UNIVERSITEIT GENT

Center for Medical Genetics

BCL2 inhibition by ABT-199 in T-cell acute lymphoblastic leukemia (T-ALL)

Pieter Van Vlierberghe

Bioluminescent Cell-based Assay Seminar Day
Monday 31 march 2014
UCL De Duve institute
Brussels, Belgium

Short Bio sketch

PhD 2003-2008
Pediatric Oncology
Erasmus Medical Center
Rotterdam, The Netherlands

Postdoc 2008-2013
Institute for Cancer Genetics
Columbia University Medical Center
New York, USA

Short Biosketch

COLUMBIA UNIVERSITY MEDICAL CENTER

Discover. Educate. Care. Lead.

Turner BioSystems Modulus® II Microplate Multimode Reader

Short Bio sketch

Odysseus type II grant
2013-2017
Center for Medical Genetics
Ghent University

GloMax®-Multi
Microplate Reader

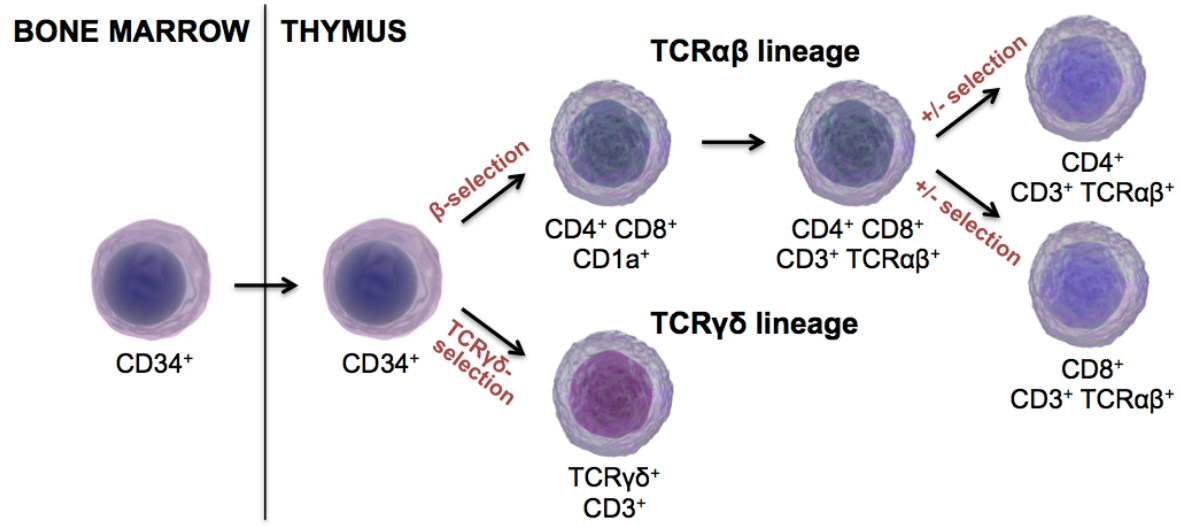
CellTiter-Glo®
Luminescent Cell Viability

Caspase-Glo® 3/7
Apoptosis Assay

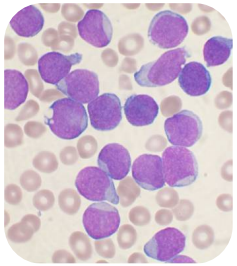
BCA protein assays

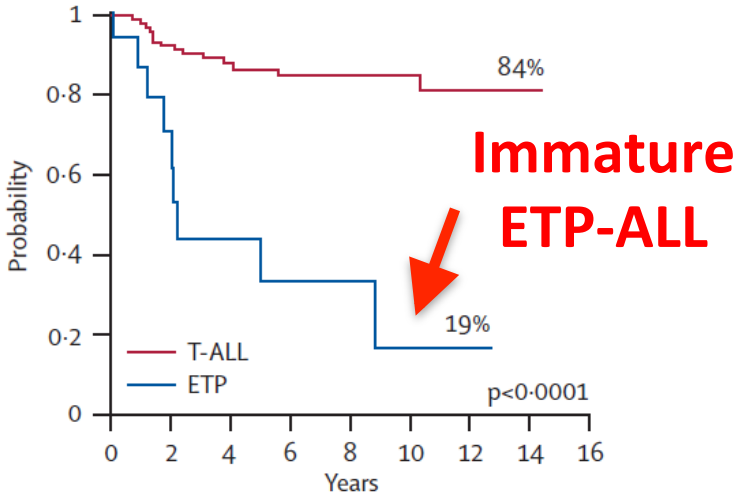
Luciferase assays
miRNA-mRNA regulation

From normal development to malignant transformation



Early T-cell precursor ALL (ETP-ALL)

