Histone Deacetylase Inhibitors: Assessing Their Potential for Clinical Use in Neurodegenerative Disorders

Elizabeth A. Thomas, Ph.D.

Associate Professor

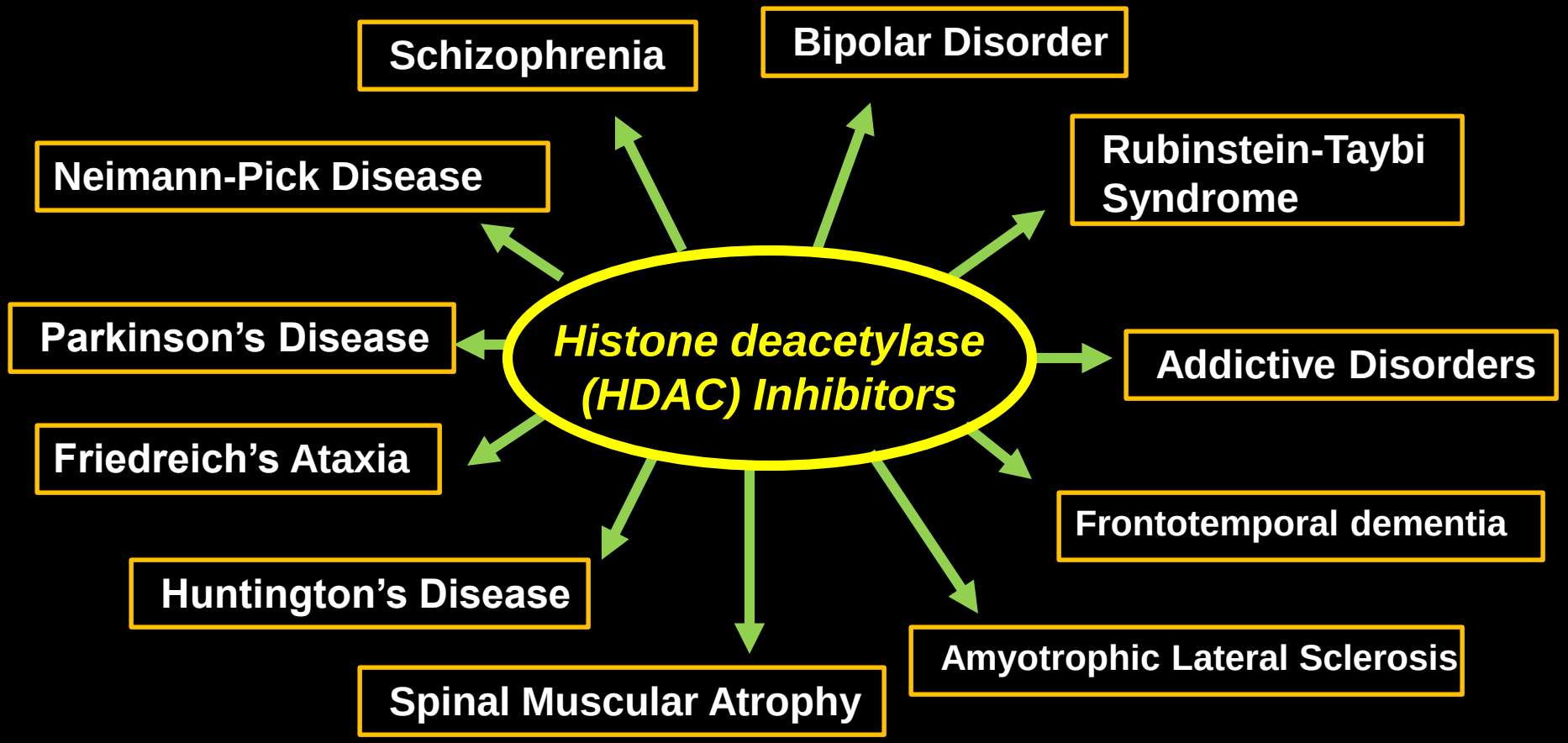
Department of Molecular and Cellular Neuroscience

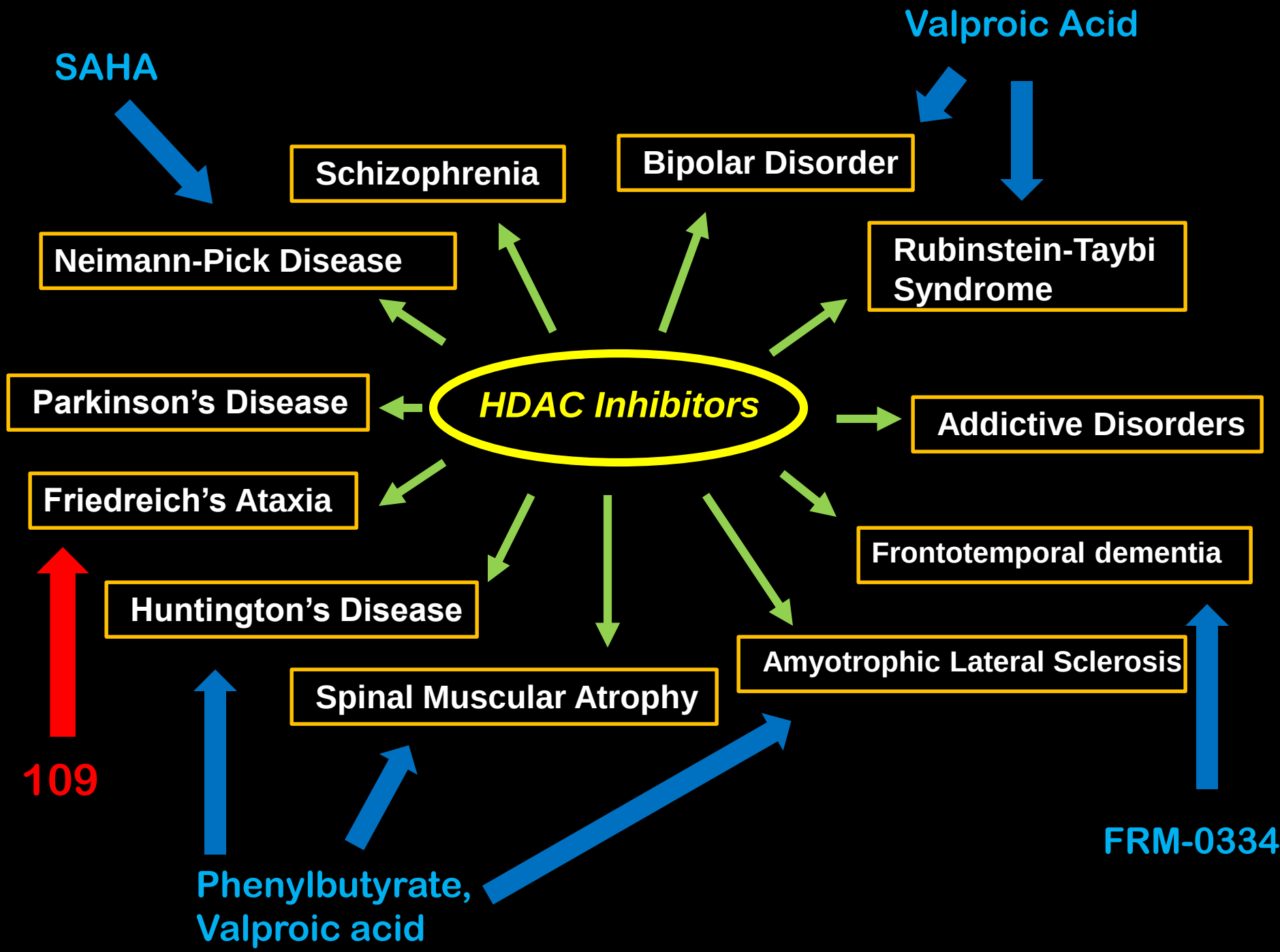
The Scripps Research Institute

La Jolla, CA

ASCPT 2016 Annual Meeting

March 10th 2016


Schizophrenia
Bipolar Disorder
Neimann-Pick Disease
Rubinstein-Taybi Syndrome
Parkinson's Disease
Histone deacetylase (HDAC) Inhibitors
Addictive Disorders
Friedreich's Ataxia
Frontotemporal dementia
Huntington's Disease
Amyotrophic Lateral Sclerosis
Spinal Muscular Atrophy


Valproic Acid
SAHA
Schizophrenia
Bipolar Disorder
Neimann-Pick Disease
Rubinstein-Taybi Syndrome
Parkinson's Disease
HDAC Inhibitors
Addictive Disorders
Friedreich's Ataxia
Frontotemporal dementia
Huntington's Disease
Amyotrophic Lateral Sclerosis
Spinal Muscular Atrophy
109
Phenylbutyrate,
Valproic acid
FRM-0334

Clinical trials with HDAC inhibitors (non-cancer)

- Phase I – Phenylbutyrate in Huntington's disease. 60 patients; 20 weeks.
- Phase I –Phenylbutyrate in Amyotrophic Lateral Sclerosis (ALS). 26 patients; 20 weeks.
- Phase I – Valproate in Spinal Muscular Atrophy (SMA). 33 patients; 6 months.
- Phase I/IIa – Valproate and Phenylbutyrate in Spinal Muscular Atrophy (SMA). 10 patients; 14 weeks.
- Phase I/II – Vorinostat (SAHA) in Niemann-Pick Disease. 15 patients; 3 months.
- Phase IIa - FRM-0334 in Frontotemporal Dementia. 30 patients; 28 days.
- Phase II – Valproate in Rubinstein-Taybi Syndrome. 60 children; 1 year.
- Phase I – 109 in Friedreich's ataxia. 20 patients; 29 days.

Goals

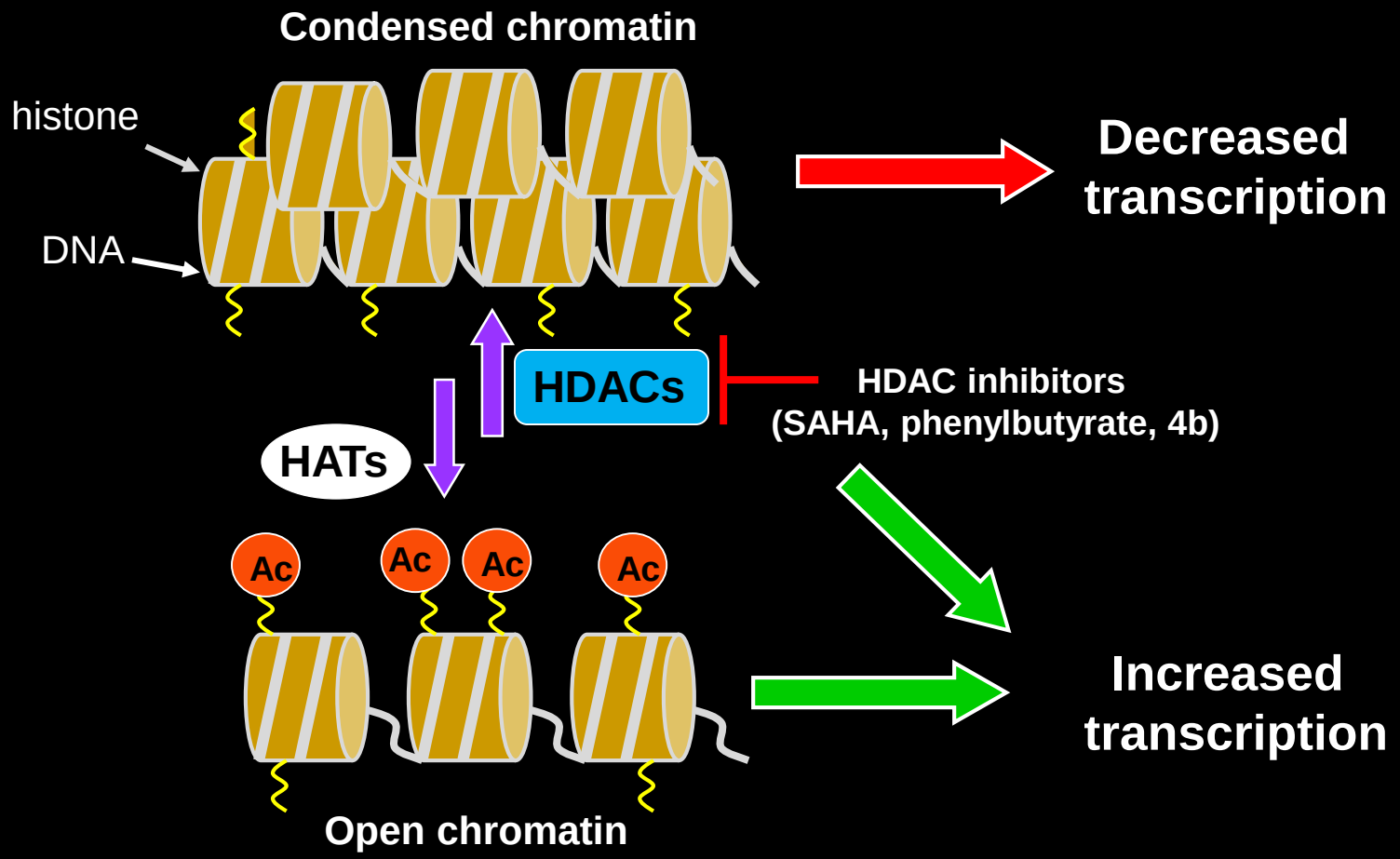
- Understanding the role(s) of epigenetic modifications in disease mechanisms and response to therapies.

  - HDAC inhibitors in Huntington's disease; preclinical studies from mouse models.

- Discover how epigenetic analysis can be applied in clinical trials to identify markers of response.

  - HDAC inhibitors in Friedreich's ataxia; clinical data from patients.

