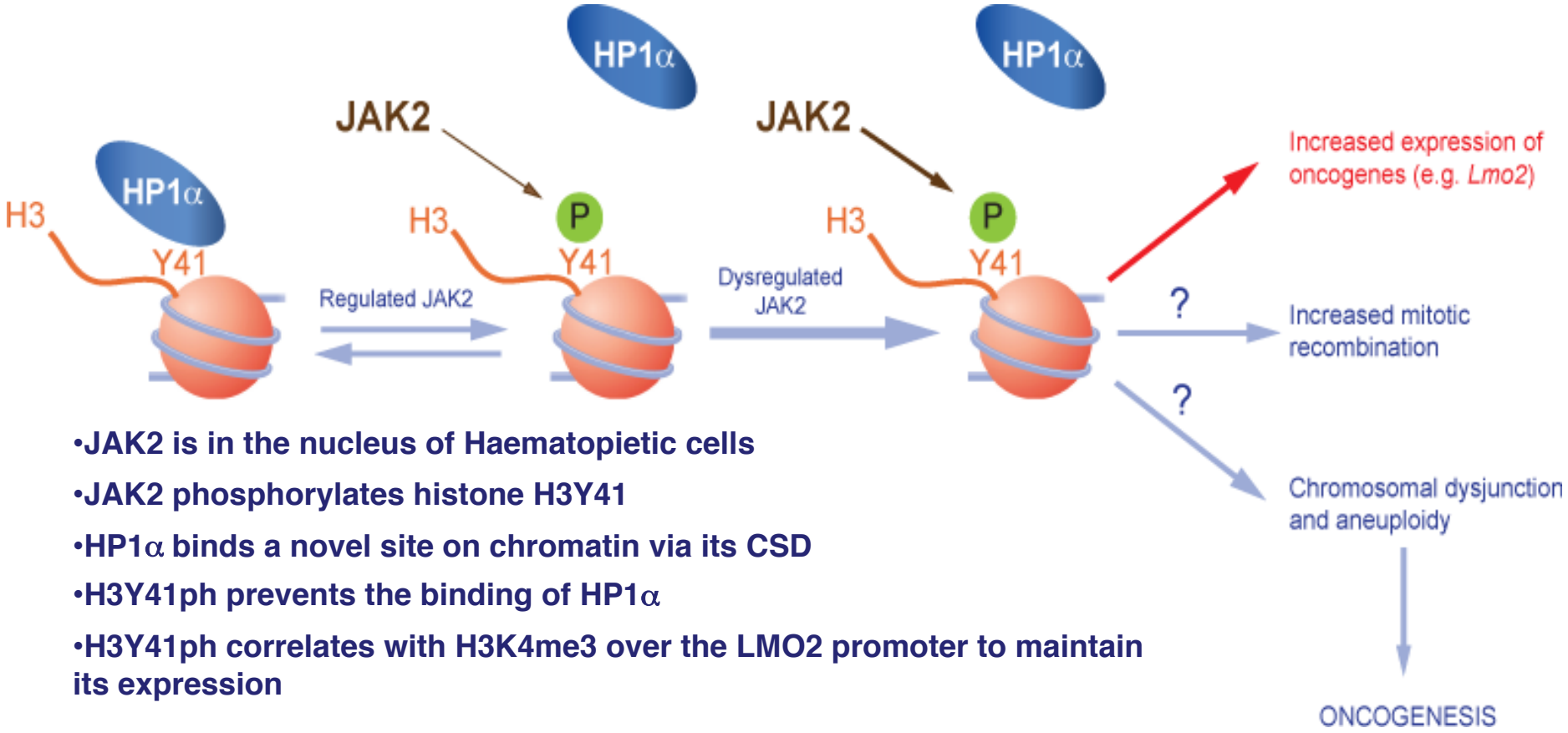


JAK2 at chromatin

Vol 461|8 October 2009|doi:10.1038/nature08448 nature

JAK2 phosphorylates histone H3Y41 and excludes HP1α from chromatin

Mark A. Dawson[1,2]*, Andrew J. Bannister[3]*, Berthold Göttgens[1], Samuel D. Foster[1], Till Bartke[3], Anthony R. Green[1,2]* & Tony Kouzarides[3]*



- JAK2 is in the nucleus of Haematopietic cells
- JAK2 phosphorylates histone H3Y41
- HP1α binds a novel site on chromatin via its CSD
- H3Y41ph prevents the binding of HP1α
- H3Y41ph correlates with H3K4me3 over the LMO2 promoter to maintain its expression

JAK2 at chromatin

nature cell biology

ARTICLES

LIF-independent JAK signalling to chromatin in embryonic stem cells uncovered from an adult stem cell disease

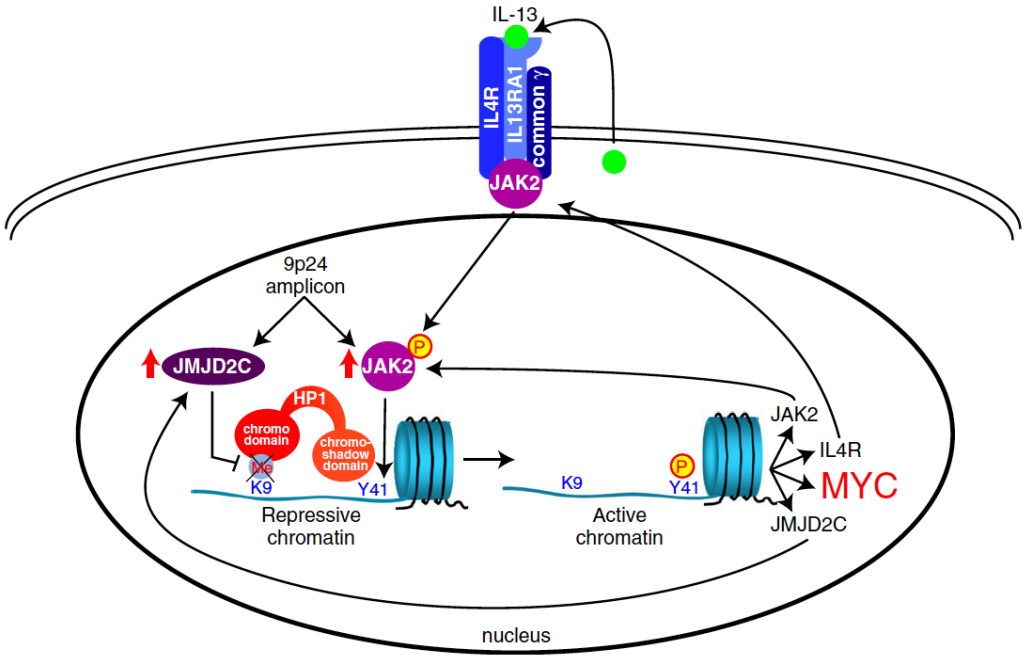
Dean S. Griffiths[1,7], Juan Li[1], Mark A. Dawson[1,2,6], Matthew W. B. Trotter[3], Yi-Han Cheng[1], Aileen M. Smith[1], William Mansfield[4], Pentao Liu[5], Tony Kouzarides[6], Jennifer Nichols[4], Andrew J. Bannister[6], Anthony R. Green[1,2] and Berthold Göttgens[1,7]

JAK2 at chromatin

Cancer Cell
Article

Cooperative Epigenetic Modulation by Cancer Amplicon Genes

Lixin Rui,[1] N.C. Tolga Emre,[1] Michael J. Kruhlak,[2] Hye-Jung Chung,[3] Christian Steidl,[4] Graham Slack,[4] George W. Wright,[5] Georg Lenz,[1] Vu N. Ngo,[1] Arthur L. Shaffer,[1] Weihong Xu,[1] Hong Zhao,[1] Yandan Yang,[1] Laurence Lamy,[1] R. Eric Davis,[1] Wenming Xiao,[6] John Powell,[6] David Maloney,[7] Craig J. Thomas,[7] Peter Möller,[8] Andreas Rosenwald,[9] German Ott,[10] Hans Konrad Muller-Hermelink,[9] Kerry Savage,[4] Joseph M. Connors,[4] Lisa M. Rimsza,[11,12] Elias Campo,[13] Elaine S. Jaffe,[3] Jan Delabie,[14] Erlend B. Smeland,[15,16] Dennis D. Weisenburger,[17] Wing C. Chan,[17] Randy D. Gascoyne,[4] David Levens,[3] and Louis M. Staudt[1,*]



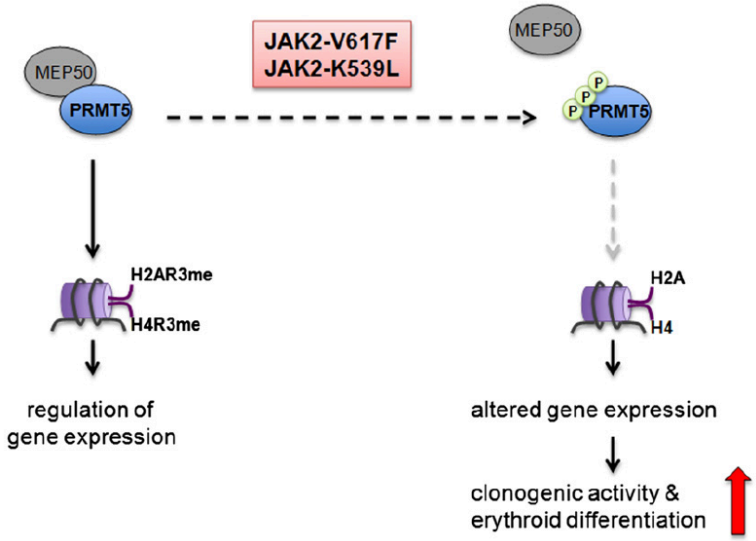
JAK2 at chromatin

Cell PRESS

Cancer Cell
Article

JAK2V617F-Mediated Phosphorylation of PRMT5 Downregulates Its Methyltransferase Activity and Promotes Myeloproliferation