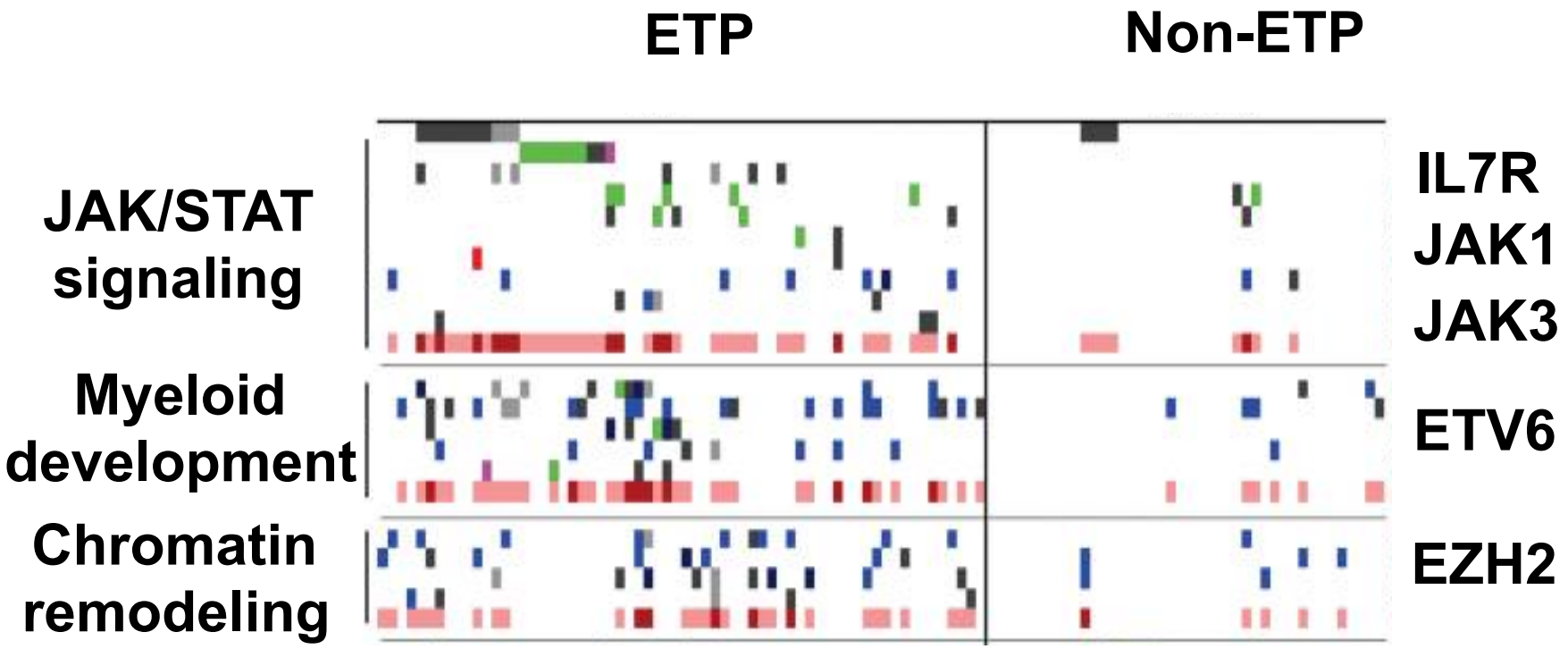




Immature ETP-ALL

Coustan-Smith *et al.*, Lancet Oncology, 2009

Genetic heterogeneity in human ETP-ALL

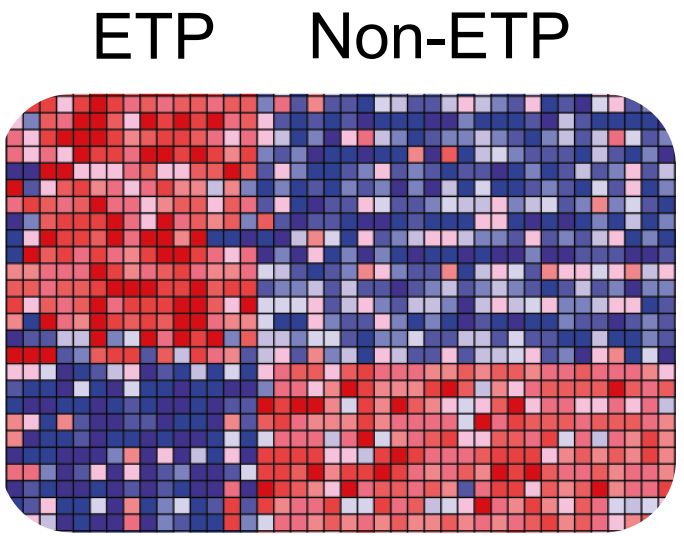


Genetic heterogeneity provides challenges for the implementation of a uniform targeted therapy in ETP-ALL