HDAC inhibitors activate gene expression by changing chromatin structure

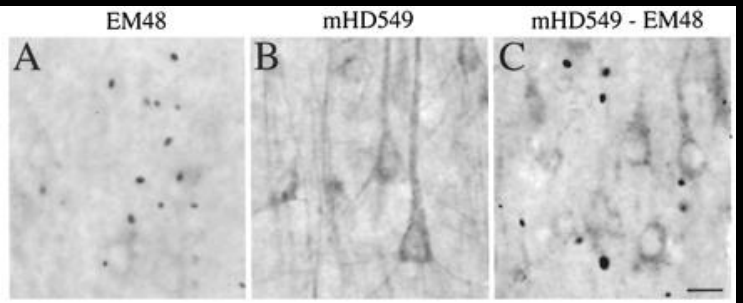


* Several neurodegenerative disorders are association with histone hypoacetylation and altered gene expression, including Huntington's disease

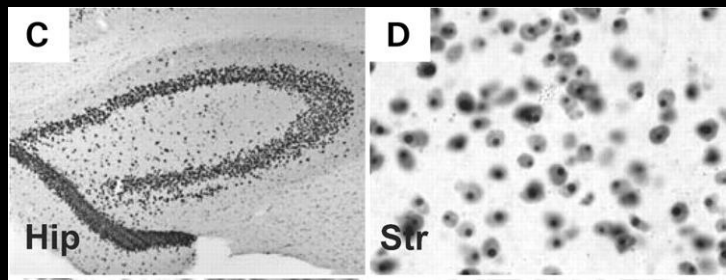
Huntington's disease (HD)

- Caused by CAG repeat expansion in exon 1 of the HD gene, resulting in a translated huntingtin protein with an expanded polyQ tract.
- Autosomal dominant; afflicts ~1 in 10,000 people.
- Obvious symptoms are random, uncontrollable movements called *chorea*, lack of coordination, unsteady gait. Other cognitive and psychiatric symptoms are often present.
- Hallmark feature of disease is the formation of huntingtin aggregates in the brain.
- Largely adult-onset. Typically, patients live 15 years after diagnosis.
- No cure; no good therapies.

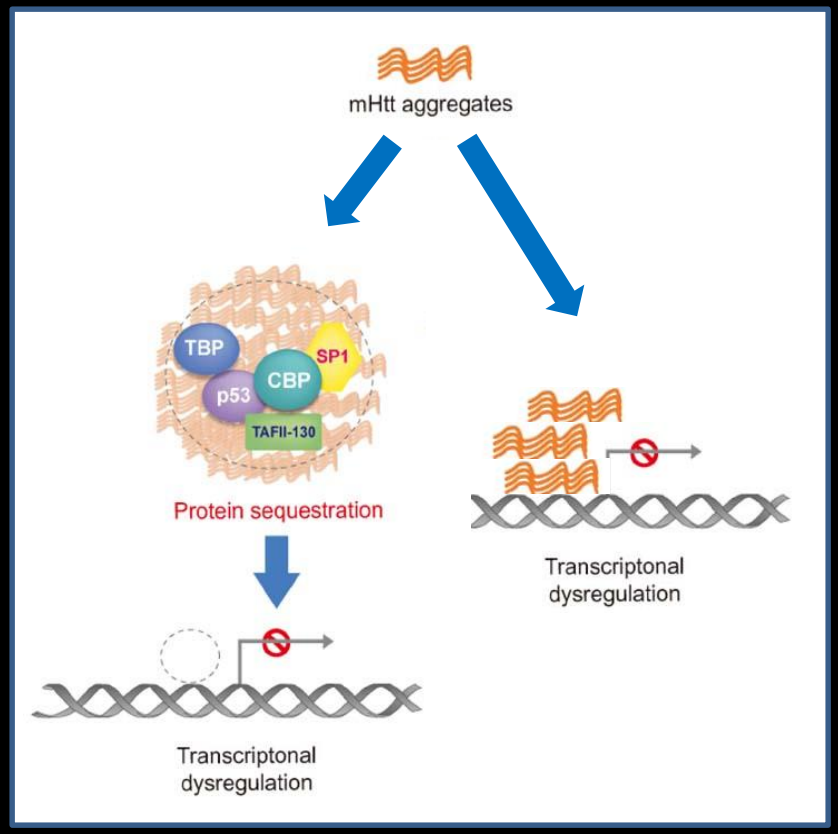
Human brain



Mouse brain



Chromatin and gene expression abnormalities in HD



Abnormal gene regulation

(Steffan et al., 2001; McCampbell et al., 2001; Cong et al., 2005; Ferrante et al., 2003; Stack et al. 2007; Sadri-Vakili et al., 2007; McFarland et al., 2012)

		Log2 ratios:			
R6/1-24 wks	R6/2-6 wks	CHL2-15 mos	Q92-18 mos	Human	Gene symbol
–0.55	–0.28	–0.31	–0.89	–1.71	*CNR1*
–0.71	–1.42	–1.08	–0.84	–1.68	*ARPP-19*
–0.83	–0.79	–0.63	–0.80	–1.68	*COCH*
–0.68	–0.53	–0.32	–0.64	–1.60	*KCNAB1*
–1.17	–0.25	–1.35	–0.81	–1.56	*RGS4*
–0.75	–0.54	–0.31	–0.74	–1.52	*PTPN5*
–0.51	–0.83	–1.03	–0.51	–1.47	*MYT1L*
–0.81	–1.41	–0.24	–1.26	–1.38	*PENK*
–0.39	–0.35	–0.32	–0.58	–1.36	*PPP3CA*
–0.56	–0.79	–0.44	–0.49	–1.34	*HPCA*
–0.41	–0.91	–0.24	–0.38	–1.28	*NGEF*
–0.70	–1.12	–0.78	–1.08	–1.23	*ADORA2A*
–0.26	–0.30	–0.20	–0.28	–1.15	*PPP1R1A*
–0.38	–0.69	–0.26	–0.70	–1.13	*RBP4*
–0.64	–0.61	–1.02	–0.43	–1.11	*PRKCB1*
–0.24	–0.42	–0.37	–0.36	–1.10	*KCNQ2*
–0.71	–0.33	–0.37	–0.83	–1.09	*CA12*
–0.69	–0.87	–0.77	–0.44	–1.08	*ITPR1*
–0.25	–0.54	–0.76	–0.96	–1.07	*RGS14*
–0.57	–0.63	–0.40	–0.54	–1.04	*CACNA2D3*
–0.47	–0.41	–0.24	–0.54	–1.03	*RAP1GAP*
–0.38	–0.23	–0.44	–0.49	–1.00	*PLCB1*
–0.21	–0.34	–0.46	–0.33	–0.92	*GNAO1*
–0.62	–1.00	–0.26	–0.75	–0.91	*PDE1B*
–0.50	–0.51	–0.74	–0.93	–0.83	*ST8SIA3*
–0.83	–0.93	–0.94	–0.99	–0.80	*RASGRP2*
–0.33	–0.35	–0.34	–0.40	–0.80	*MANIA1*
–0.34	–0.50	–0.34	–0.39	–0.80	*GNB5*
–0.57	–0.42	–0.47	–0.35	–0.79	*ATP2A2*
–0.69	–0.83	–0.40	–0.69	–0.77	*GABRD*
–0.44	–0.81	–0.20	–0.44	–0.75	*BAIAP2*
–0.61	–0.73	–0.55	–0.40	–0.75	*CX3CL1*
–0.54	–0.74	–0.55	–0.48	–0.72	*HOMER1*
–0.25	–0.39	–0.19	–0.27	–0.70	*AP1S1*
–0.39	–0.46	–0.25	–0.36	–0.66	*CAMK2B*
–0.38	–0.65	–0.53	–0.54	–0.64	*MAST3*
–0.34	–0.55	–0.49	–0.32	–0.62	*CYFIP2*
–0.48	–0.33	–0.46	–0.63	–0.60	*B3GNT2*
–0.64	–0.55	–0.61	–0.40	–0.58	*ATP2B2*
–0.35	–0.43	–0.23	–0.25	–0.57	*DIO2*
–0.53	–0.72	–0.51	–0.81	–0.56	*DRD2*
–0.50	–0.65	–0.37	–0.57	–0.55	*SEZ6*
–0.16	–0.41	–0.17	–0.49	–0.52	*ARHGEF7*
–0.32	–0.55	–0.33	–0.42	–0.51	*MEIS2*
–0.23	–0.43	–0.13	–0.42	–0.50	*HRAS*
–0.47	–0.35	–0.39	–0.51	–0.47	*SLMAP*
–0.69	–0.55	–0.72	–0.35	–0.44	*CAMK2A*
–0.34	–0.33	–0.40	–0.22	–0.43	*SRM*
–0.40	–0.78	–0.35	–0.59	–0.42	*DBP*
–0.33	–0.63	–0.49	–0.72	–0.39	*KCNK2*
–0.37	–0.51	–0.80	–0.90	–0.37	*RXRG*
–0.29	–0.52	–0.30	–0.29	–0.36	*CORO2B*
–0.30	–0.17	–0.17	–0.29	–0.36	*C16orf24*
–0.20	–0.23	–0.27	–0.19	–0.35	*MLF2*
–0.37	–0.48	–0.32	–0.32	–0.33	*KCTD 17*
–0.45	–0.32	–0.67	–0.36	–0.33	*ZNF706*
–0.18	–0.24	–0.13	–0.26	–0.30	*SEPHS1*
–0.33	–0.23	–0.38	–0.57	–0.29	*POU3F1*
–0.38	–0.15	–0.25	–0.36	–0.28	*MBTPS1*
–0.44	–0.49	–0.28	–0.41	–0.22	*USP2*

Abnormal gene expression

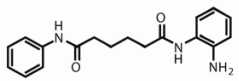
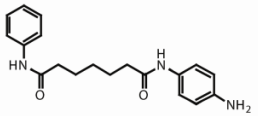
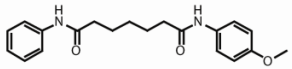
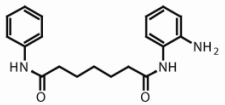
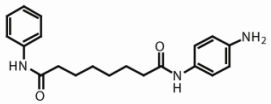
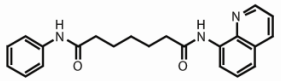
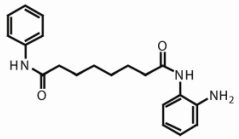
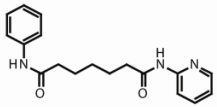
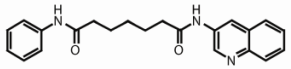
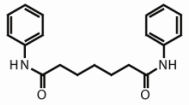
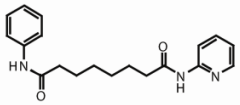
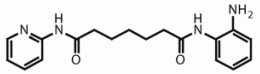
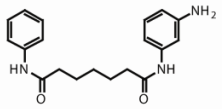
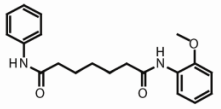
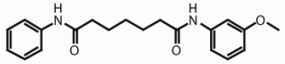
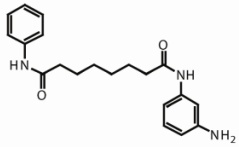
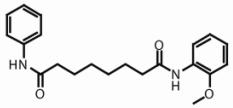
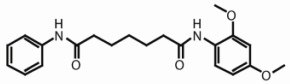
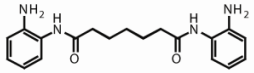
(Luthi-Carter, 2000; Luthi-Carter, 2002; Chan et al. 2002; Desplats et al., 2006; Hodges et al., 2006; Kuhn et al., 2007; Friedrich et al., 2012)

Huntington's disease is associated with a range of chromatin/gene expression abnormalities

New targets for drug treatment are aimed at correcting faulty transcription:
"histone deacetylase (HDAC) inhibitors"

Novel benzamide-type HDAC inhibitors show low toxicity

Table 1. Activities and IC_{50} values of HDAC inhibitors. Structures for each compound (numbers in bold) are shown with corresponding transcriptional change in *frataxin* mRNA in the FRDA lymphoid cell line and IC_{50} for inhibition of histone deacetylation activity in a HeLa extract.

Compound	Fold-change[1] (IC_{50})[2]	Compound	Fold-change (IC_{50})	Compound	Fold-change (IC_{50})
4a 	1.4 ± 0.06 (238 μM)	**7b** 	2.6 ± 0.14 123 μM)	**10b** 	2.5 ± 0.17 (438 μM)
4b 	2.5 ± 0.24 (78 μM)	**7c** 	2.0 ± 0.08 (186 μM)	**11b** 	3.0 ± 0.17 (17 μM)
4c 	1.4 ± 0.06 (87 μM)	**8b** 	2.6 ± 0.14 (140 μM)	**12b** 	2.5 ± 0.17 (84 μM)
5b 	1.4 ± 0.15 (204 μM)	**8c** 	2.0 ± 0.08 (99 μM)	**13b** 	2.4 ± 0.10 (91 μM)
6b 	1.5 ± 0.13 (500 μM)	**9b** 	2.3 ± 0.11 (54 μM)	**14b** 	1.8 ± 0.12 (>1 mM)
6c 	2.1 ± 0.15 (85 μM)	**9c** 	1.8 ± 0.07 (470 μM)	**15b** 	1.5 ± 0.06 (387 μM)
				16b 	3.1 ± 0.19 (14 μM)

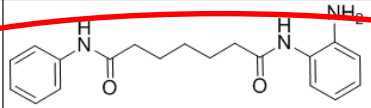
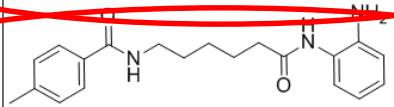
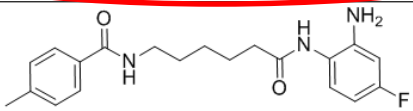
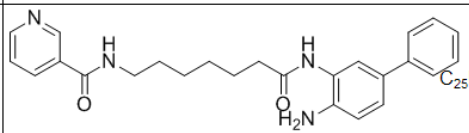
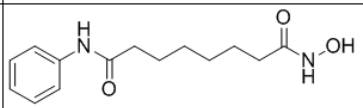
[1] Fold-change of *frataxin* mRNA in affected GM15850 cells, normalized to *GAPDH* mRNA, were determined in triplicate by real-time quantitative RT-PCR after incubation with each compound at 5 μM for 96 h. Values are relative to untreated control cells.