Fan Liu,[1,4] Xinyang Zhao,[1,4] Fabiana Perna,[1] Lan Wang,[1] Priya Koppikar,[2] Omar Abdel-Wahab,[2] Michael W. Harr,[1] Ross L. Levine,[2] Hao Xu,[1] Ayalew Tefferi,[3] Anthony Deblasio,[1] Megan Hatlen,[1] Silvia Menendez,[1] and Stephen D. Nimer[1,*]



Targeting chromatin readers

ARTICLE

doi:10.1038/nature09504

Selective inhibition of BET bromodomains

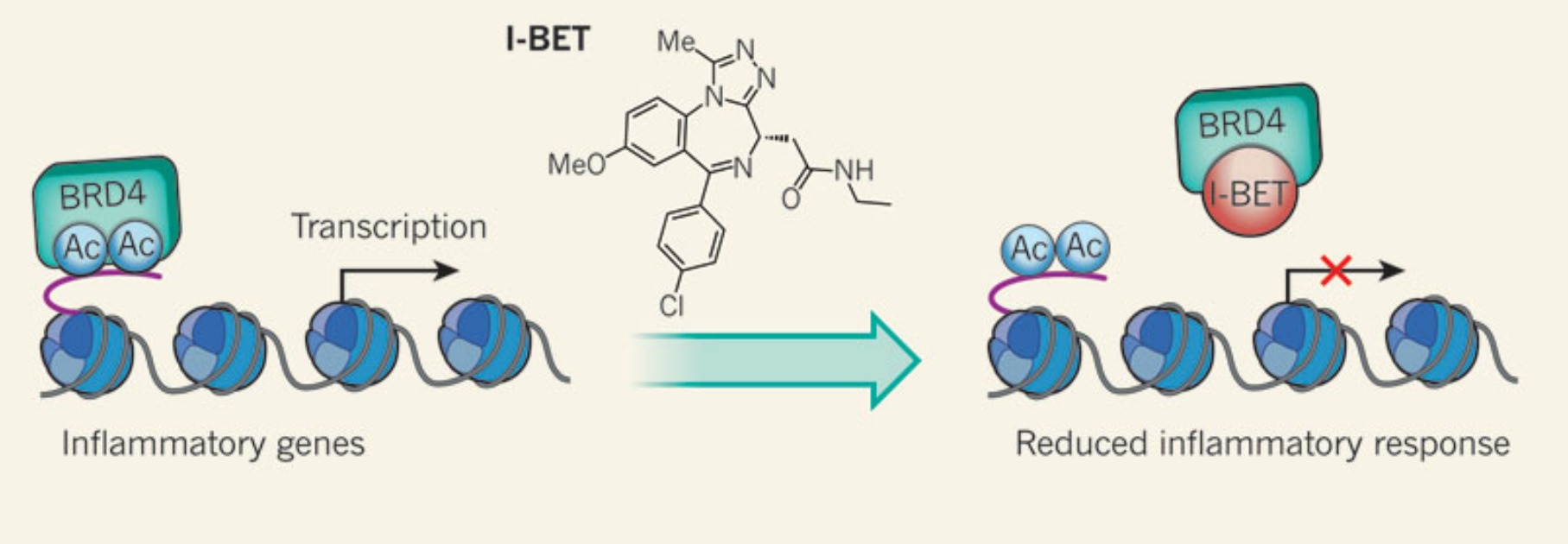
Panagis Filippakopoulos[1]*, Jun Qi[2]*, S
Tracey Keates[1], Tyler T. Hickman[4], Il
Amanda L. Christie[8], Nathan West[2], M
Christopher A. French[4], Olaf Wiest[3],

LETTER

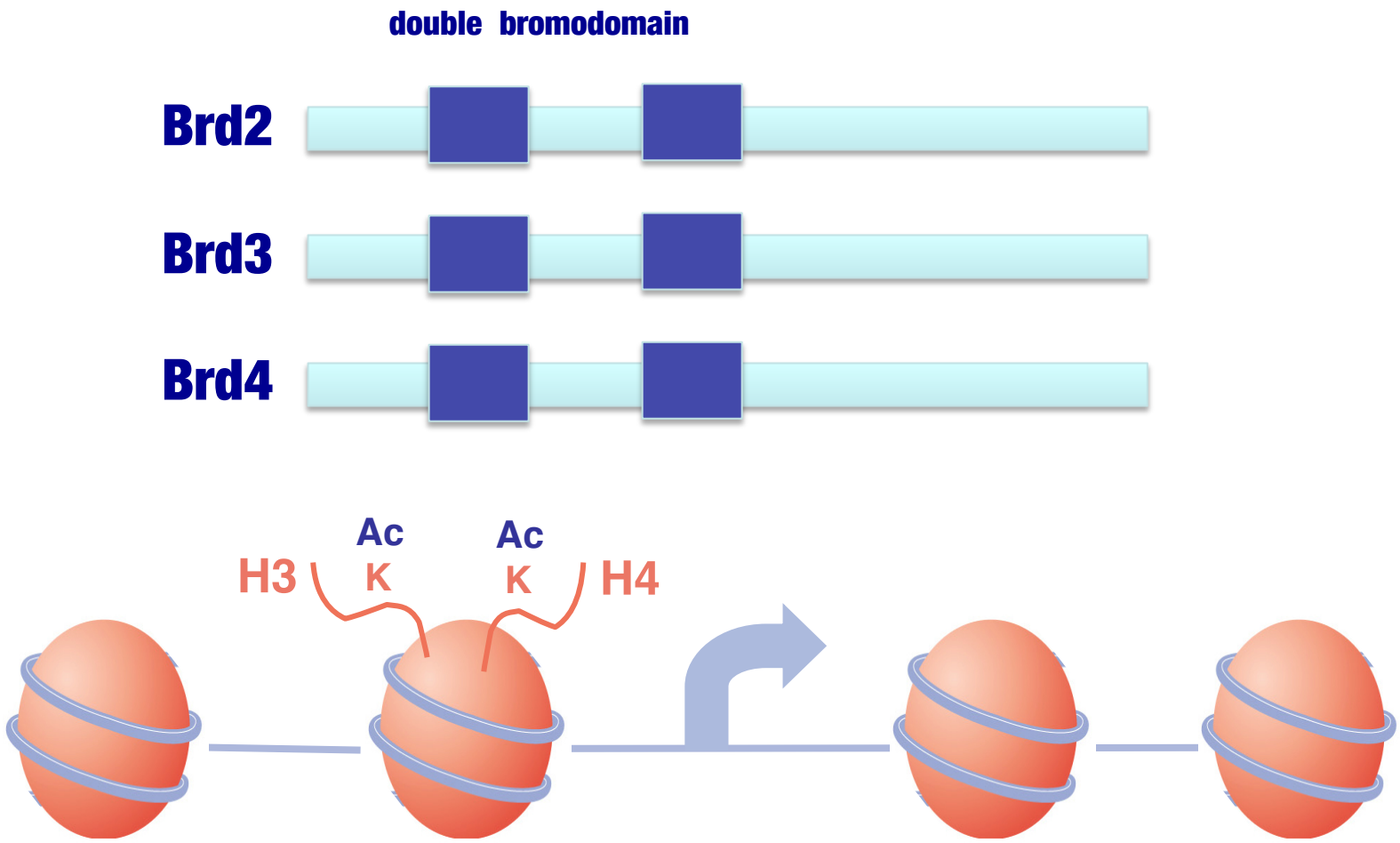
doi:10.1038/nature09589

Suppression of inflammation by a synthetic histone mimic

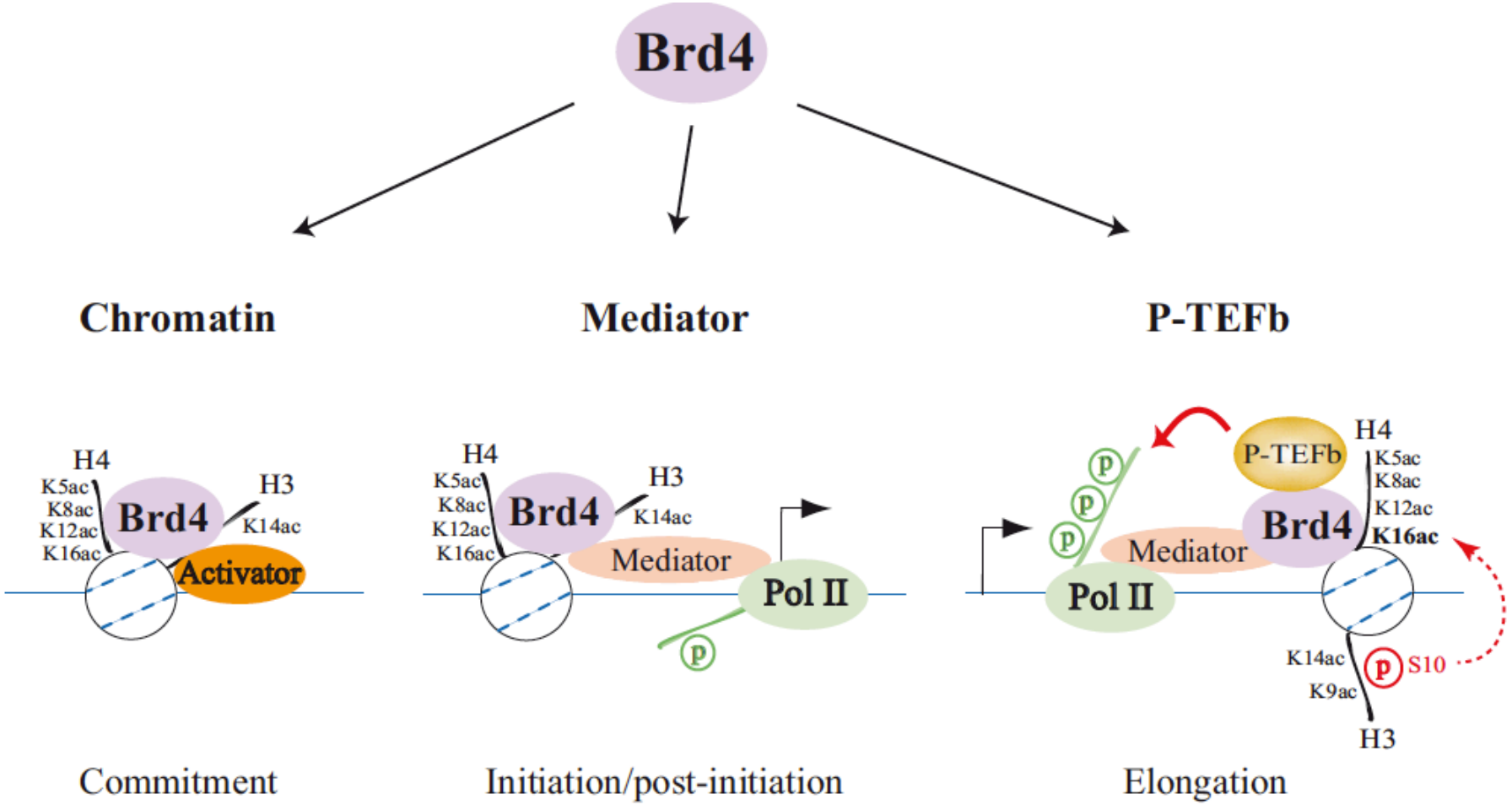
Edwige Nicodeme[1]*, Kate L. Jeffrey[2]*, Uwe Schaefer[2]*, Soren Beinke[3]*, Scott Dewell[4], Chun-wa Chung[5], Rohit Chandwani[2], Ivan Marazzi[2], Paul Wilson[5], Hervé Coste[1], Julia White[5], Jorge Kirilovsky[1], Charles M. Rice[6], Jose M. Lora[5], Rab K. Prinjha[3], Kevin Lee[3] & Alexander Tarakhovsky[2]