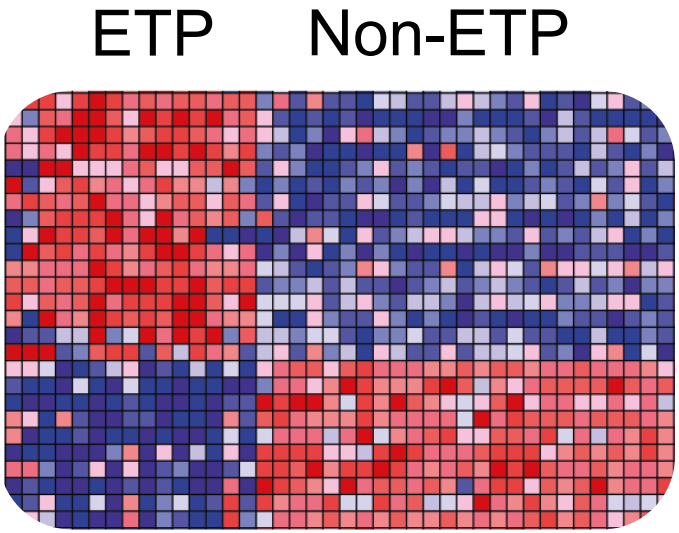
Zhang et al., Nature 2012

Unique features of human ETP-ALL

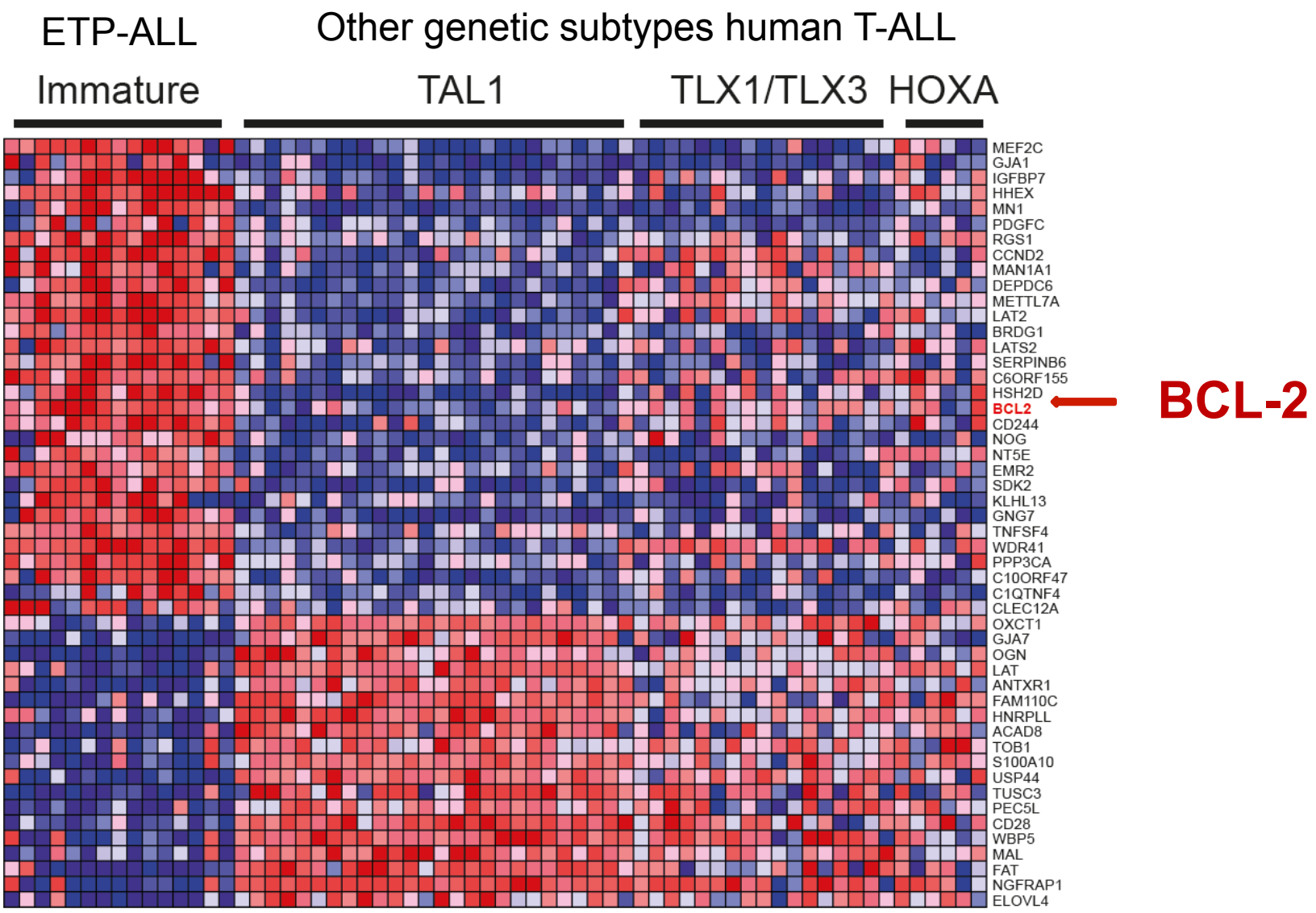
Human ETP-ALLs
have a unique
gene expression signature