[2] IC_{50} values (in parenthesis below fold-change values) were determined by total histone deacetylation inhibition in a HeLa nuclear extract.

Herman et al., Nature Chem. Biol. 2:551-558, 2006

Benzamide-type HDAC inhibitors preferentially target HDAC1 and/or HDAC3

Table 1. HDAC subtype selectivity profiles for the HDAC inhibitors tested in qPCR analysis (plus SAHA as a reference).

		IC50 for Class I HDAC enzymes:				IC50 for Class II HDAC enzymes:			Selectivity:	Proliferation inhibition Hct116, IC50:	Proliferation inhibition IMR90, IC50:
Compound	**Structure**	**HDAC1**	**HDAC2**	**HDAC3**	**HDAC8**	**HDAC4**	**HDAC5**	**HDAC7**			
4b	 $C_{19}H_{23}N_3O_2$	199 nM	1.59 μM	69 nM	5 μM	>180 μM	>180 μM	>180 μM	HDAC1/3	10 μM	ND
109	 $C_{20}H_{25}N_3O_2$	300 nM	1.28 μM	63 nM	10.7 μM	>180 μM	>180 μM	>180 μM	HDAC1/3	6.5 μM	50 μM
136	 $C_{20}H_{24}FN_3O_2$	5.2 μM	3.0 μM	400 nM	13.2 μM	>180 μM	>180 μM	>180 μM	HDAC3	40 μM	>50 μM
228	 $C_{25}H_{28}N_4O_2$	61 nM	314 nM	3.33 μM	>100 μM	>180 μM	>180 μM	>180 μM	HDAC1	1.8 μM	ND
SAHA*	 $C_{14}H_{20}N_2O_3$	171 nM	389 nM	200 nM	306 nM	282 nM	400 nM	510 nM	NS	1.0 μM	1.0 μM

HDACs 6, 9, and 10 were not tested.

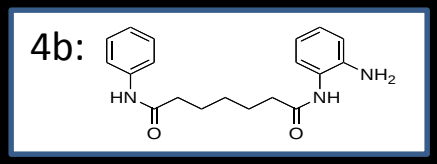
* IC_{50} values for SAHA taken from Reaction Biology Corp (www.reactionbiology.com)

NS, non-selective. ND, not determined.

RepliGen

Jia H et al., Neurobiol Dis. 2012; 46:351-61

Pipeline for screening novel HDAC1/3-targeting inhibitors for Huntington's disease

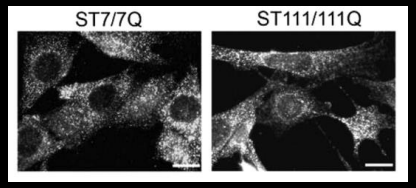


Library of ~100 novel HDAC inhibitors

RepliGen

BIOMARIN

Compounds tested for pharmacokinetics, cell permeability, metabolic stability, receptor cross-reactivity, cytotoxic properties, etc.



Striatal ell culture model

Drosophila

Mouse models (R6/2, N171-82Q, CAG140 KI)

Investigational New Drug (IND) Application

Selective HDAC1/3 inhibitors ameliorate disease phenotypes in Huntington's disease model systems

PNAS

The HDAC inhibitor 4b ameliorates the disease phenotype and transcriptional abnormalities in Huntington's disease transgenic mice

Elizabeth A. Thomas*†, Giovanni Coppola‡, Paula A. Desplats*, Bin Tang*, Elisabetta Soragni*, Ryan Burnett*, Fuying Gao‡, Kelsey M. Fitzgerald*, Jenna F. Borok*, David Herman*, Daniel H. Geschwind‡, and Joel M. Gottesfeld*

*Department of Molecular Biology, The Scripp
Institute, University of California, Los Angeles,

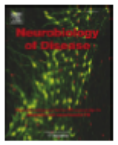
Edited by Floyd E. Bloom, The Scripps Research

Neurobiology of Disease 46 (2012) 351–361

Contents lists available at SciVerse ScienceDirect

Neurobiology of Disease

journal homepage: www.elsevier.com/locate/ynbdi



Histone deacetylase (HDAC) inhibitors targeting HDAC3 and HDAC1 ameliorate polyglutamine-elicited phenotypes in model systems of Huntington's disease

Haiqun Jia [a], Judit Pallos [b], Vincent Jacques [c], Alice Lau [d], Bin Tang [a], Andrew Cooper [c], Adeela Syed [b], Judith Purcell [b], Yi Chen [c], Shefali Sharma [c], Gavin R. Sangrey [e], Shayna B. Darnell [e], Heather Plasterer [c], Ghazaleh Sadri-Vakili [e], Joel M. Gottesfeld [a], Leslie M. Thompson [d,f], James R. Rusche [c], J. Lawrence Marsh [b], Elizabeth A. Thomas [a,*]

[a] Department of Molecular Biology, The Scripps Research Institute, La Jolla, CA, USA
[b] Department of Developmental and Cell Biology, University of California, Irvine, CA, USA
[c] Repligen Corporation, Research and Development, Waltham, MA, USA
[d] Department of Psychiatry and Human Behavior, University of California, Irvine, CA, USA
[e] NeuroEpigenetics Laboratory, MassGeneral Institute for Neurodegenerative Disease, Massa
[f] Department of Neurobiology and Behavior, University of California, Irvine, CA, USA

Human Molecular Genetics, 2012 **1–14**
doi:10.1093/hmg/dds379

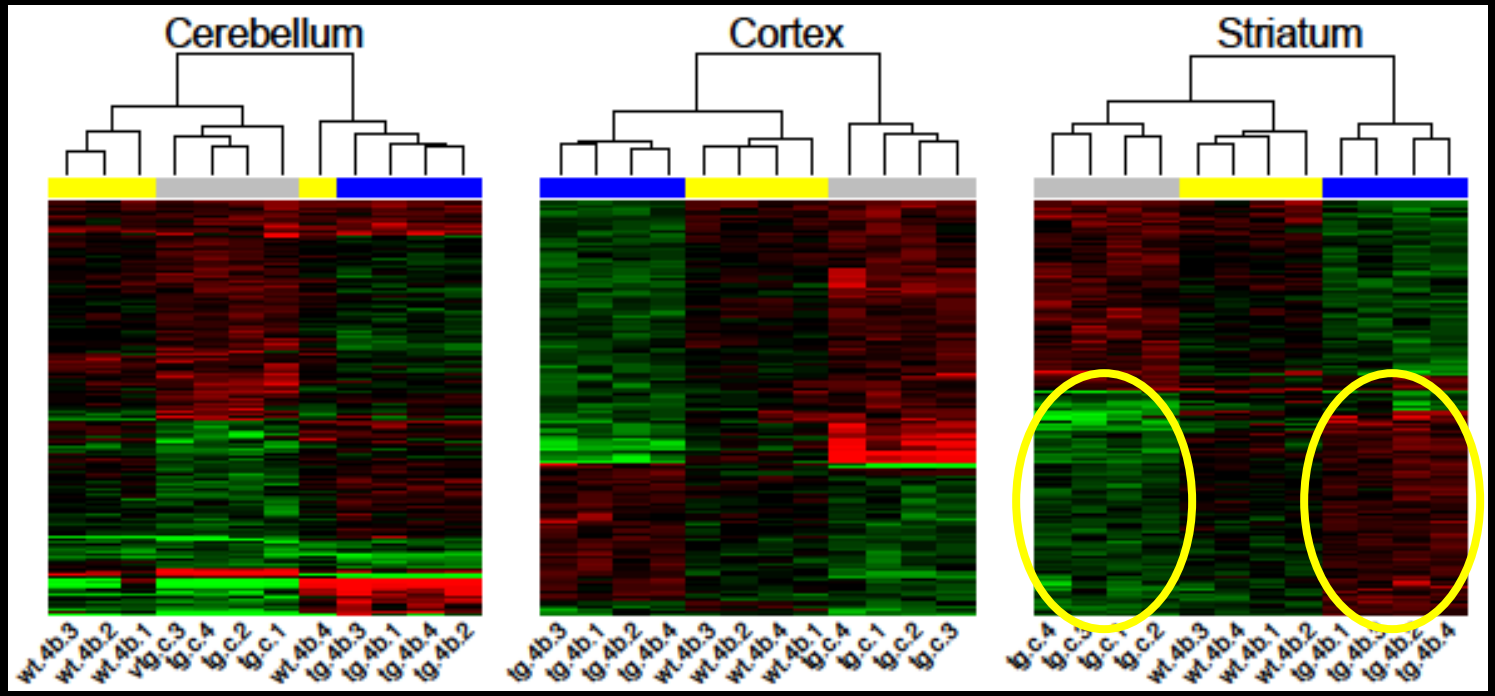
Selective histone deacetylase (HDAC) inhibition imparts beneficial effects in Huntington's disease mice: implications for the ubiquitin–proteasomal and autophagy systems

Haiqun Jia[1], Ryan J. Kast[1], Joan S. Steffan[2] and Elizabeth A. Thomas[1,*]

[1]Department of Molecular Biology, The Scripps Research Institute, La Jolla, CA, USA and [2]Department of Psychiatry and Human Behavior, University of California, Irvine, CA, USA

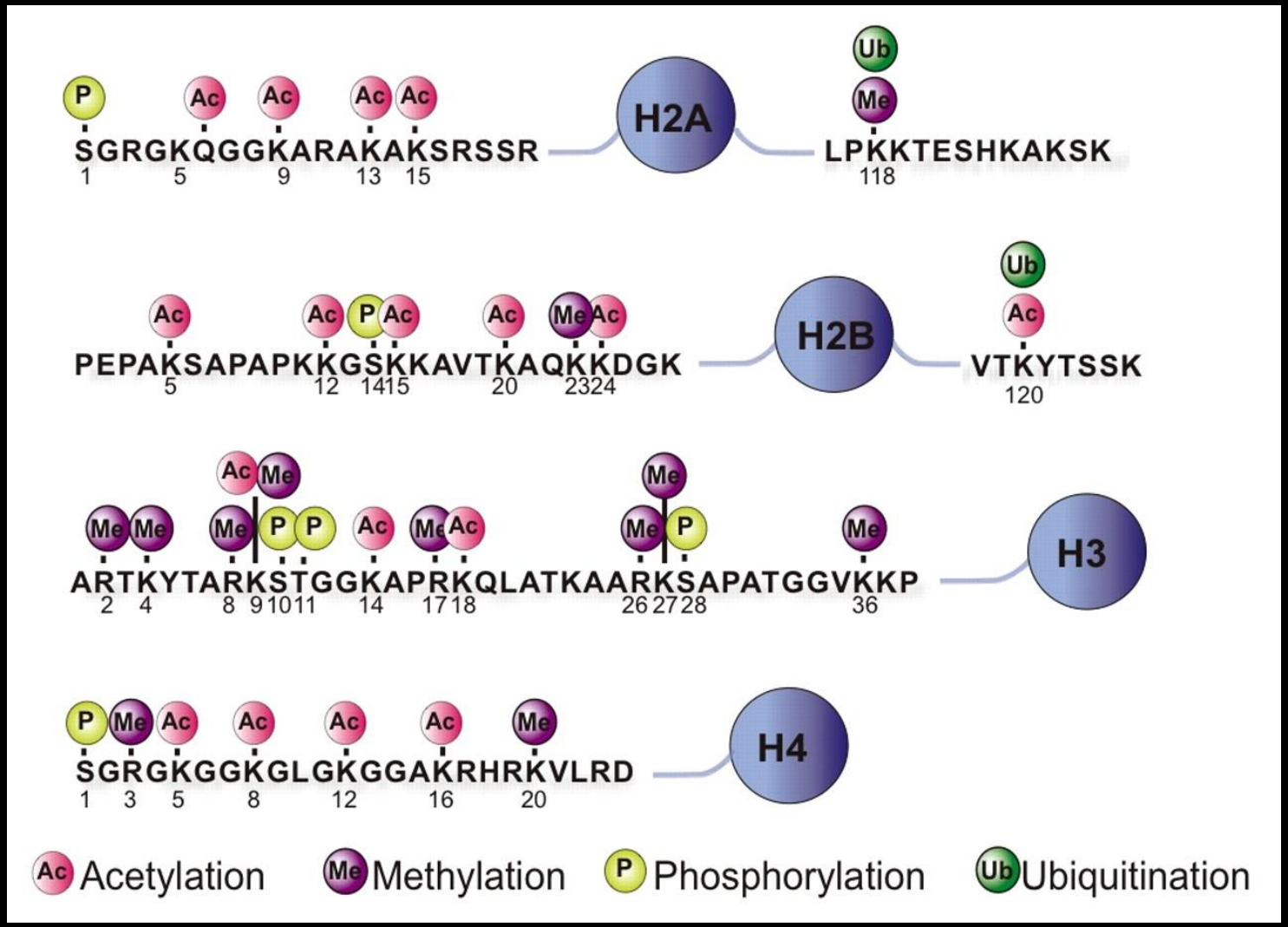
Received August 14, 2012; Revised and Accepted September 5, 2012