The BET domain proteins

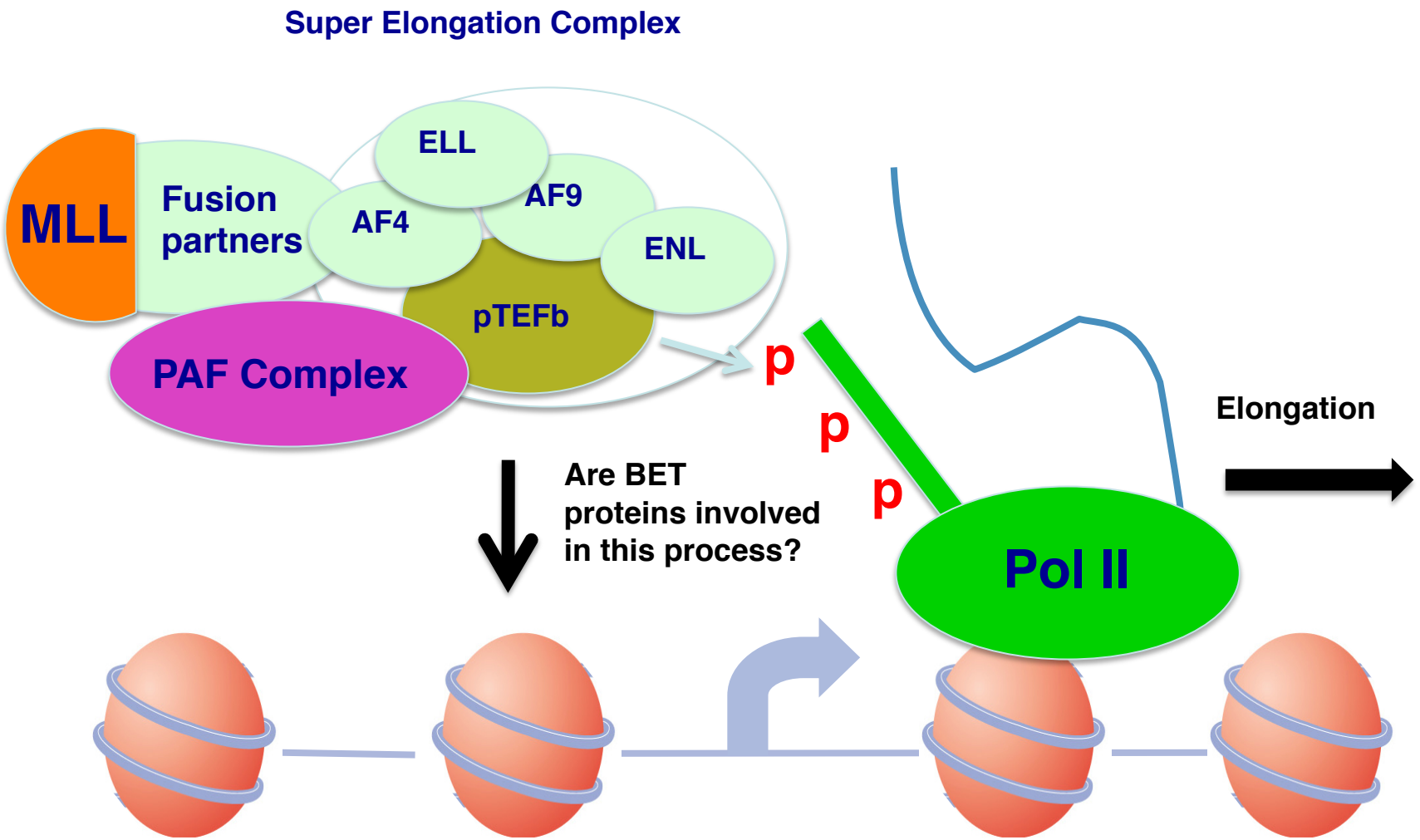


The BET domain proteins



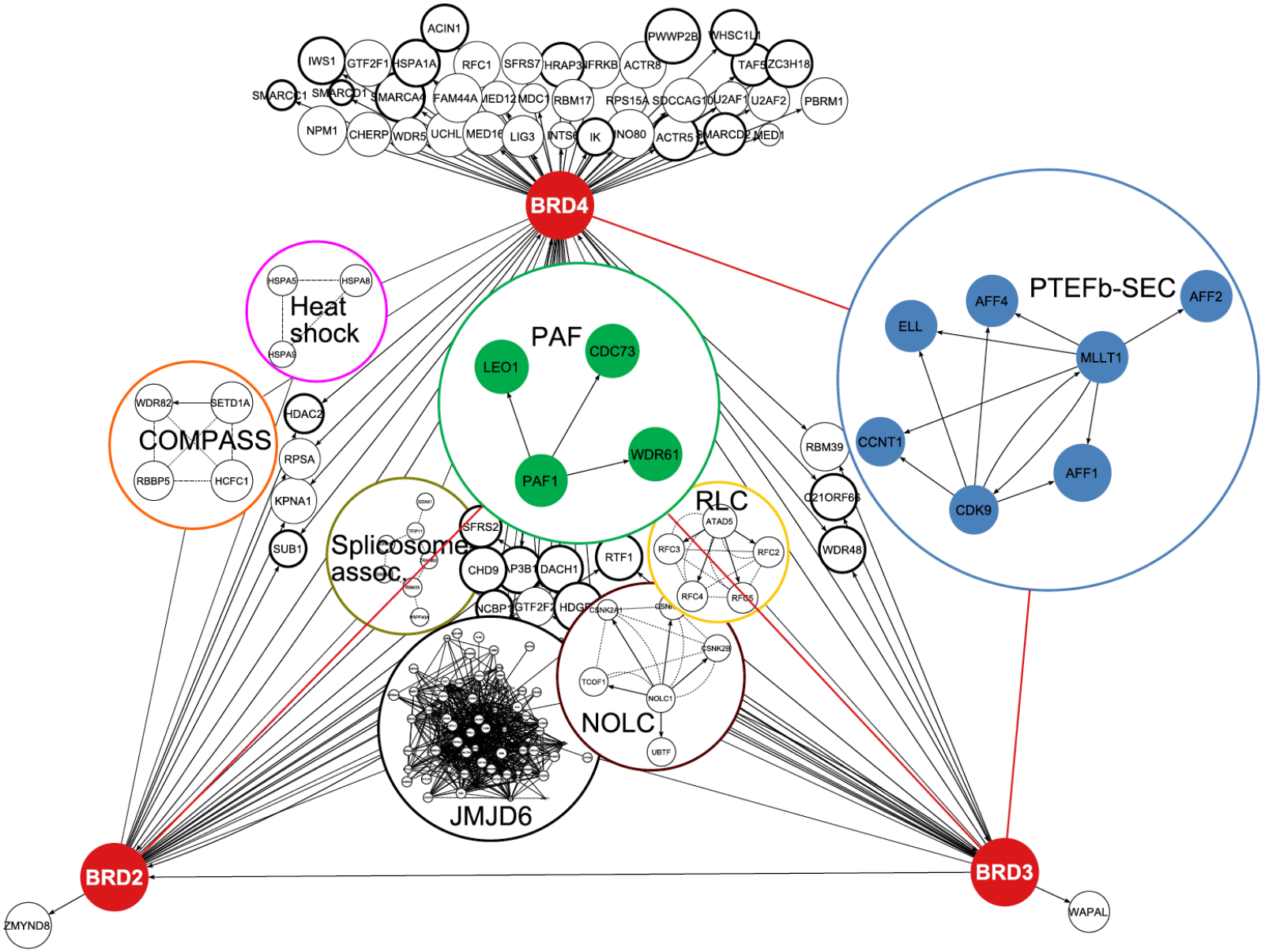
Chiang; F1000 Biol Reports; 2009

The pathogenesis of MLL Leukaemia

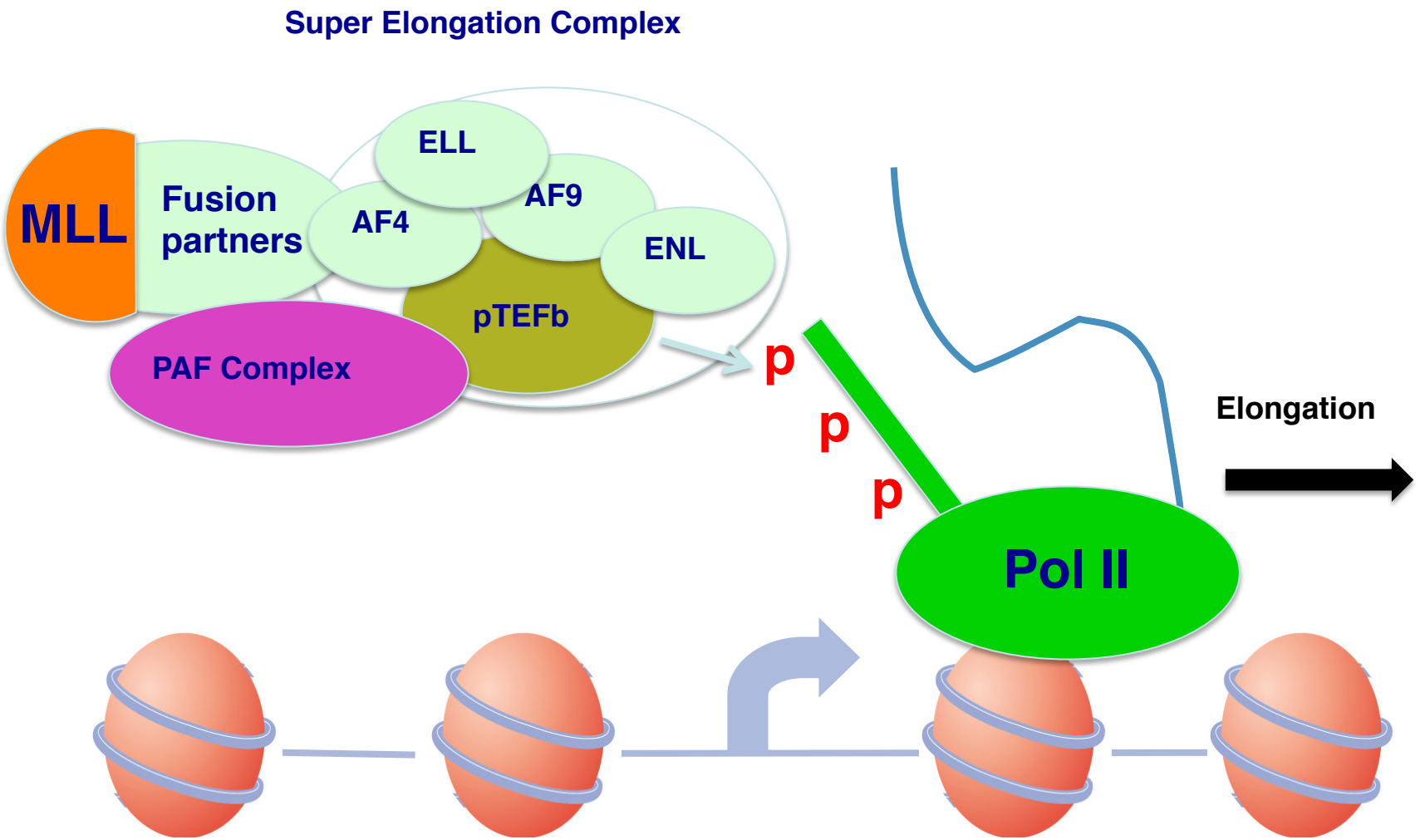