Therapeutic targets in human ETP-ALL gene expression profile

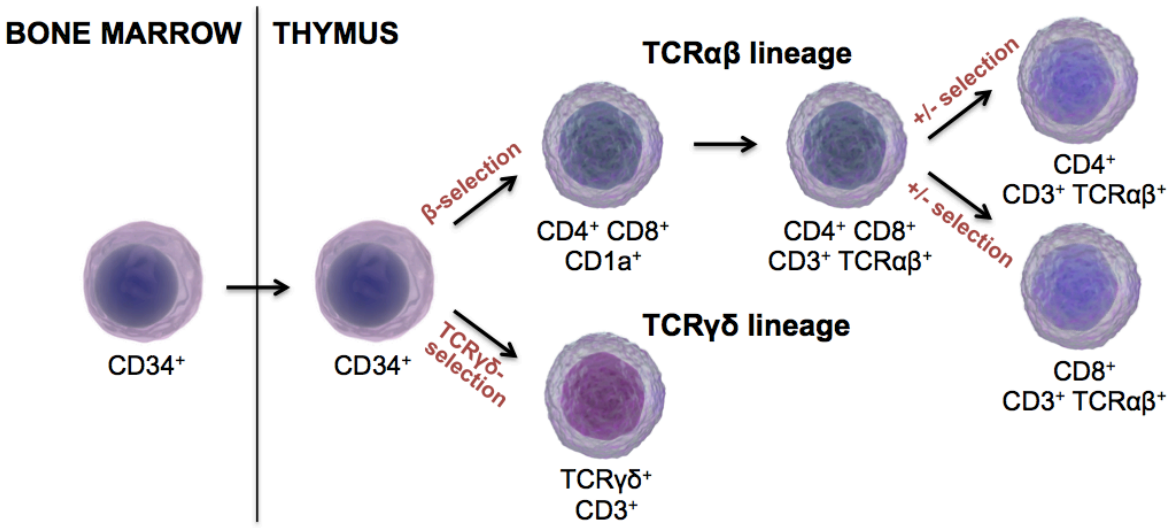


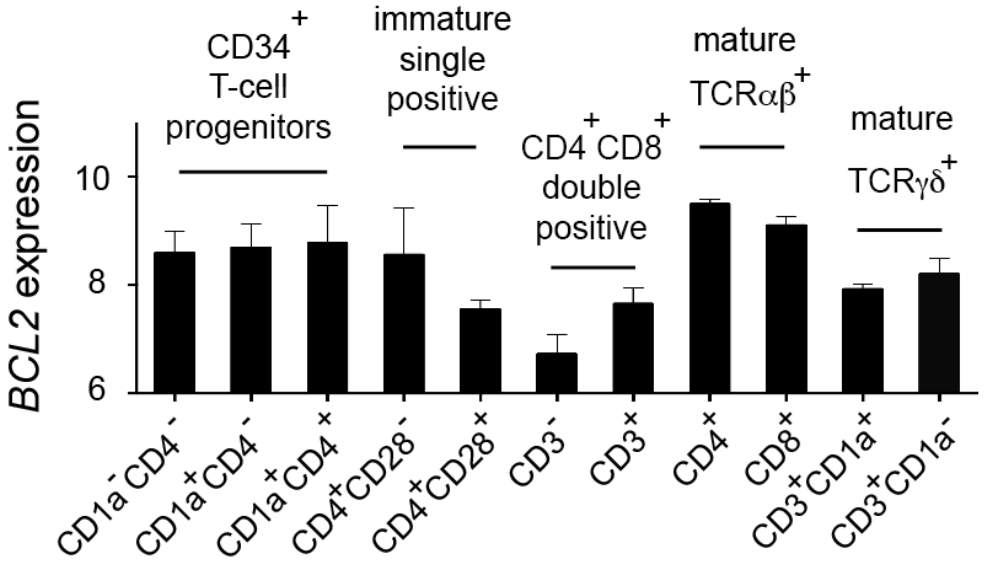
High *BCL2* expression in human ETP-ALL