Gene expression signatures associated with HDACi 4b treatment

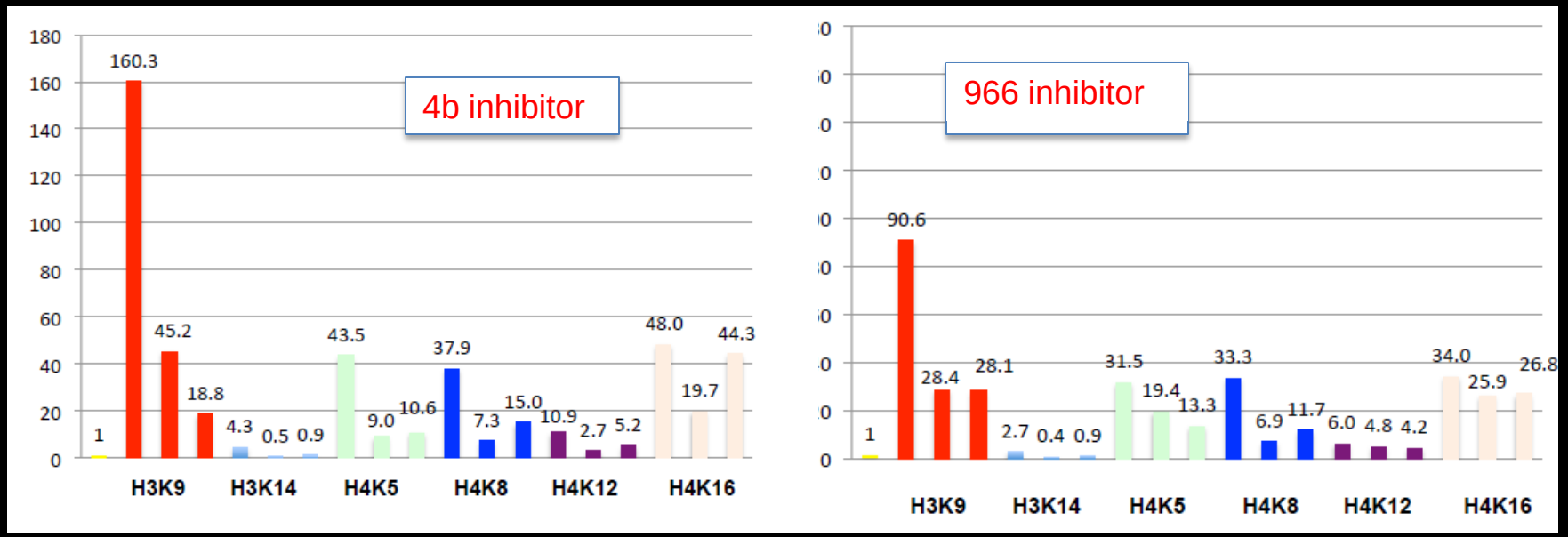


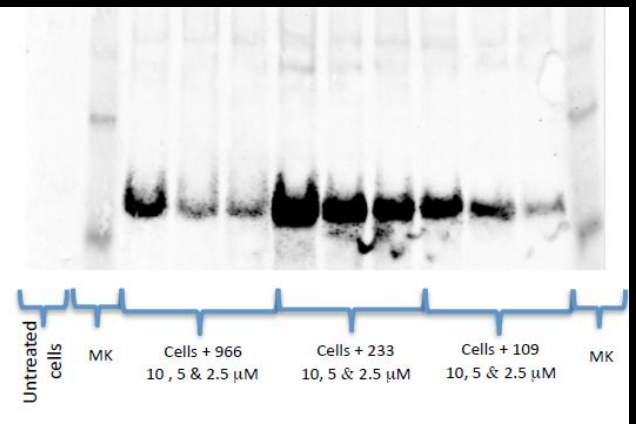
Thomas et al., PNAS USA 105:15564-9 (2008)

Histone modifications



Benzamide-type HDAC inhibitors increase histone acetylation at specific sites



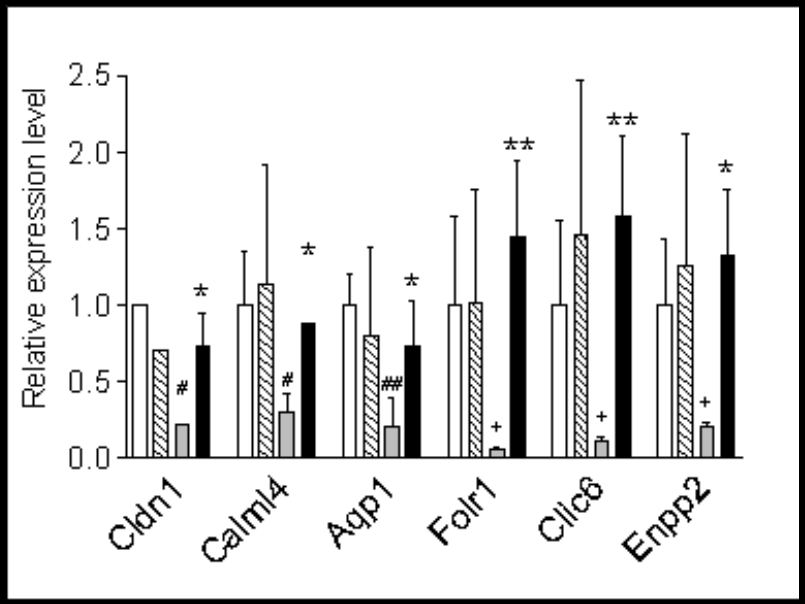


Western blot of SHSY5Y neuroblastoma cells treated with HDAC inhibitors

Unpublished

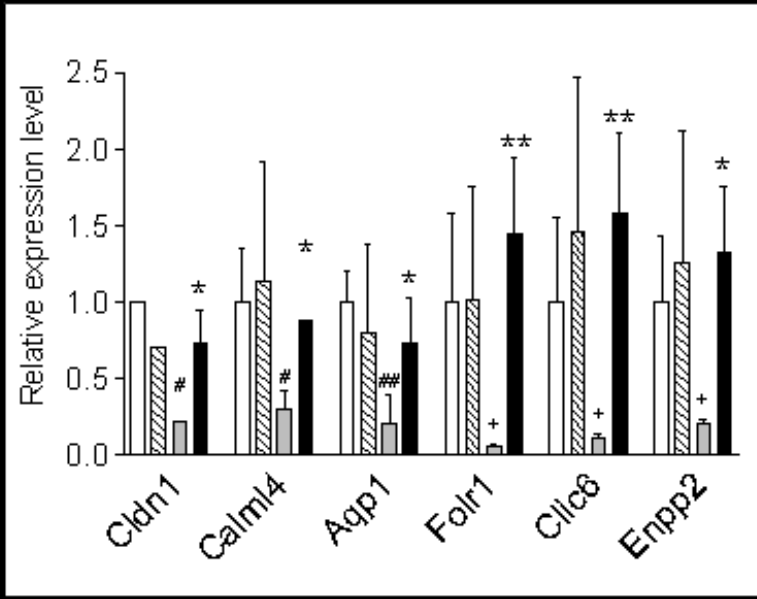
HDACi 4b treatment reverses expression downregulation coincident with increased histone H3K9 acetylation at promoters of key genes

qPCR validation:

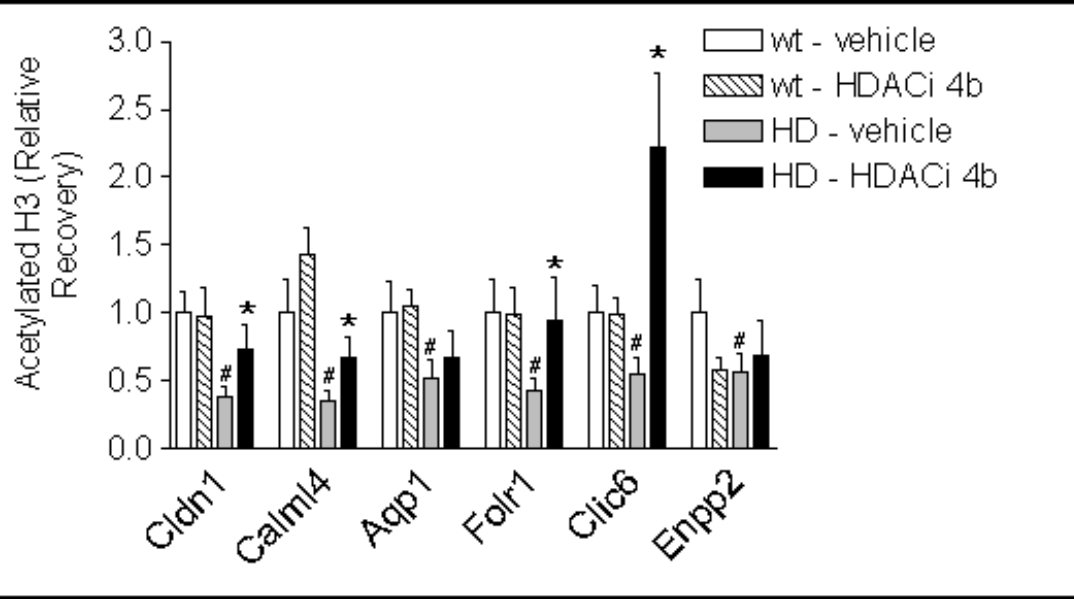


HDACi 4b treatment reverses expression downregulation coincident with increased histone H3K9 acetylation at promoters of key genes

qPCR validation:



Chromatin immunoprecipitation (ChIP) for acetylated H3 (AcH3K9):



HDAC1/3 inhibition alters the expression of DNA methylation-related genes in WT and HD mouse brain

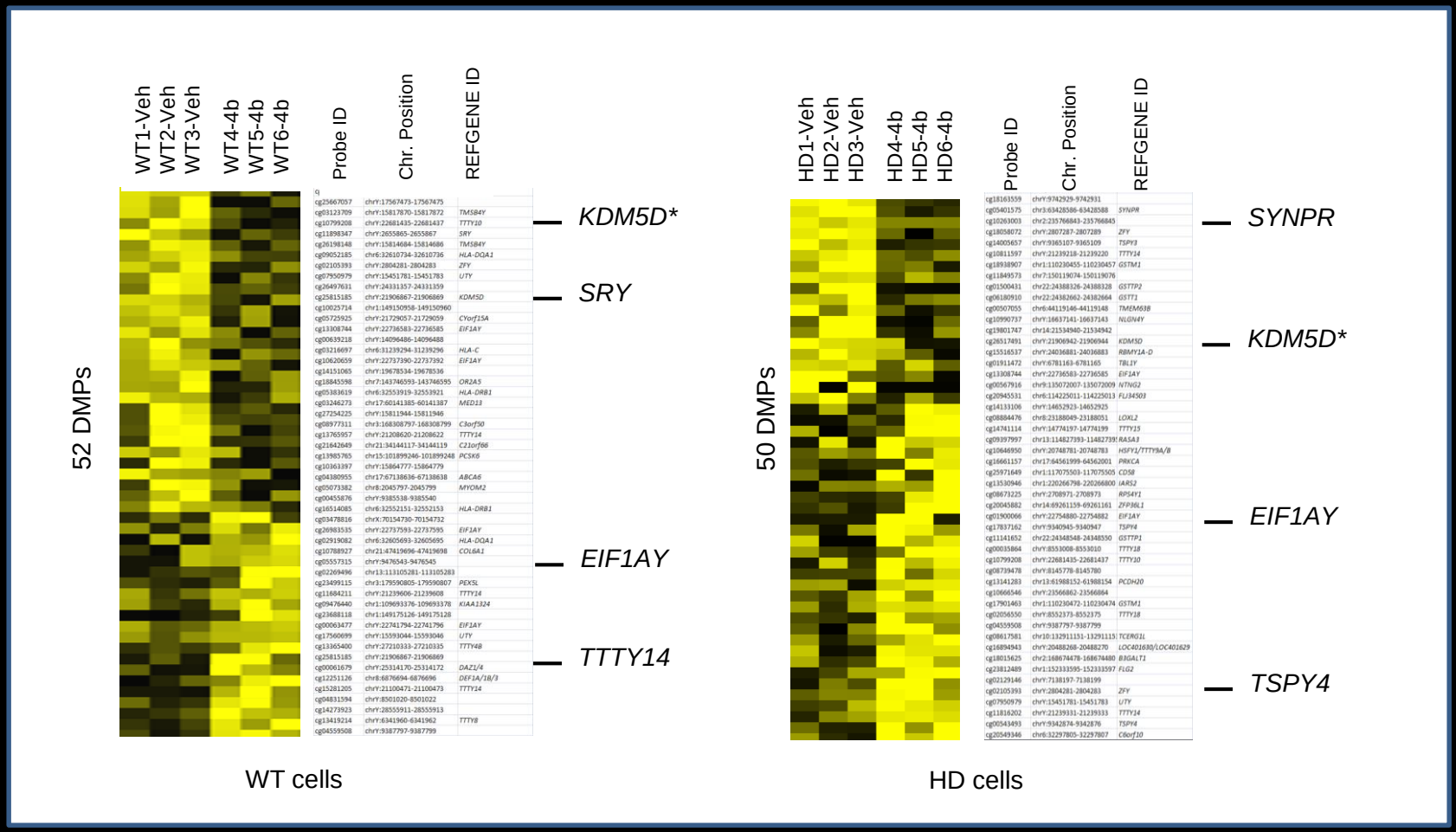
Table 1. qPCR validation of DNA methylation related genes altered by HDACi 4b treatment in cortex, striatum and muscle.