Global Proteomic characterization of BET complexes

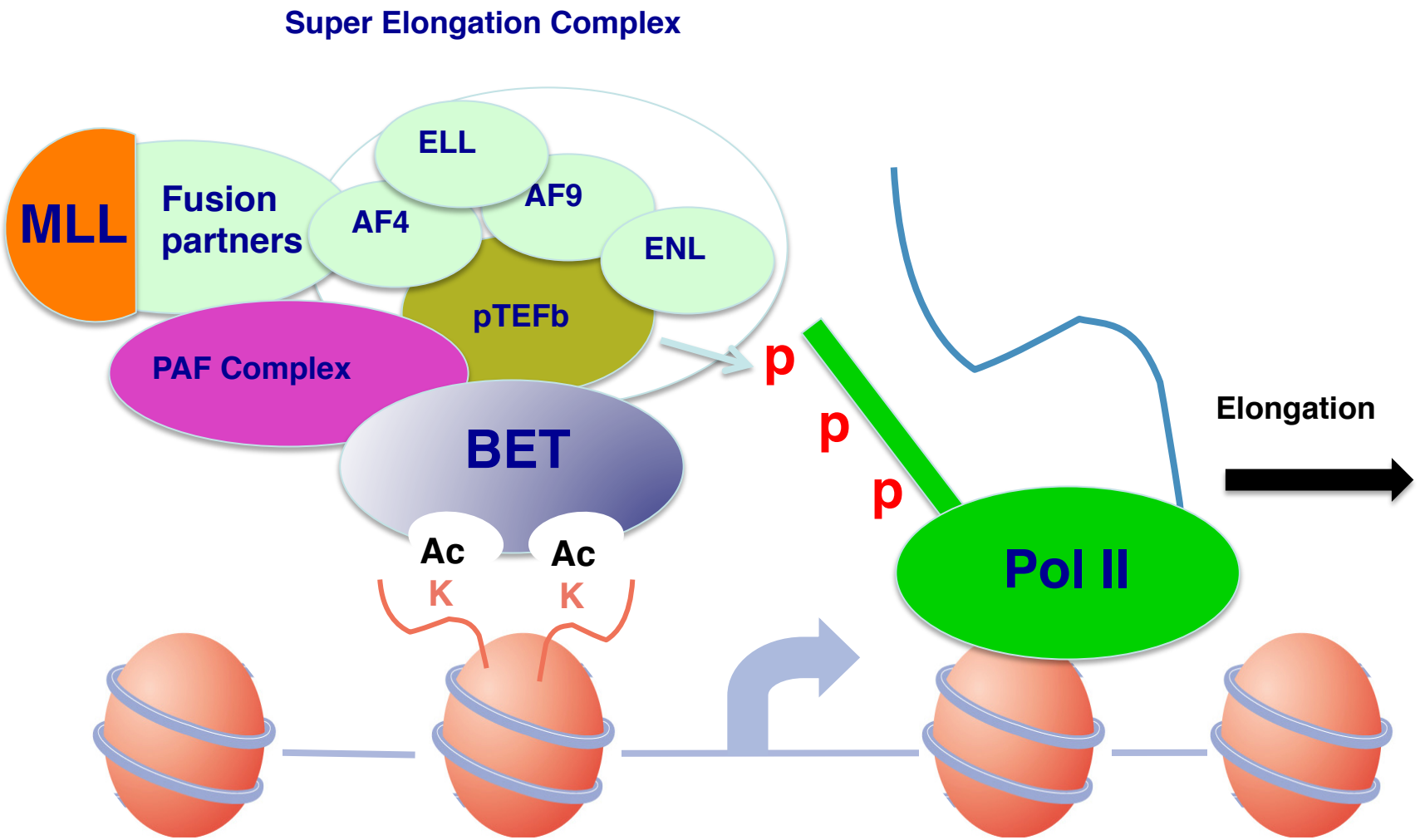
Global Proteomic characterization of BET complexes



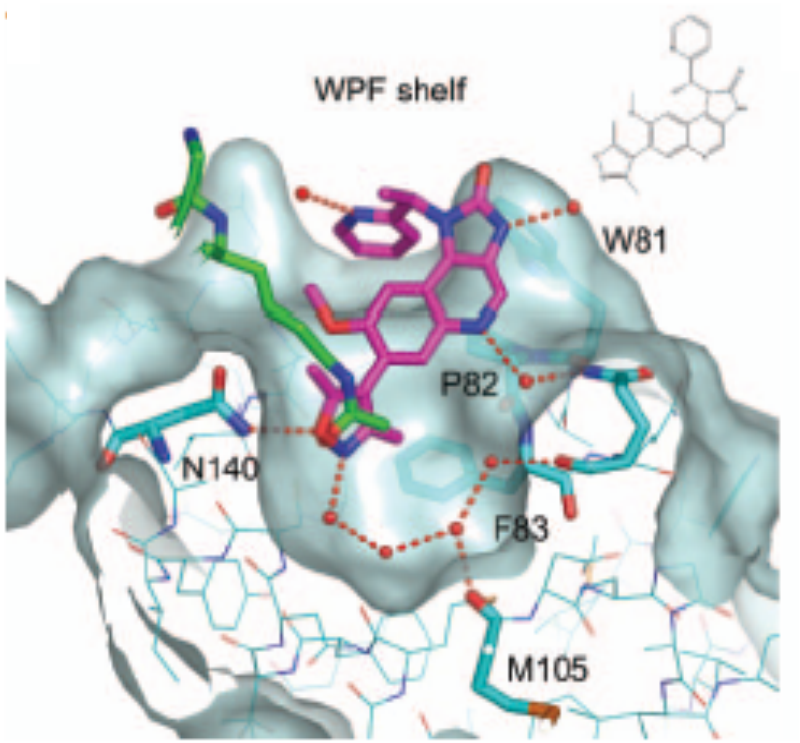
MLL-fusion partners are in transcriptional elongation complexes