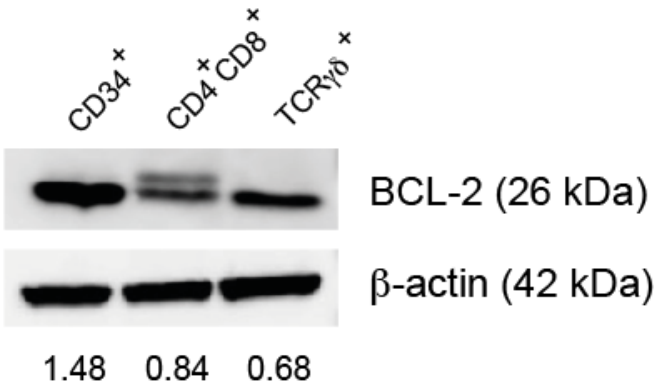


Fold change > 2; p<0.05

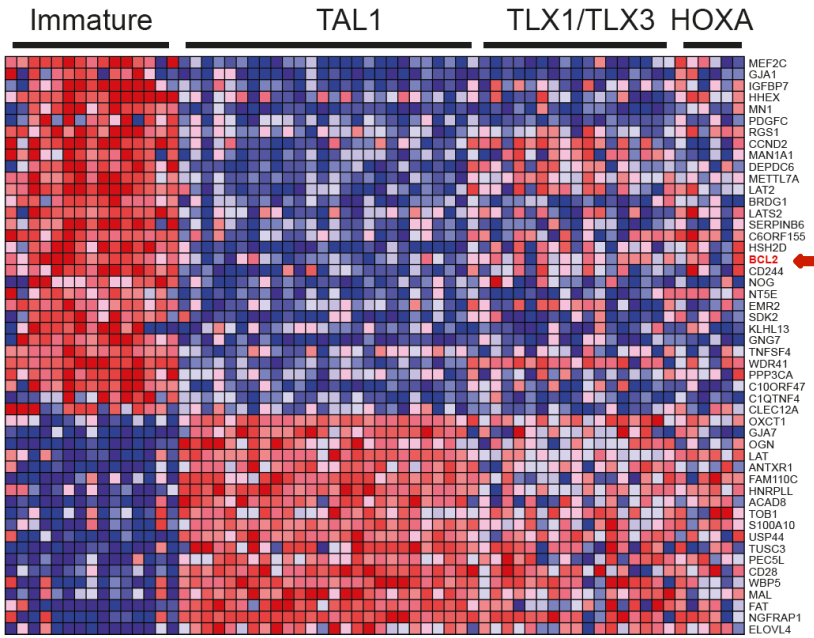
High *BCL2* expression a reflection of T cell differentiation?