N171-82Q		**Cortex**		**Striatum**		**Muscle**	
Symbol:	**Entrez Gene Name:**	**FC:**	**p-val:**	**FC:**	**p-val:**	**FC:**	**p-val:**
Dnmt1	DNA (cytosine-5-)-methyltransferase 1	0.91	0.140	0.90	0.143	**1.61***	**0.025**
Dnmt3a	DNA (cytosine-5-)-methyltransferase 3 alpha	0.94	0.330	**1.40***	**0.032**	**1.23***	**0.011**
Gadd45b	Growth arrest and DNA-damage-inducible 45 beta	**1.44***	**0.049**	1.01	0.460	0.96	0.338
Hdac1	histone deacetylase 1	**0.81***	**0.040**	**0.57***	**0.011**	**0.81***	**0.049**
Hdac2	histone deacetylase 2	0.80	0.210	0.97	0.441	1.11	0.160
Hdac3	histone deacetylase 3	0.88	0.310	**0.44***	**0.031**	0.98	0.437
Mbd3	methyl-CpG binding domain protein 3	**1.57****	**0.006**	0.90	0.135	1.11	0.110
Mecp2	methyl CpG binding protein 2 (Rett syndrome)	0.97	0.890	1.05	0.360	**1.21***	**0.047**
Parp1	Poly (ADP-ribose) polymerase family, member 1	**1.41***	**0.007**	1.22	0.107	0.92	0.186
Rnf4	RING finger protein 4	**1.11***	**0.030**	**1.24***	**0.035**	**0.87***	**0.049**
WT		**Cortex**		**Striatum**		**Muscle**	
Symbol:	**Entrez Gene Name:**	**FC:**	**p-val:**	**FC:**	**p-val:**	**FC:**	**p-val:**
Dnmt1	DNA (cytosine-5-)-methyltransferase 1	1.00	0.48	0.98	0.43	0.79	0.13
Dnmt3A	DNA (cytosine-5-)-methyltransferase 3 alpha	0.78	0.16	**1.37***	**0.03**	0.90	0.21
Gadd45b	Growth arrest and DNA-damage-inducible 45 beta	1.11	0.12	**1.32***	**0.03**	0.92	0.31
Hdac1	histone deacetylase 1	0.90	0.29	0.97	0.40	0.98	0.45
Hdac2	histone deacetylase 2	0.98	0.46	1.09	0.26	1.12	0.12
Hdac3	histone deacetylase 3	0.90	0.31	**1.26***	**0.04**	1.10	0.25
Mbd3	methyl-CpG binding domain protein 3	**0.74****	**0.01**	0.94	0.31	0.91	0.33
Mecp2	methyl CpG binding protein 2 (Rett syndrome)	0.85	0.17	1.08	0.33	0.78	0.08
Parp1	Poly (ADP-ribose) polymerase family, member 1	**1.25***	**0.03**	0.99	0.48	0.95	0.35
Rnf4	RING finger protein 4	1.07	0.20	1.11	0.28	0.86	0.13

Bold font indicates fold-change (FC) that was significantly different, as determined by Student's t test (unpaired; two-tailed).

Jia et al., PNAS, 112(1):E56-64, 2015

HDACi 4b elicits DNA methylation changes in human fibroblasts- *(Infinium HumanMethylation450 BeadChip)*

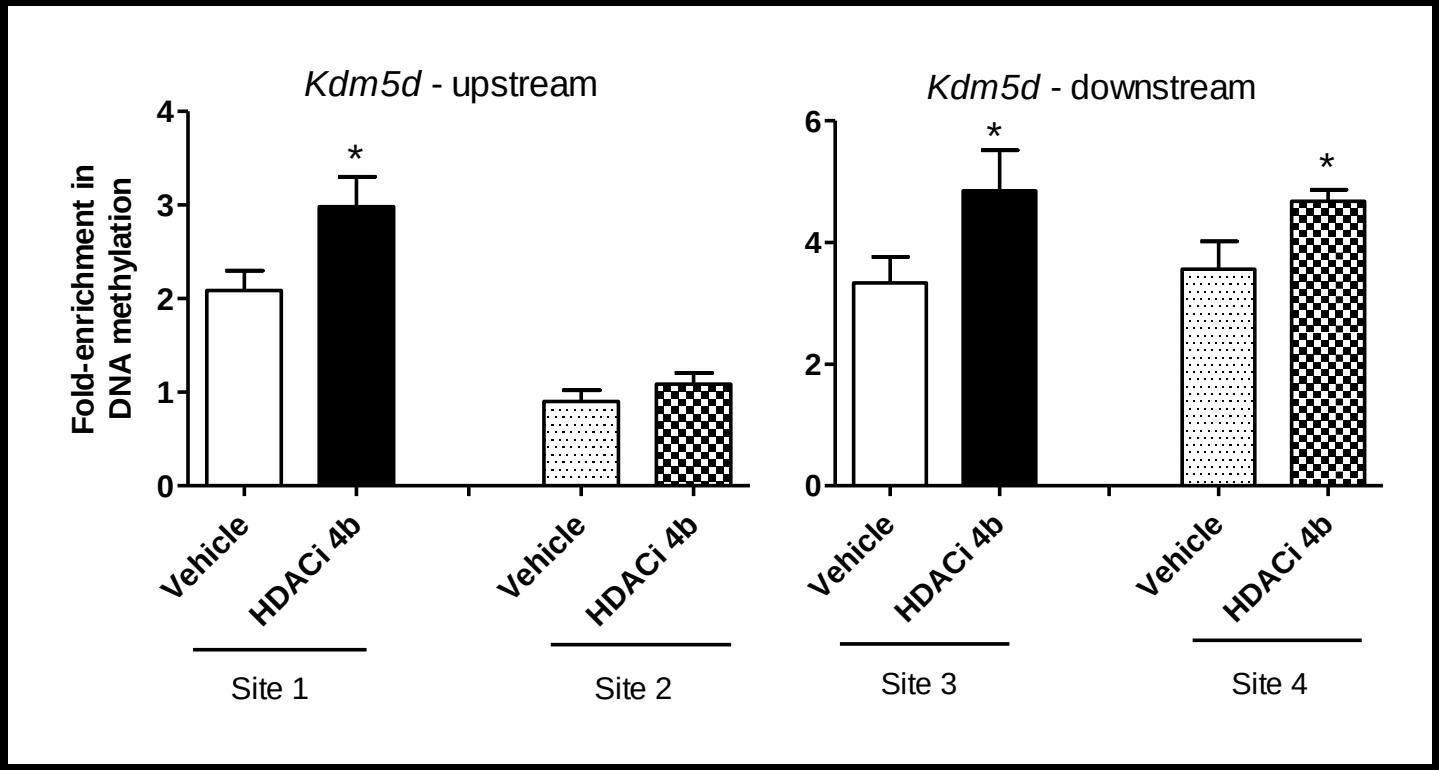


**KDM5D*: Lysine (K)-specific demethylase 5D

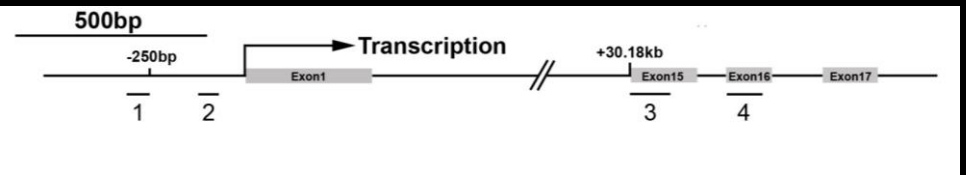
Jia et al., PNAS, 112(1):E56-64 2015

HDACi 4b elicits increased methylation at several sites at the Kdm5d locus

MeDIP RT-PCR analysis



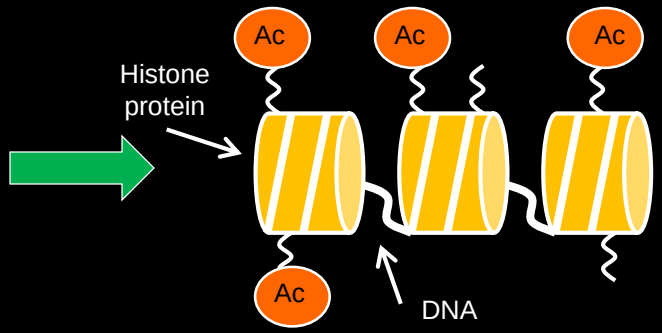
***Kdm5d* locus:**



Jia et al., PNAS, 112(1):E56-64 2015

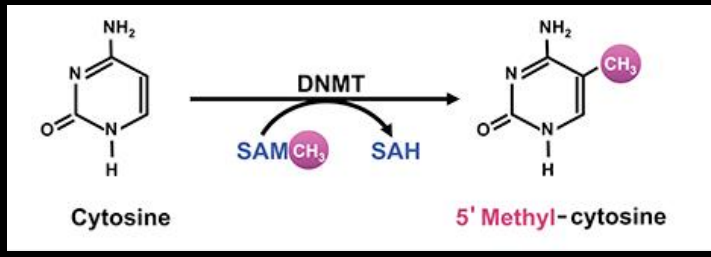
Epigenetic markers of HDAC inhibition

HDAC inhibitors



Increased Expression of DNA methylation-related genes

Increased histone acetylation marks – H3K9



Altered DNA methylation – KDM5D

Goals

- Understanding the role(s) of epigenetic modifications in disease mechanisms and response to therapies.

  - HDAC inhibitors in Huntington's disease; preclinical studies from mouse models.

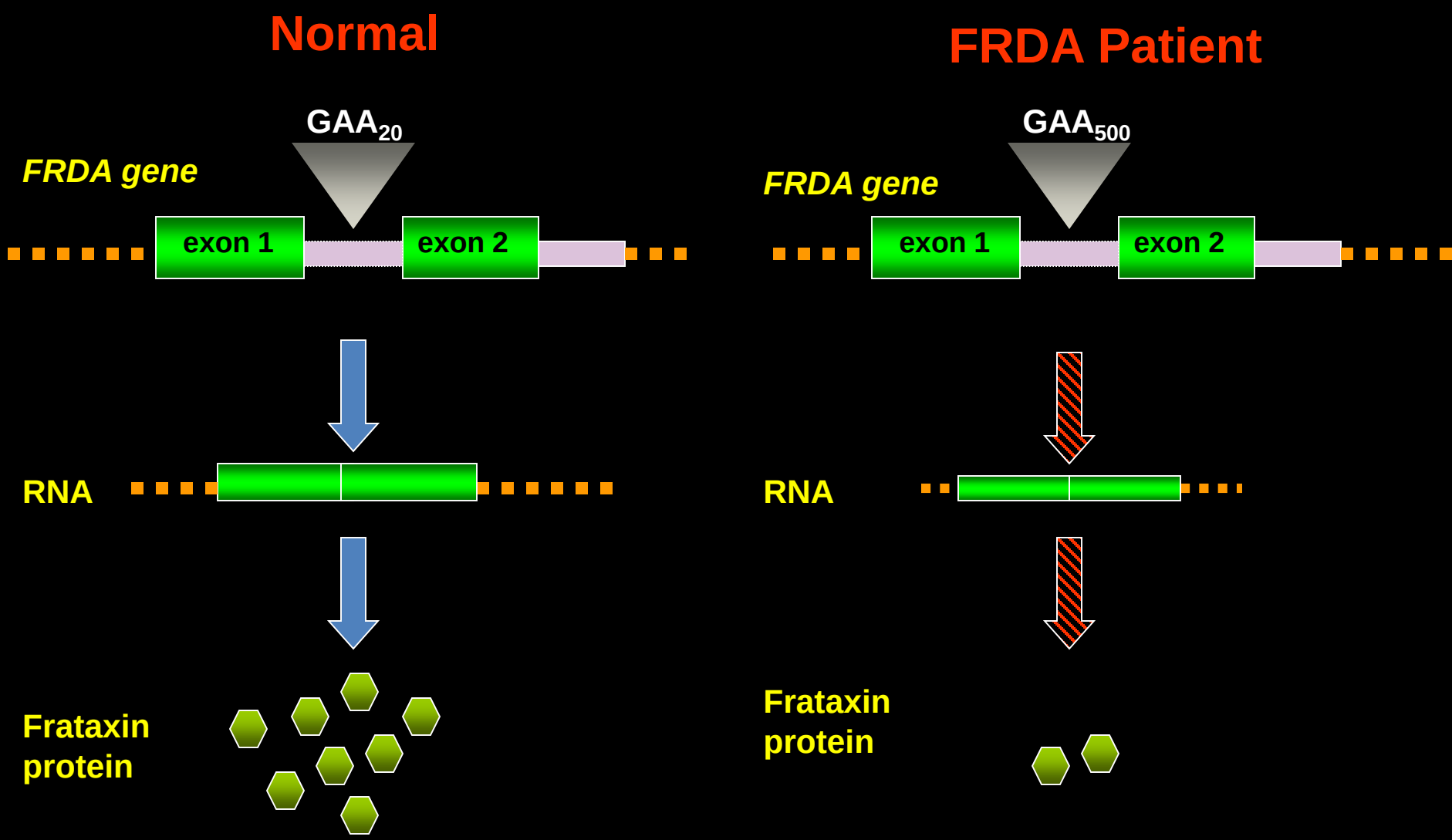
- Discover how epigenetic analysis can be applied in clinical trials to identify markers of response.

  - HDAC inhibitors in Friedreich's ataxia; clinical data from patients.