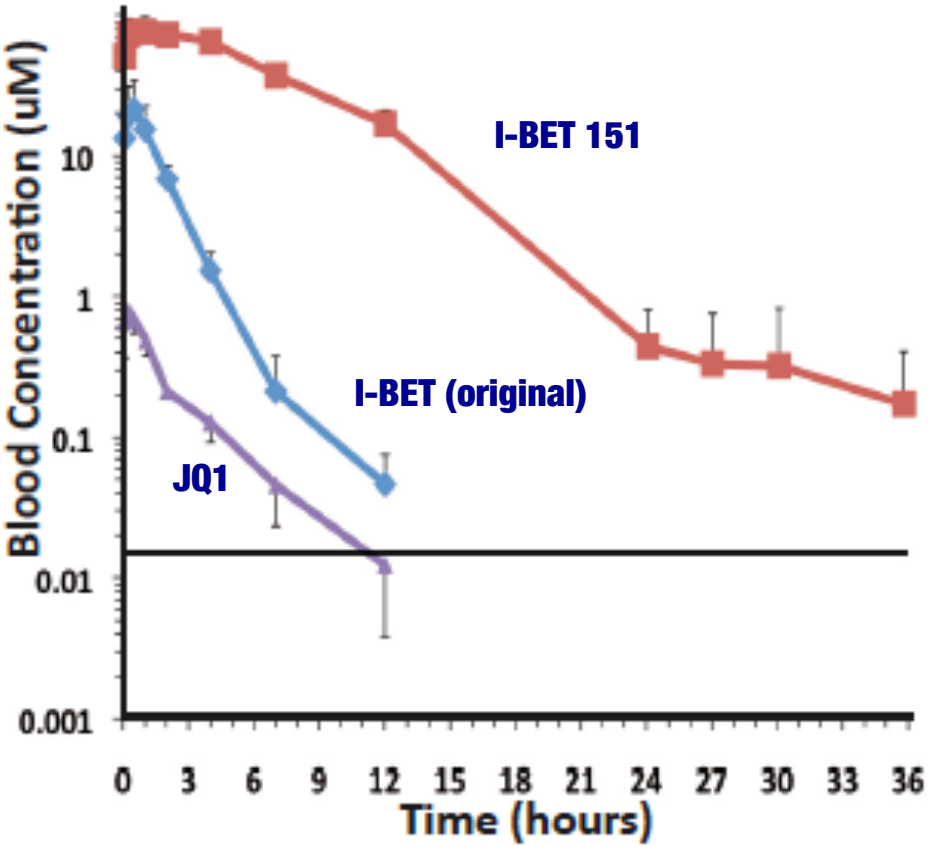


MLL-fusion partners are in transcriptional elongation complexes



Developing a novel and more potent BET-Inhibitor I-BET151





I-BET 151 has selectivity for MLL leukaemias

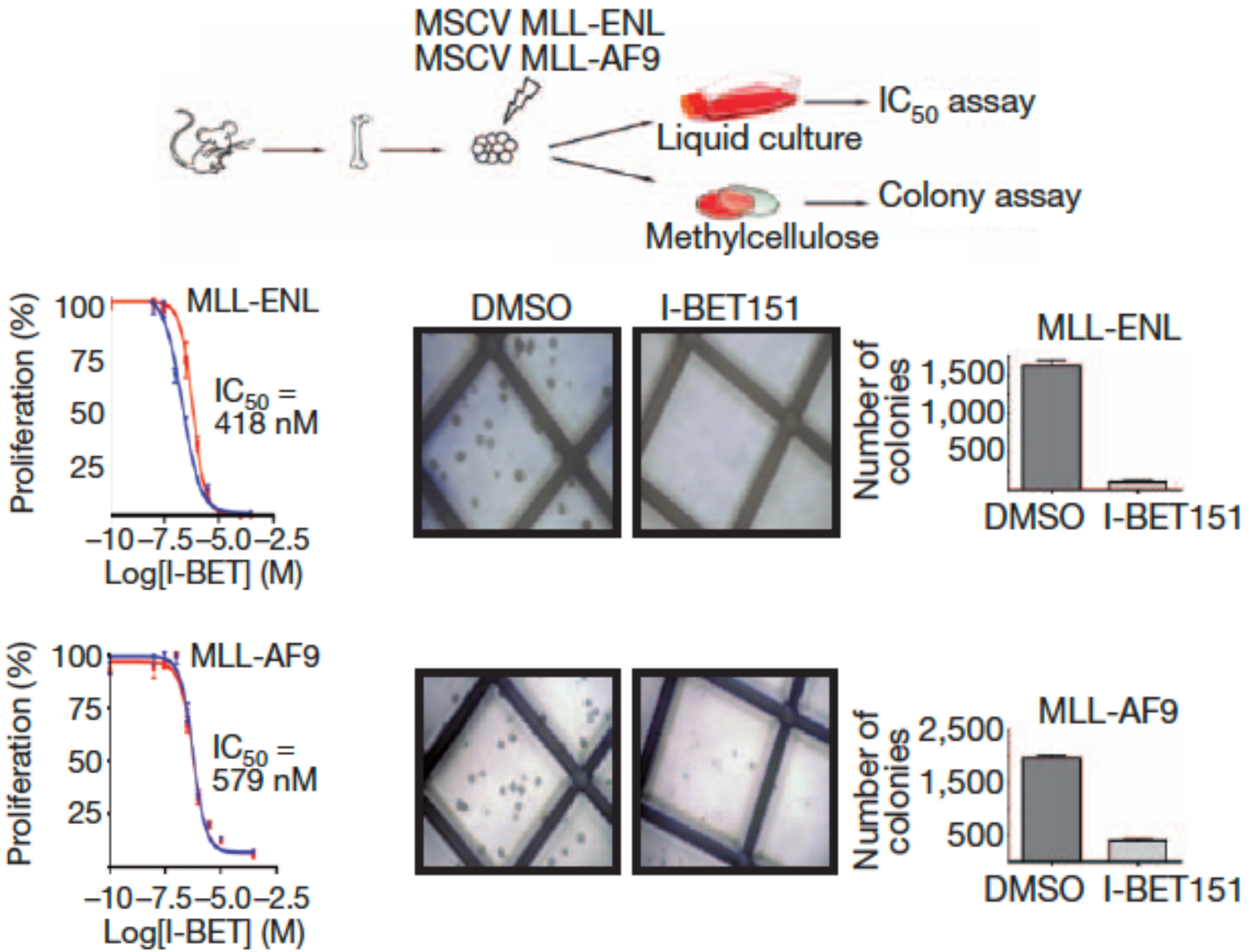
Cell Line	Oncogenic Driver	IC50
MV4;11	MLL-AF4	26nM
RS4;11	MLL-AF4	192nM
MOLM13	MLL-AF9	120nM
NOMO1	MLL-AF9	15nM
HEL	JAK2V617F	1uM
K562	BCR-ABL	>100uM
MEG01	BCR-ABL	25uM
HL60	N-RAS	890nM