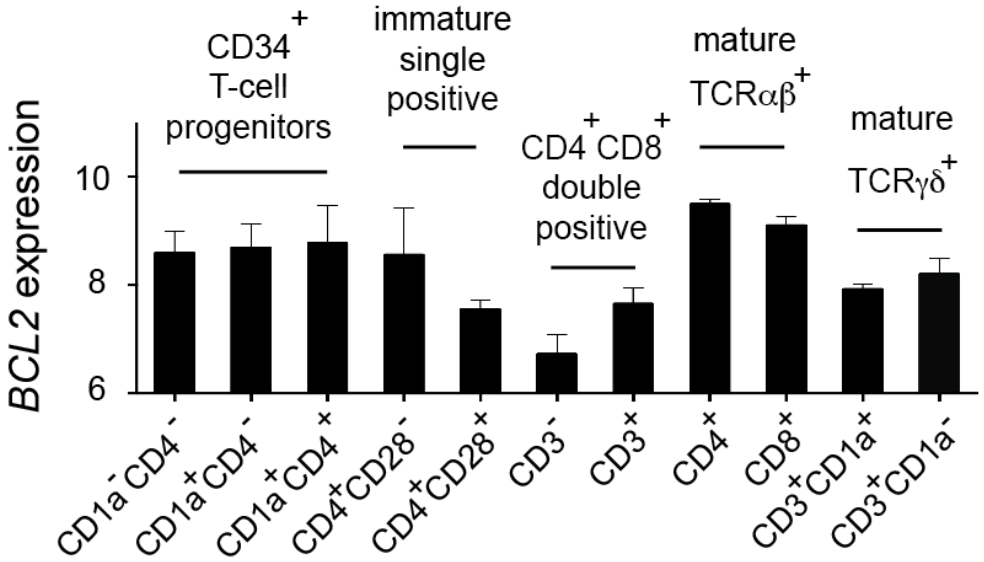


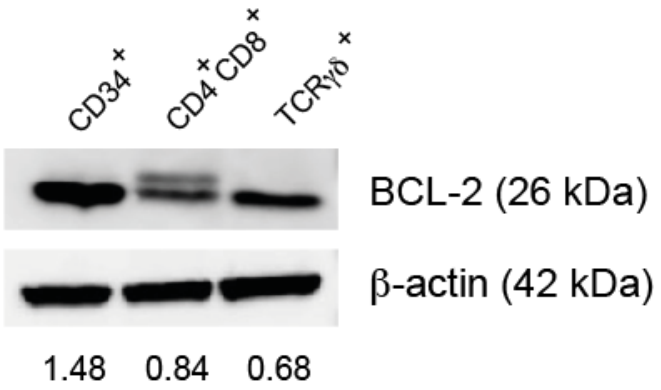


High *BCL2* expression a reflection of T cell differentiation?