Friedreich's ataxia (FRDA)

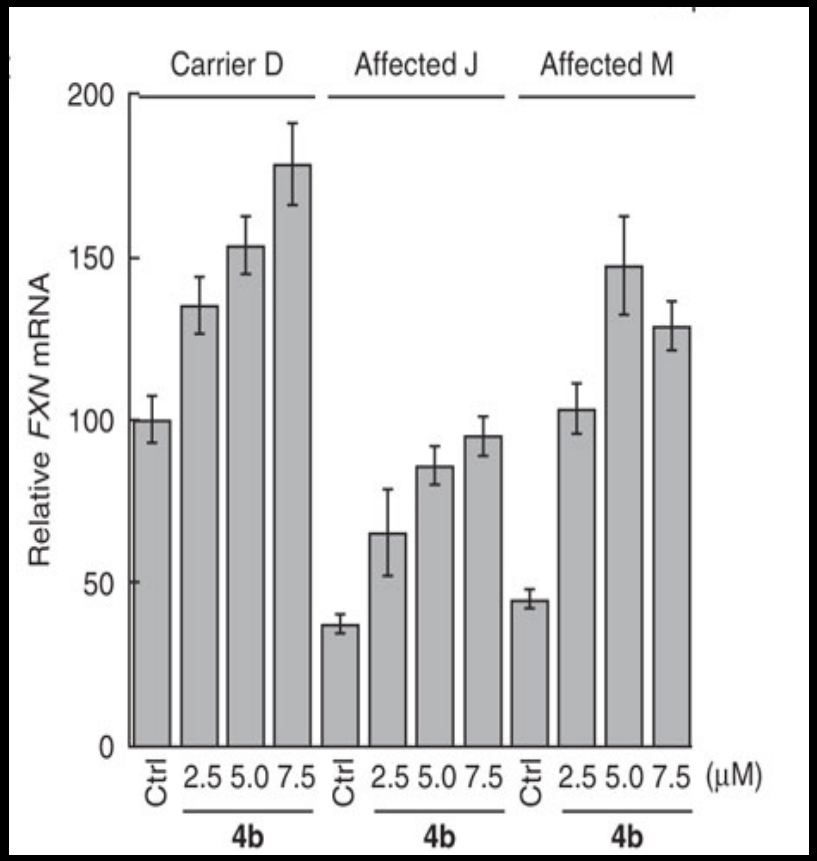
- Caused by an expansion of a GAA triplet repeat in the first intron of the FXN gene, which encodes the essential mitochondrial protein, frataxin.
- Autosomal recessive, progressive neurological disease.
- Most common form of hereditary ataxia, affecting about 1 in every 50,000 people in the United States.
- Main symptom is impaired muscle coordination (ataxia); it can also lead to scoliosis, heart disease and diabetes, but does not appreciably affect cognitive function.
- Symptoms typically begin between the ages of 5 and 15 years. Generally, within 10 to 20 years after the appearance of the first symptoms, the person is confined to a wheelchair.
- No therapies that address pathology.

Friedreich's ataxia (FRDA)



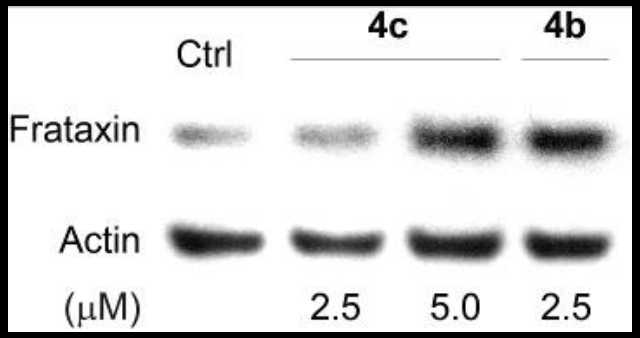
Novel HDACi 4b increases frataxin mRNA and protein in FRDA patient lymphocytes/lymphoblastoid cell lines

qPCR for frataxin mRNA

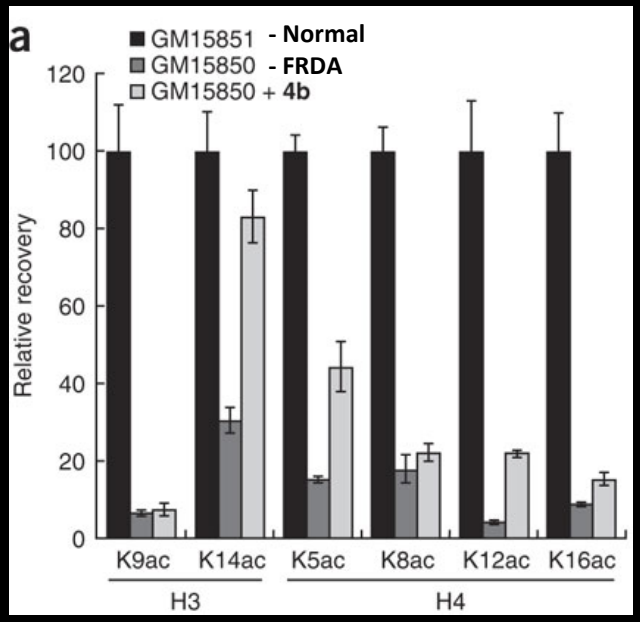


Reversal of the transcription defect to at least carrier status

Western blot for frataxin protein



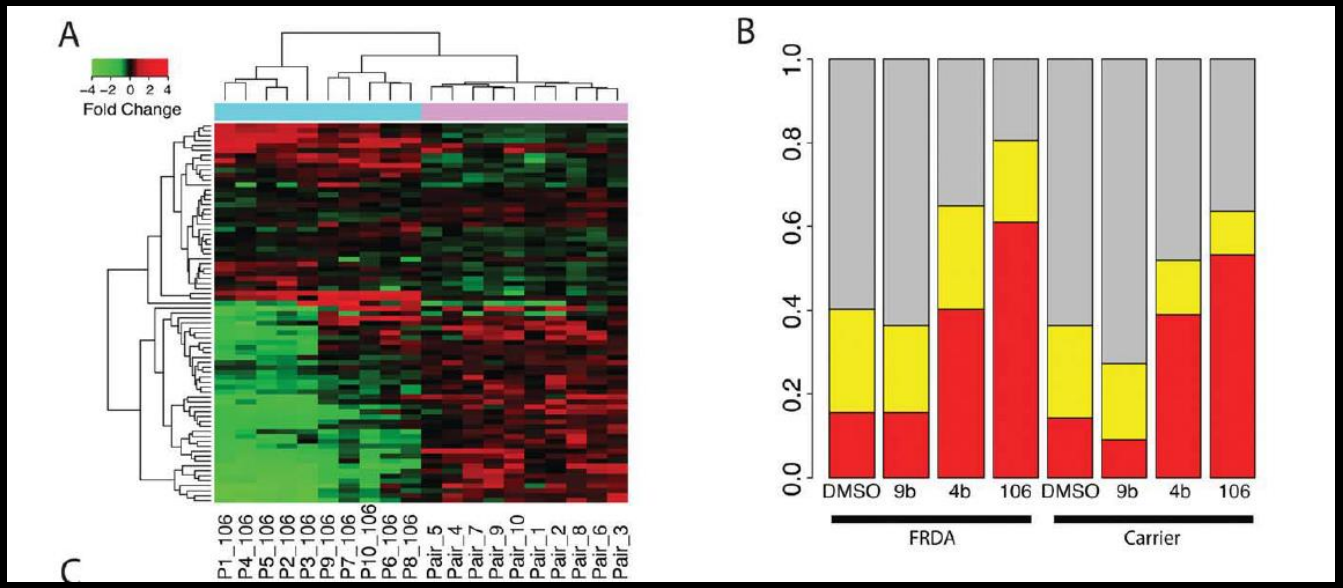
ChIP at the FXN locus



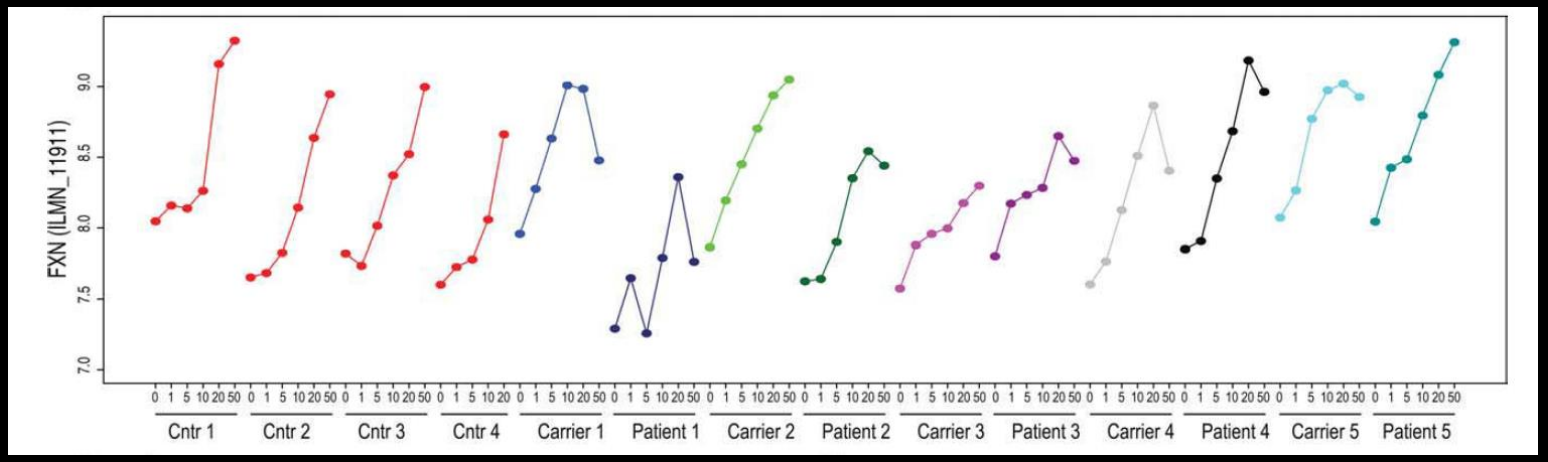
Herman et al., Nature Chem. Biol. 2:551-558, 2006

HDAC inhibitor effect on gene expression profiles in cultured peripheral blood mononuclear cells (PBMCs)

77 gene biomarker panel

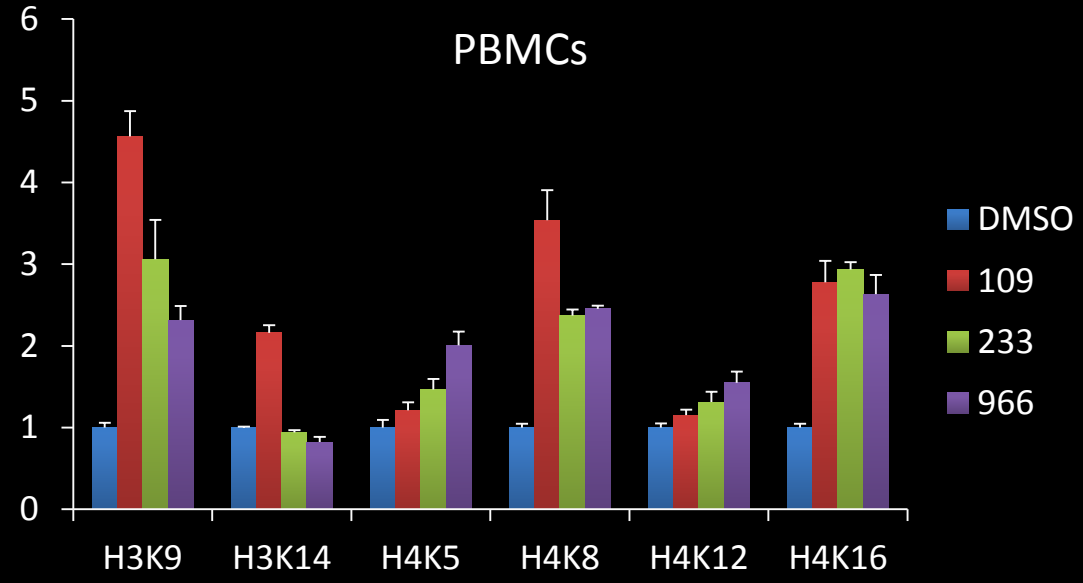


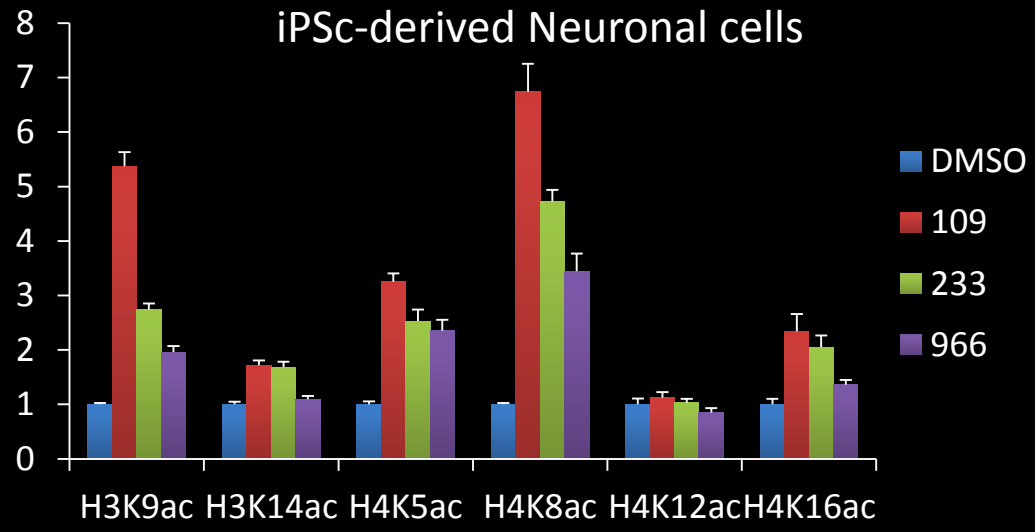
Dose-dependent increases in frataxin gene expression:



Coppola G, et al., Ann Neurol. 2011 70(5):790-804.

Chromatin immunoprecipitation experiments identify key residues for FXN activation





ChIP experiments demonstrate that H3K9 and H4K8 are critical residues for *FXN* gene activation. These could be used as a biomarker in FRDA patient trials.