10 - > 1000 Fold

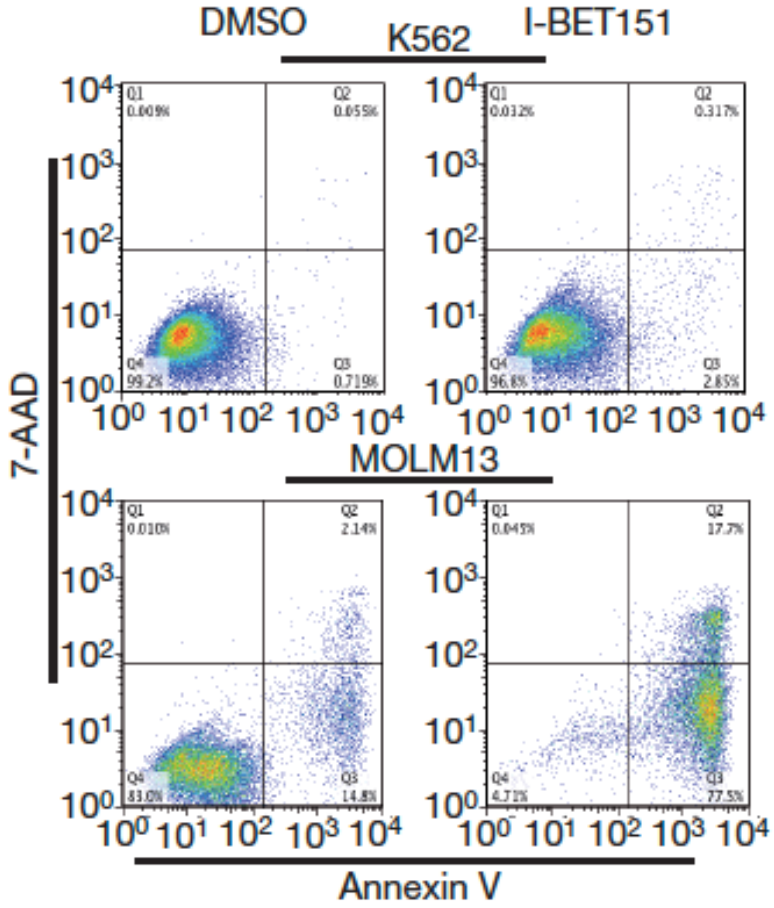
MLL-fusion cell lines

Non MLL-Fusion AML cell lines

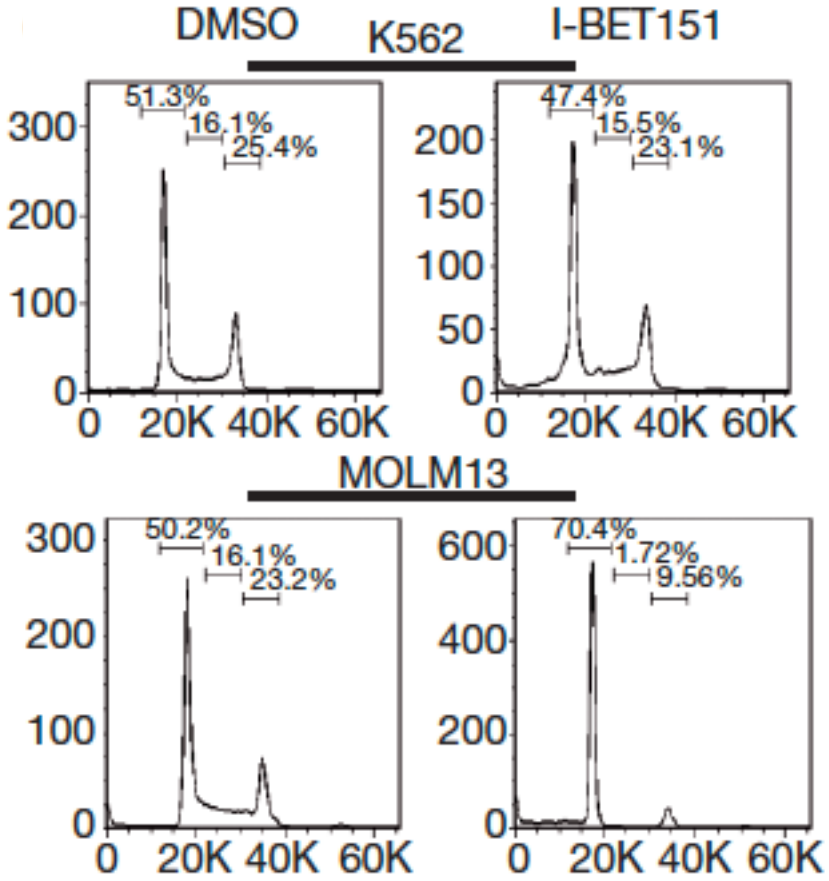
I-BET has selectivity for MLL cell lines



I-BET induces cell cycle block and apoptosis

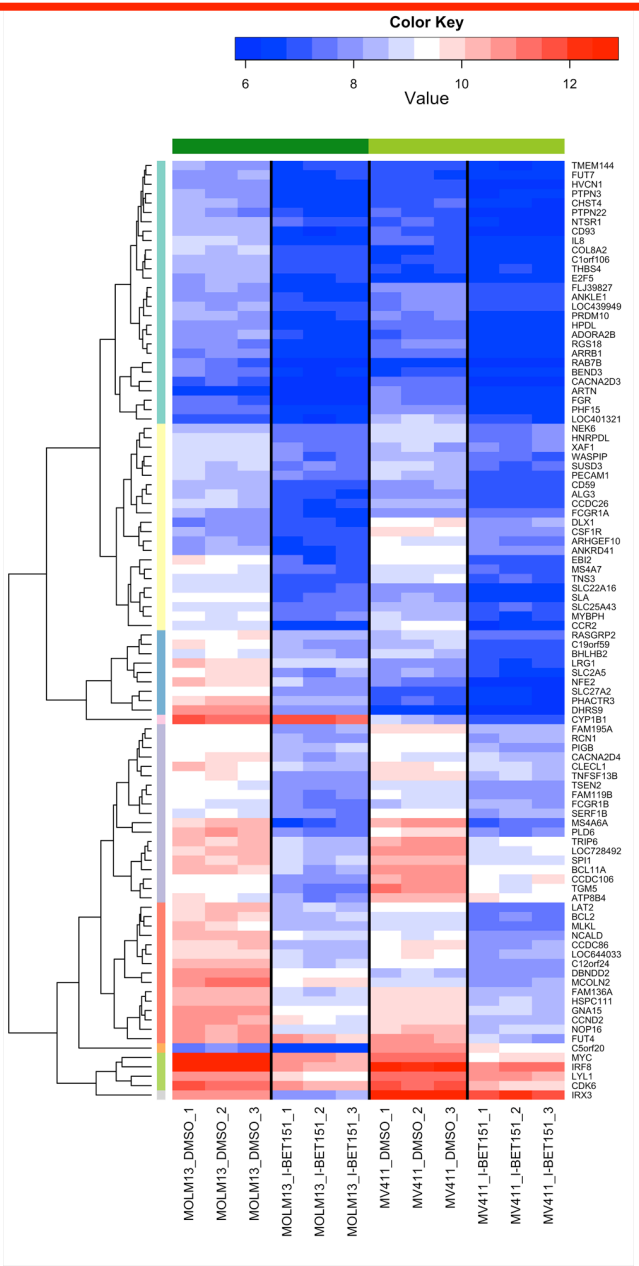


72 hours

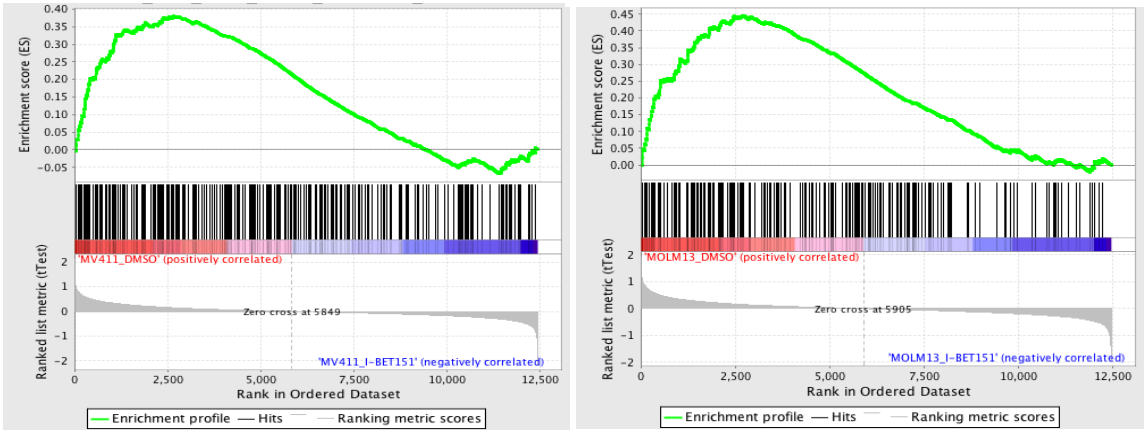


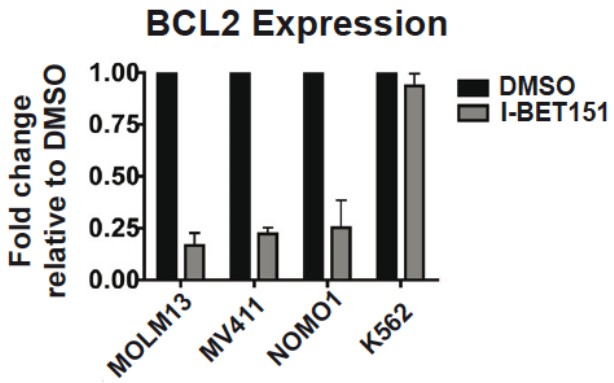
24 hours

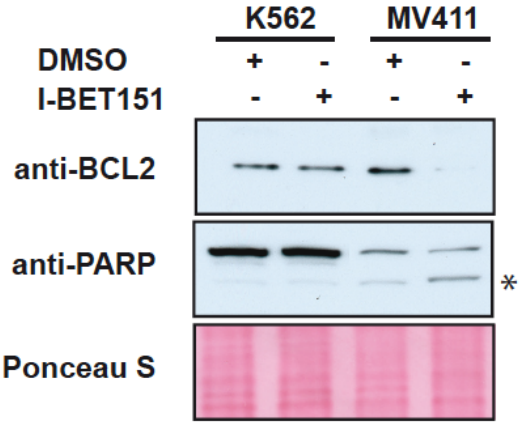
I-BET151 molecular mechanism of action



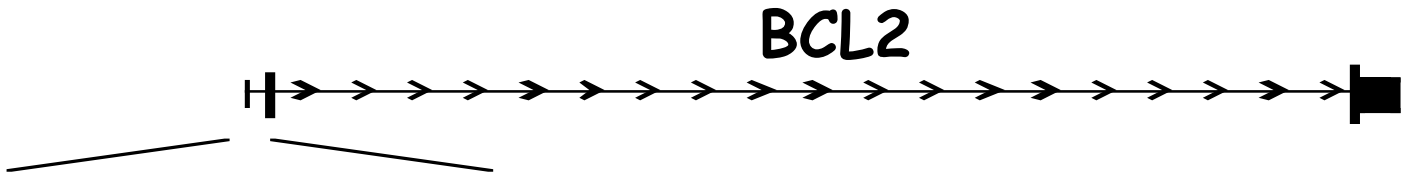
Sommervaile et al ESC self renewal signature in MLL Leukaemia

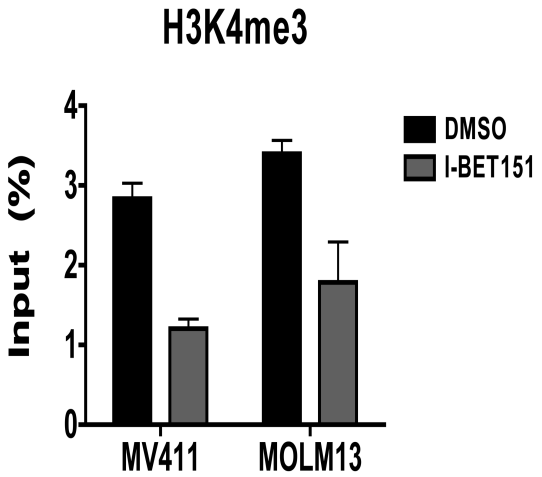


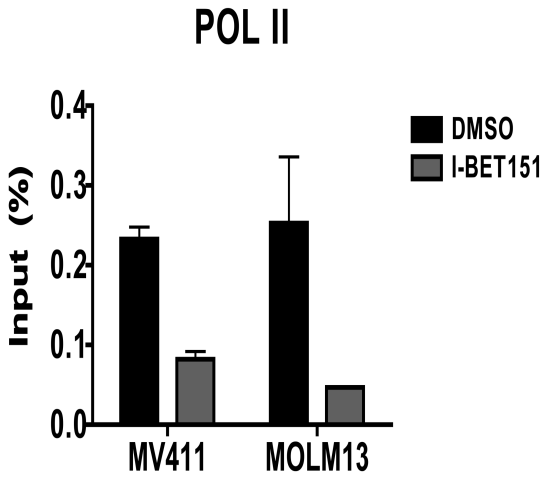


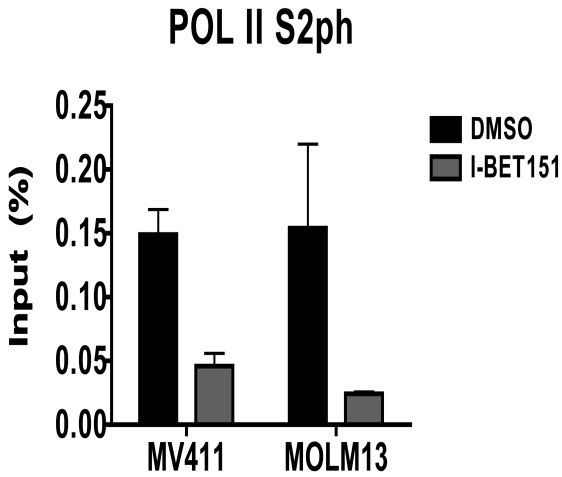


I-BET151 molecular mechanism of action

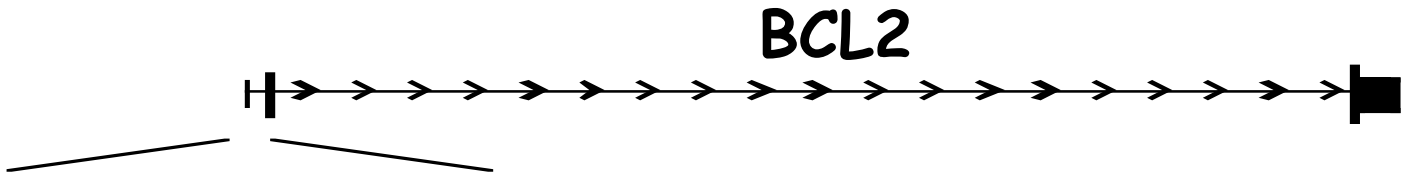


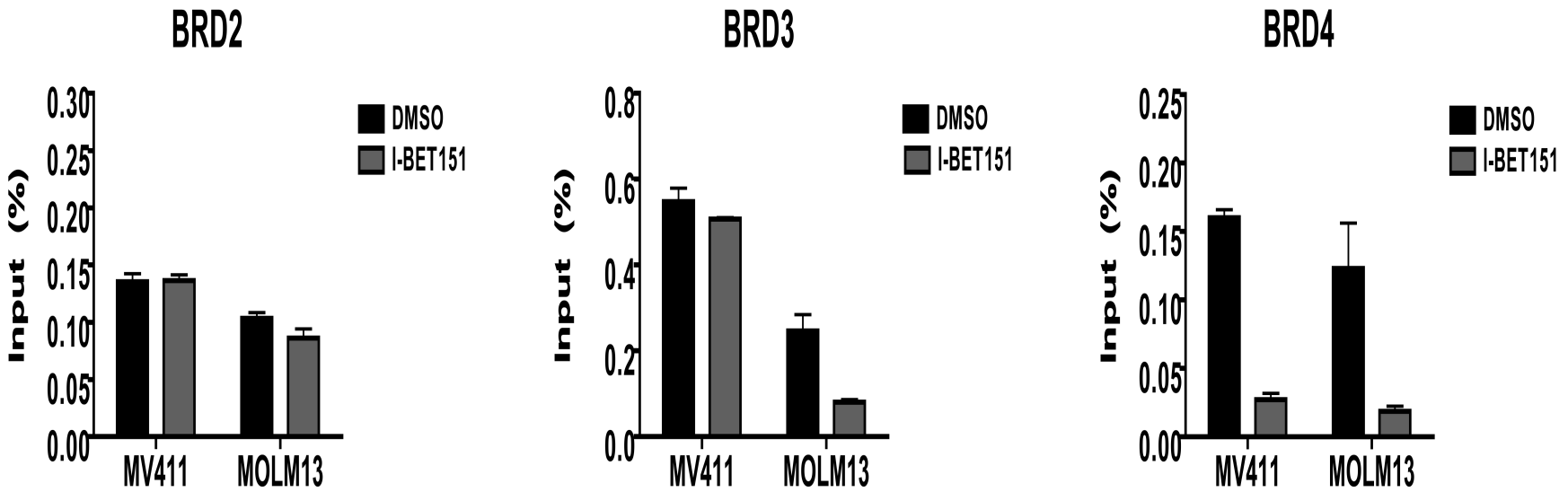




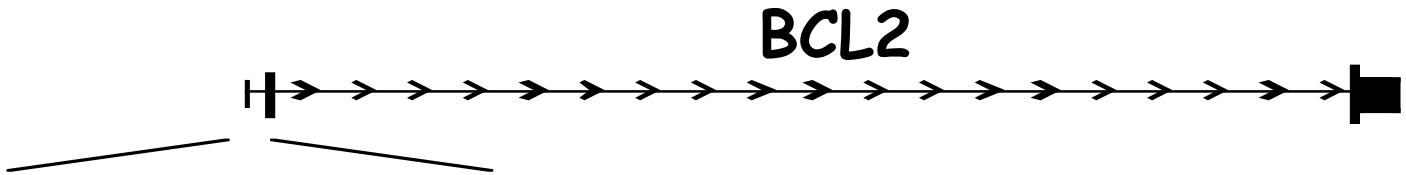


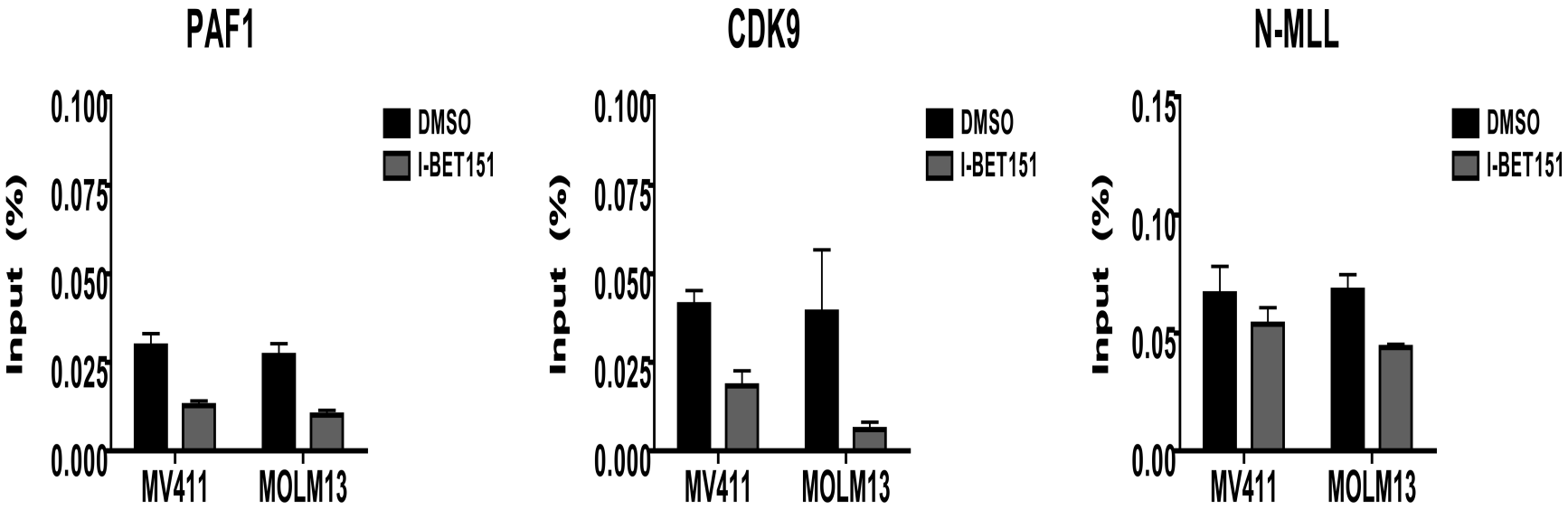
I-BET151 molecular mechanism of action





I-BET151 molecular mechanism of action





I-BET mediates disease control in MLL-AF9 leukaemia model

C57BL/6
Mice (n=20)