RG2833 (109): First in patient clinical study

- San Luigi Gonzaga Hospital, University of Turin, Italy.
- 22 patients (split into 4 cohorts receiving different doses: 30-180 mg).
- Biomarker measures:
  - Frataxin mRNA and protein in blood, PBMCs and buccal cells
  - HDAC activity in PBMCs
  - ChIP for H3K9 acetylation in PBMCs

TABLE 1

Demographic and Clinical Characteristics of Study Subjects

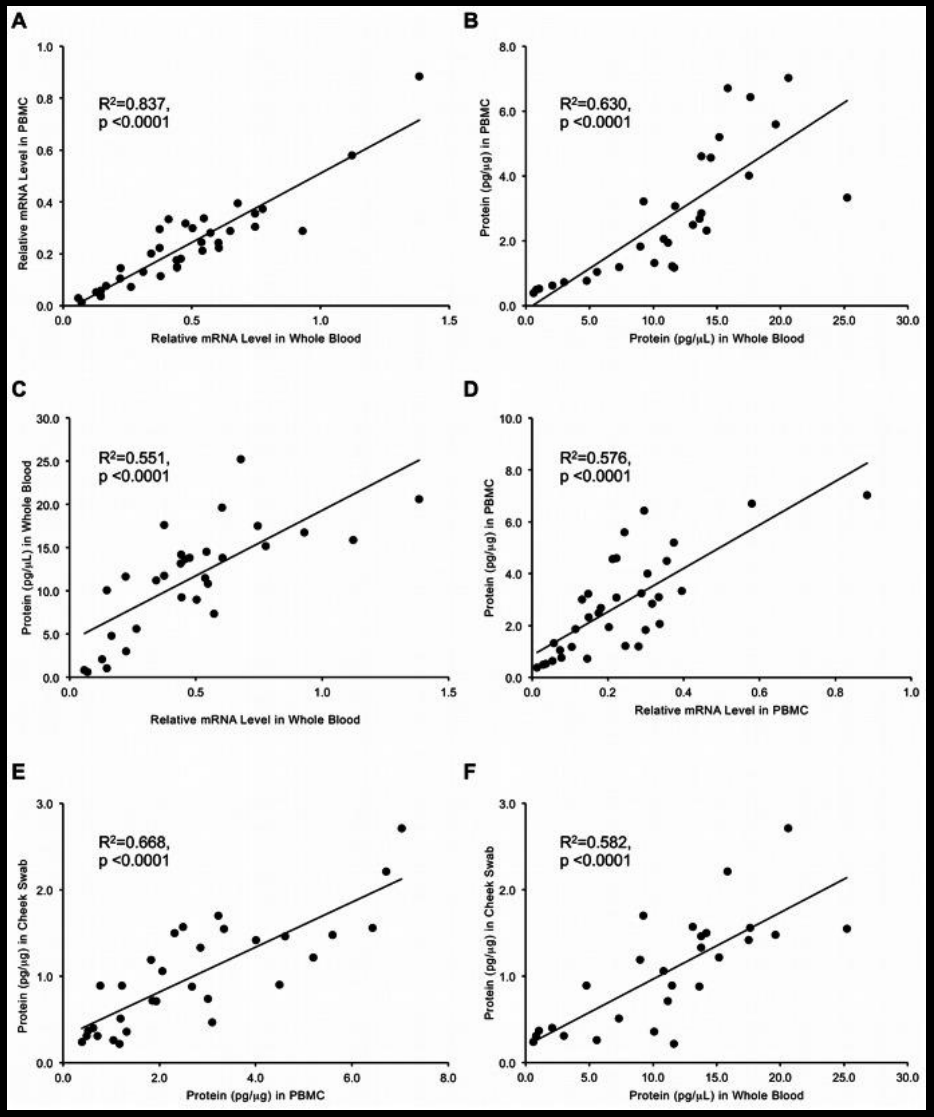
Characteristic	Value
Demographics	
Age, yr[a]	30.0±8.1
Sex, No. (%)	
M	9 (40.9)
F	13 (59.1)
Disease data	
GAA•TTC triplet expansion on shortest allele[a]	1,084.8±784.5
Age of onset, yr[a]	10.7±4.6
FARS[b] score at screening[a]	59.7±23.2
Cardiac function, ejection fraction %[a]	63.0±6.9

[a] Data are shown as the mean±standard deviation.

[b] See Beconi et al.[45]

F=female; FARS=Friedreich Ataxia Rating Scale; M=male.

Frataxin protein and mRNA levels strongly correlated in blood, PBMCs and buccal cells



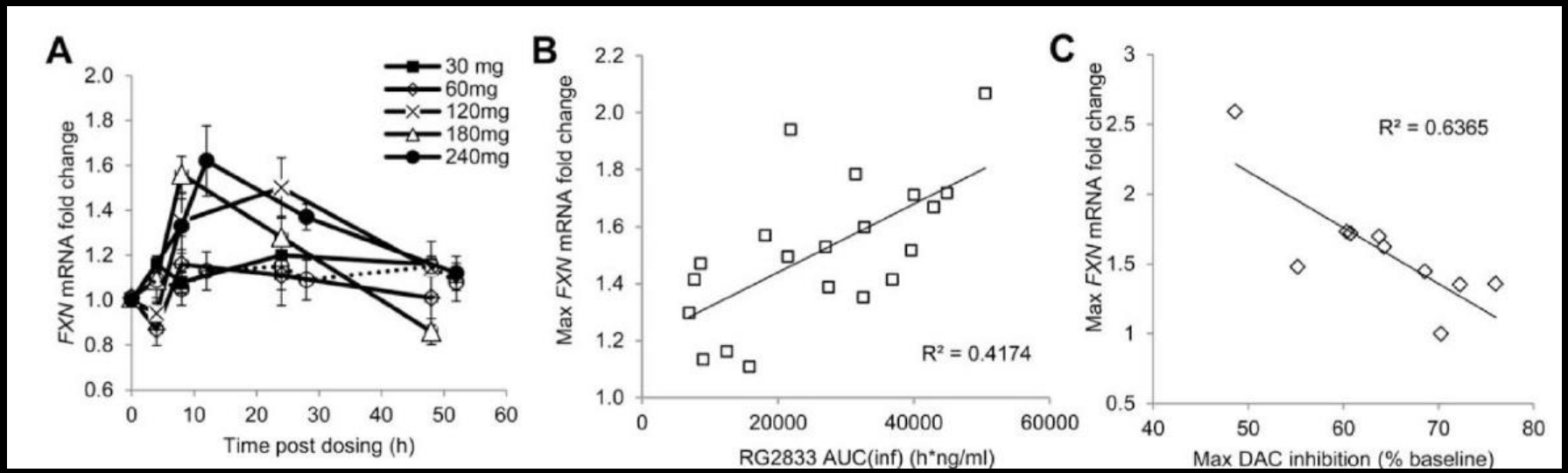
PBMCs vs. whole blood

Frataxin mRNA vs. protein

PBMCs vs. buccal cells
buccal cells vs. whole blood

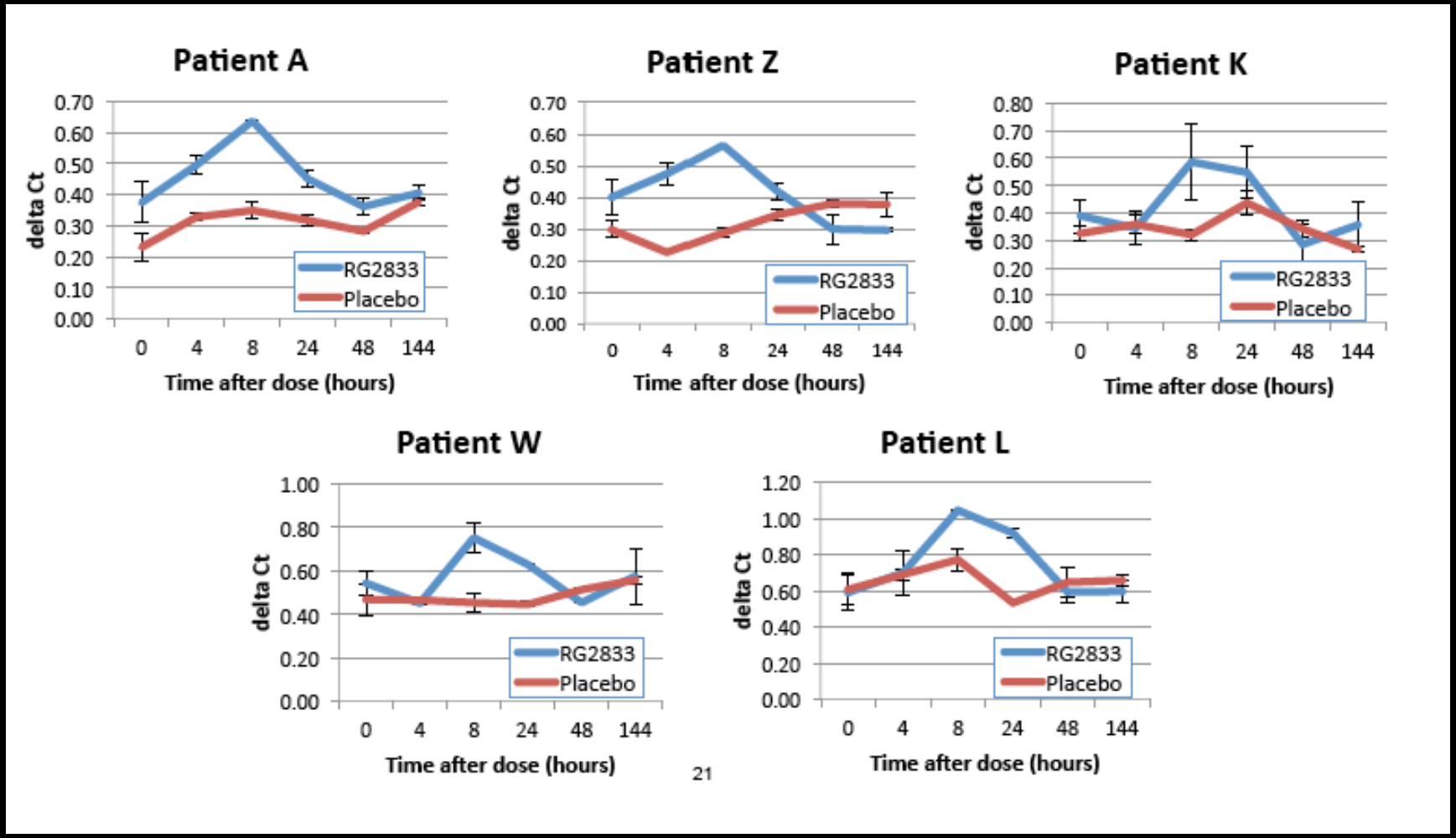
Plasterer et al., PLoS One. 2013; 8(5): e63958.

FXN mRNA in adult Friedreich ataxia patients after oral administration of RG2833/109



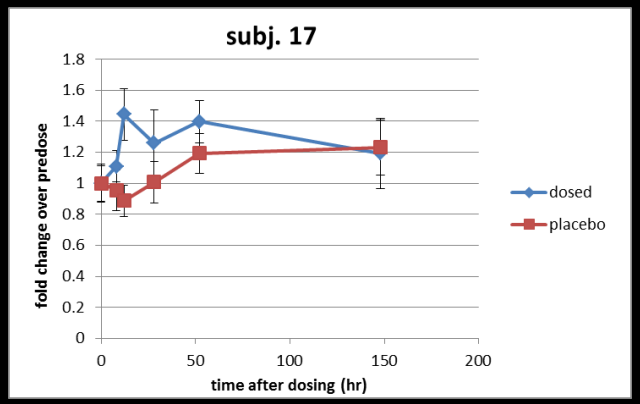
Soragni et al., Ann Neurol. 2014; 76(4): 489–508

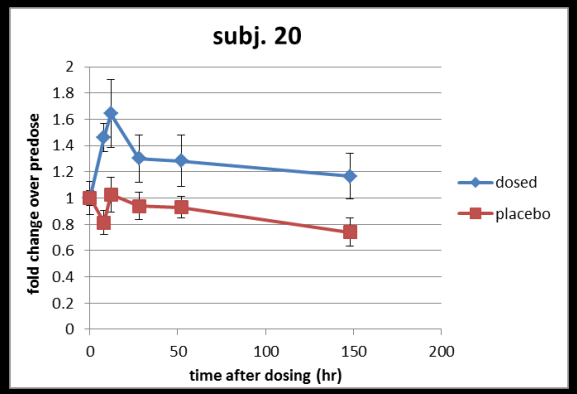
Increases in FXN mRNA in PBMCs from 5 patients after a single dose (180 mg) of RG2833

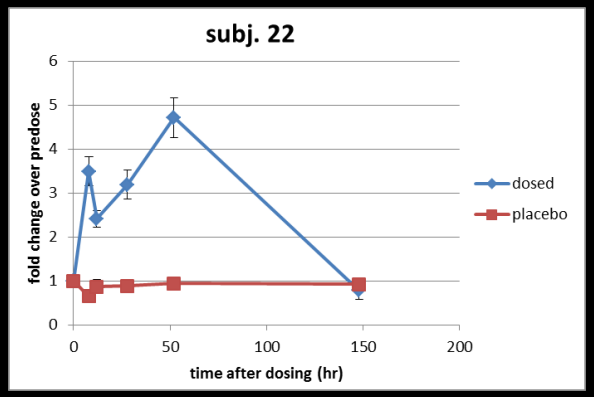


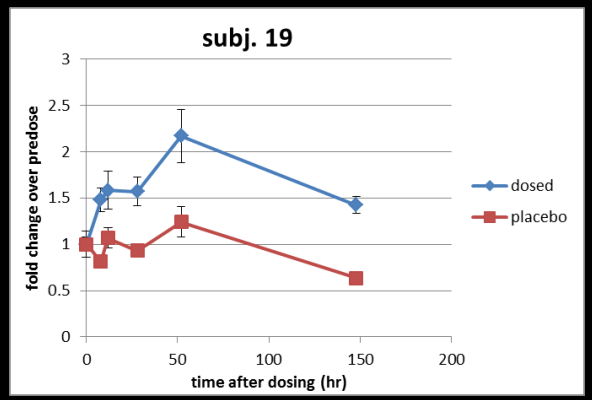
Increases in FXN mRNA observed in 9/10 patients overall