Transplant
2.5×10^6
Murine MLL-AF9
leukaemia cells

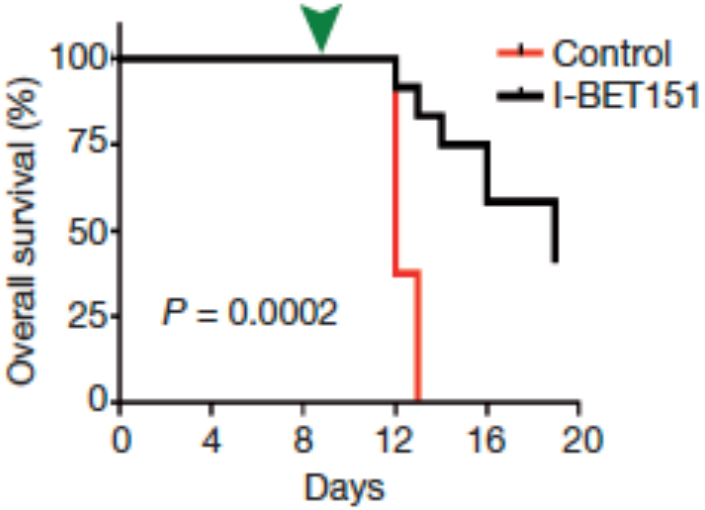
Control

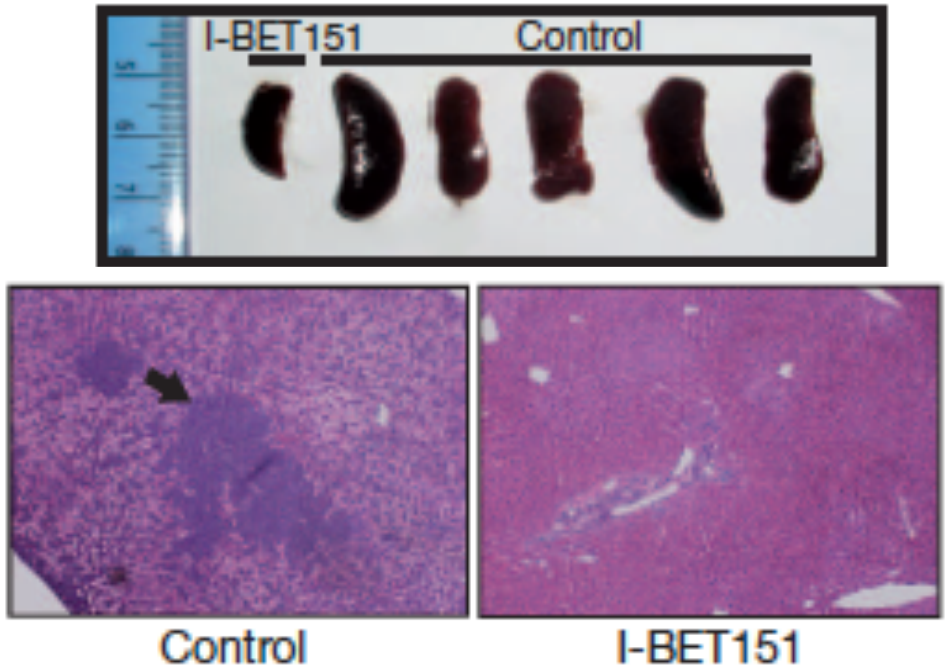
8/8 died of leukaemia

I-BET 151

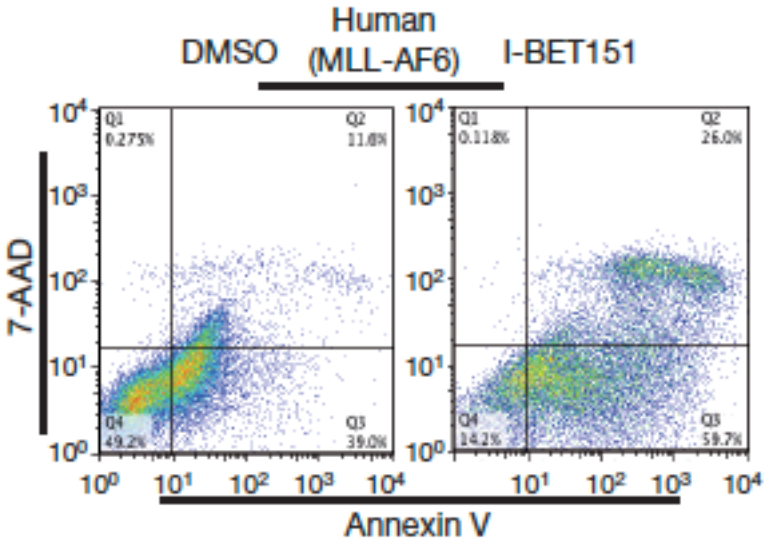
(Day 9)

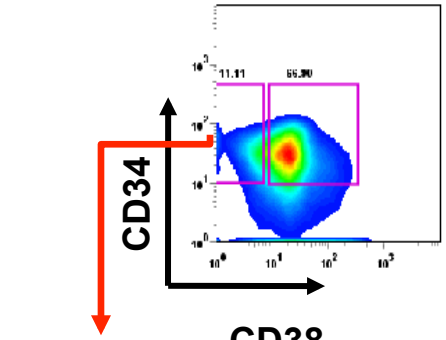
2/12 died of leukaemia
at experimental endpoint

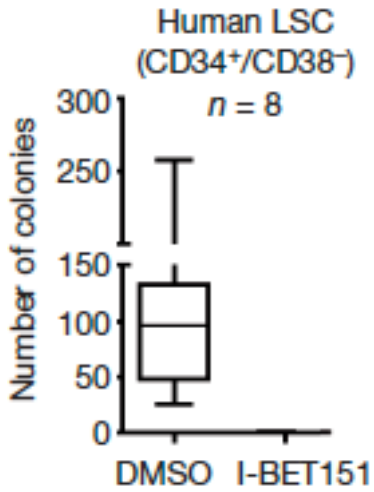


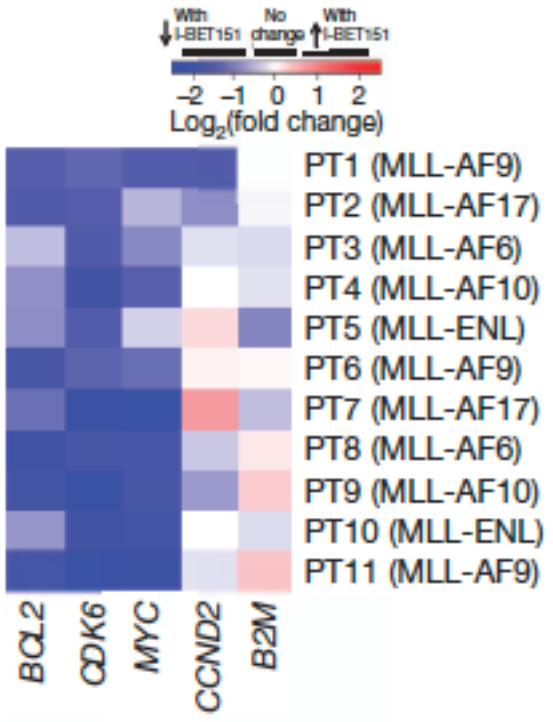


I-BET is effective in Primary Human MLL-leukaemia Samples









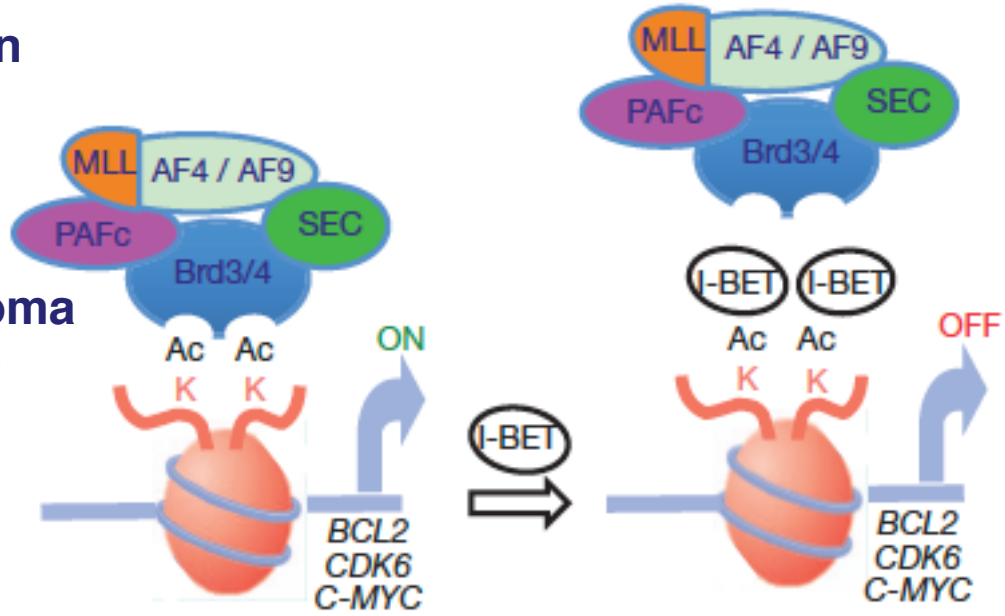
Targeting Epigenetic Readers

LETTER NATURE

Inhibition of BET recruitment to chromatin as an effective treatment for MLL-fusion leukaemia

Mark A. Dawson[1,2]*, Rab K. Prinjha[3]*, Antje Dittman[4]*, George Giotopoulos[1], Marcus Bantscheff[4], Wai-In Chan[1], Samuel C. Robson[2], Chun-wa Chung[5], Carsten Hopf[4], Mikhail M. Savitski[4], Carola Huthmacher[4], Emma Gudgin[1], Dave Lugo[3], Soren Beinke[3], Trevor D. Chapman[3], Emma J. Roberts[3], Peter E. Soden[3], Kurt R. Auger[6], Olivier Mirguet[7], Konstanze Doehner[8], Ruud Delwel[9], Alan K. Burnett[10], Phillip Jeffrey[3], Gerard Drewes[4], Kevin Lee[3], Brian J. P. Huntly[1]* & Tony Kouzarides[2]*

- **Targeting epigentic readers is an exciting new therapeutic avenue**
- **BET inhibition has therapeutic promise in AML & Multiple Myeloma**
  Zuber et al; Nature 2011, Delmore et al; Cell 2011, Mertz et al; PNAS 2011
- **Phase 1 clinical trial is being initiated**



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- Soren Beinke
- Trevor Chapman
- Emma Roberts
- Peter Soden
- Kurt Auger
- Olivier Mirguet
- Kohnstanze Döhner
- Ruud Delwel
- Alan Burnett

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- Sam Robson

- **Tony Green**

- **Bertie Gottgens**
- Sam Foster

- **All the members of the Kouzarides lab**
- **Huntly lab**
- **Green Lab**

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Haematology Society of Australia & New Zealand

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