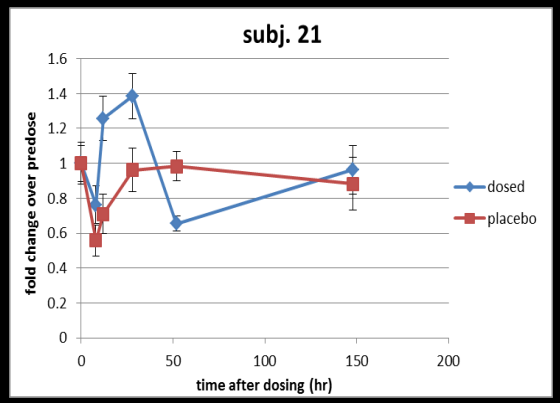
Increases in histone H3K9 acetylation in patient PBMCs after a single dose (180 mg) of RG2833





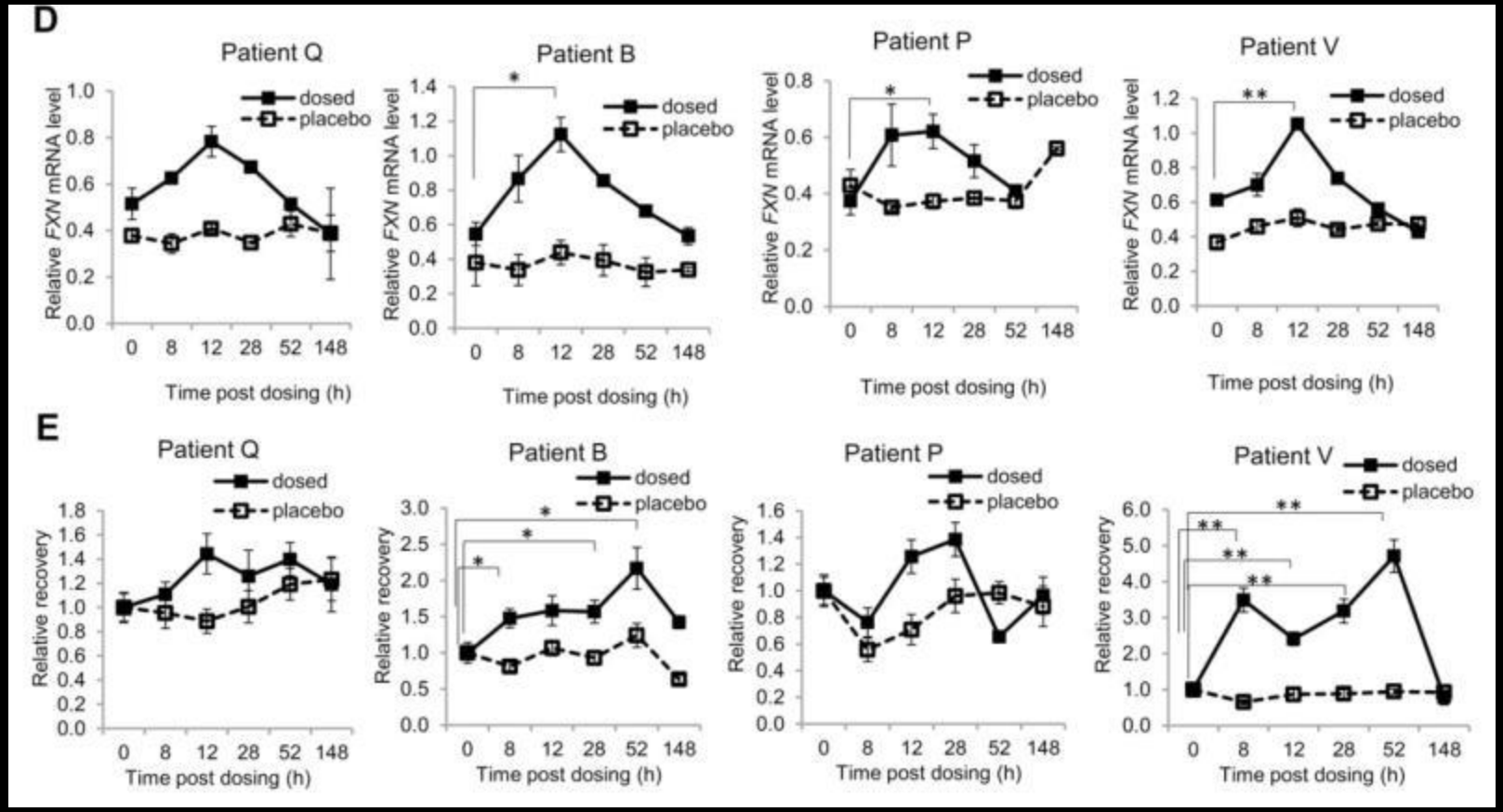






Liz Soragni/TSRI

Increases in FXN mRNA and histone H3K9 acetylation in patient PBMCs after two doses (120 mg) of RG2833



Soragni et al., Annals of Neurology. 2014; 76(4):489-508

Summary

Preclinical studies from mice:

- Show good efficacy of HDAC1/3-targeting inhibitors in HD mouse models.

- HDACi 4b treatment is associated with a reversal of histone hypoacetylation at H3K9 at the promoter of several candidate genes.

- DNA methylation may also prove useful as a marker of drug response.

In-patient clinical studies:

-Treatment with RG2833 was safe and well tolerated (at 180 mg or 120 mg twice a day) and no drug related adverse effects were reported.

- Frataxin mRNA was increased in 9/10 patients and expression levels in different cell types were correlated.

- ChIP promoter histone acetylation was increased in patient PBMCs after single and multiple doses. H3K9 acetylation is a useful epigenetic biomarker for drug response.

Second generation compounds with improved brain penetration and metabolic stability have been generated. A clinical candidate from the new generation of compounds will be taken forward for IND filing for a second round of clinical trials.

Acknowledgments

The Scripps Research Institute

Thomas Lab

Alaina Aikin
Haiqun Jia
Bin Tang
Crystal Wang

Joel M. Gottesfeld

Chunping Xu
Liz Soragni
David Herman

Jeanne Loring

Roy Williams
Kit Nazor

University of California, Irvine

J. Lawrence Marsh
Leslie Thompson

University of California, Los Angeles

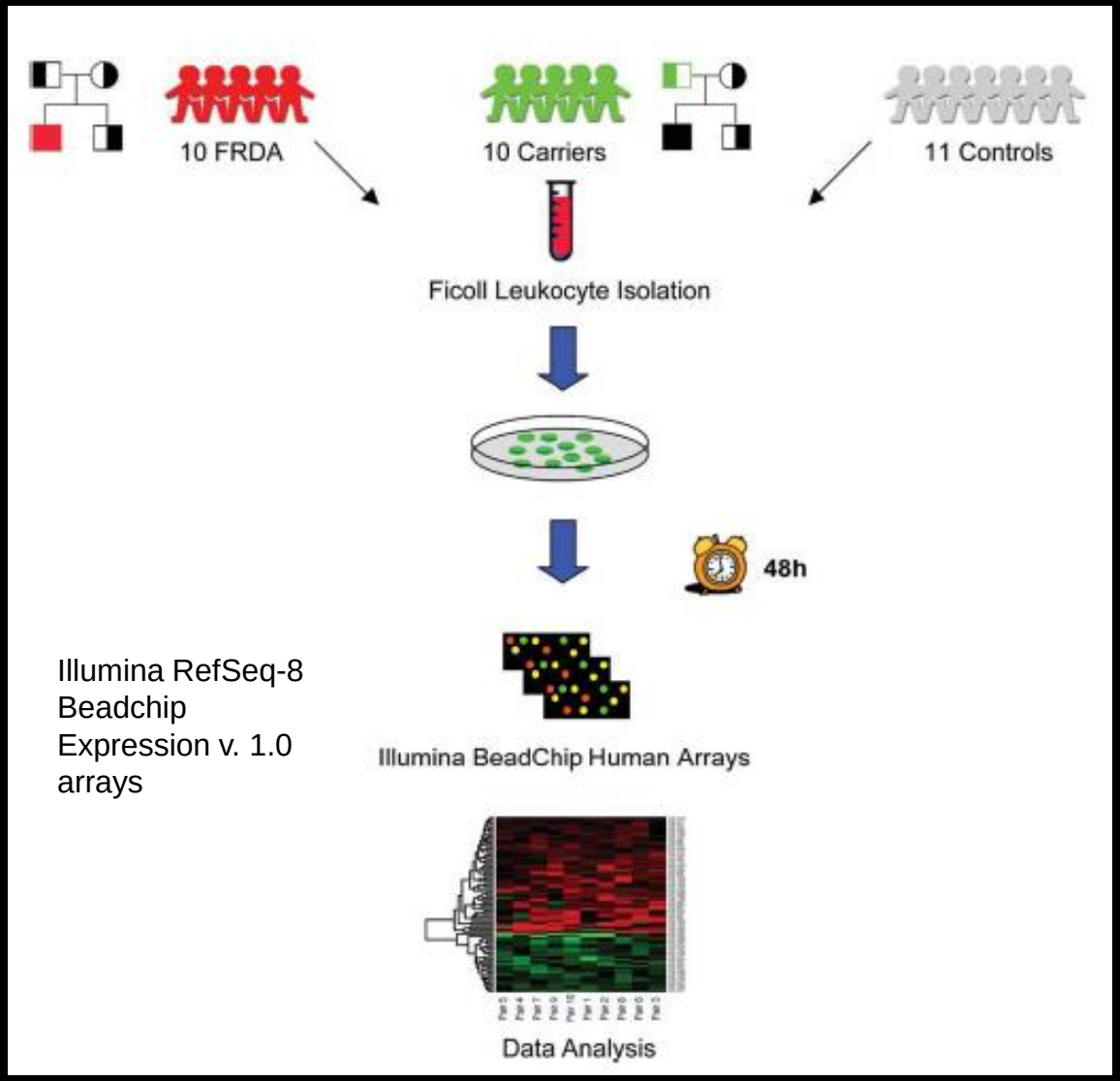
Daniel Geschwind
Giovanni Coppola

Massachusetts General Hospital

Ghazaleh Sadri-Vakili

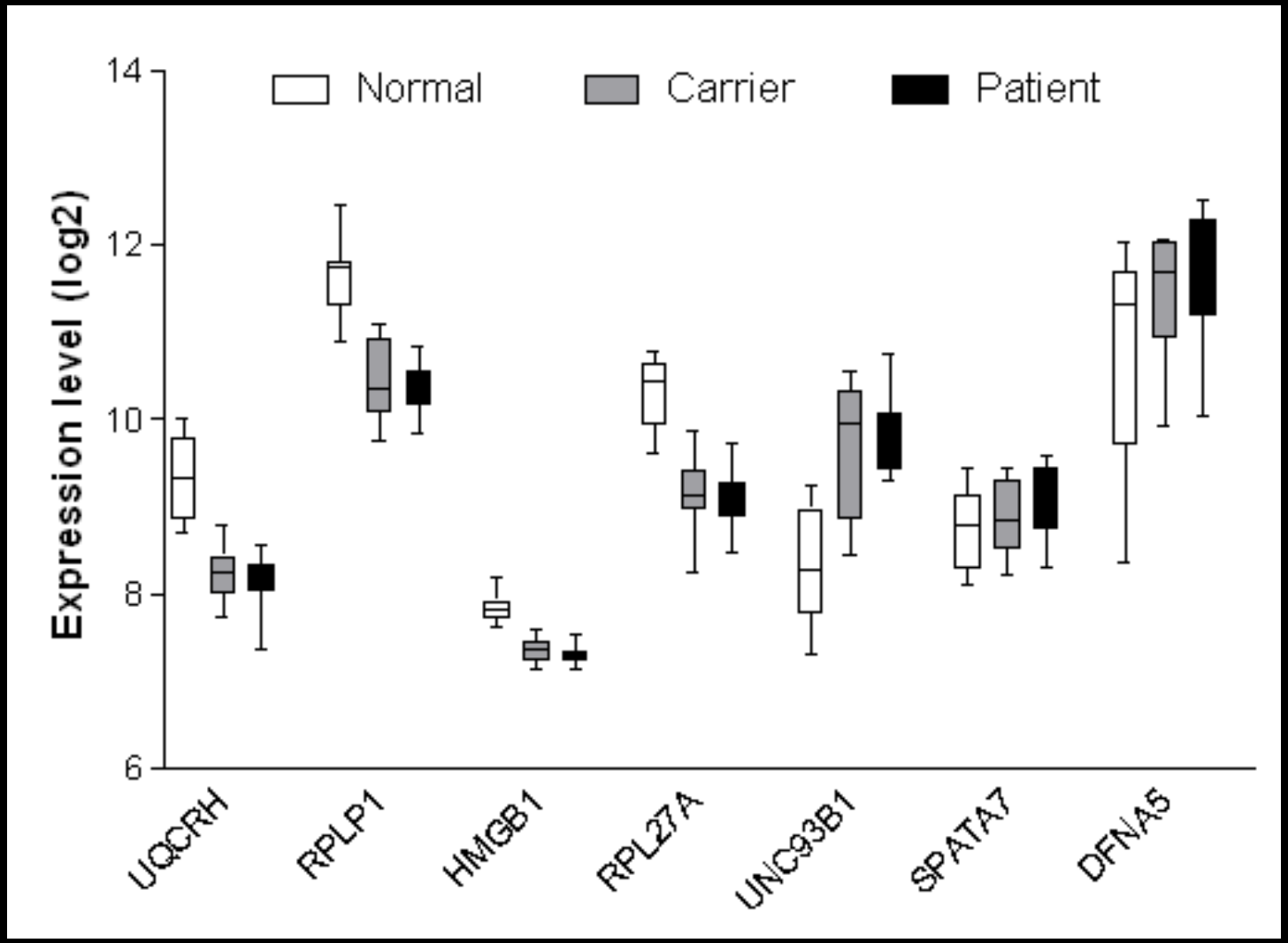
With support from :

Microarray gene expression analysis



Illumina RefSeq-8 Beadchip Expression v. 1.0 arrays

Using gene expression data for biomarker identification in PBMCs



Unpublished