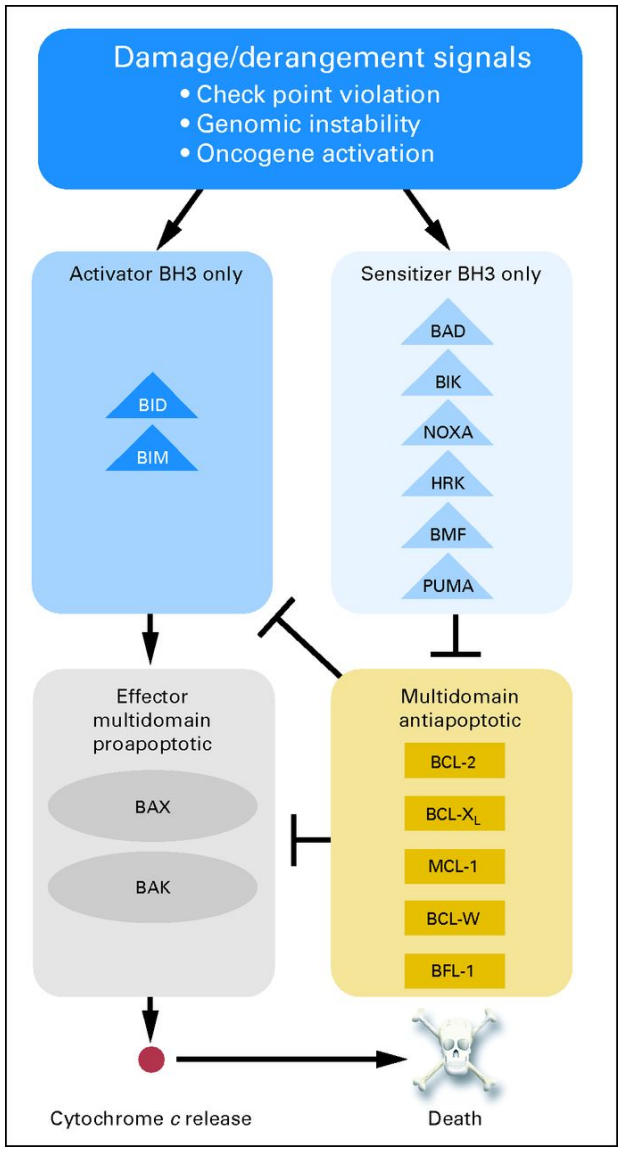


BCL-2





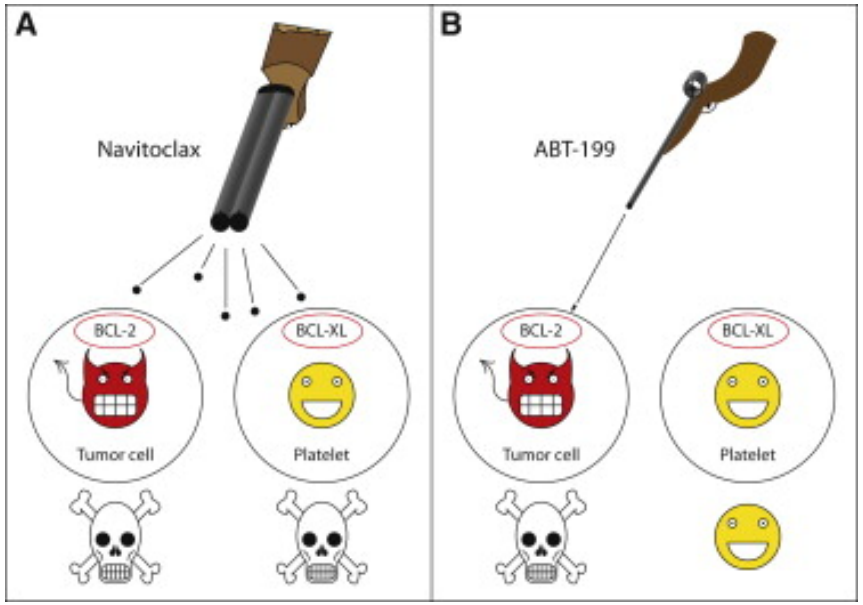
BCL2 inhibition as a therapeutic strategy



Davids & Letai, Journal of Clinical Oncology 2013

abbvie

ABT-263 (Navitoclax)	:	BCL-2, BCL-xL
ABT-199	:	BCL-2 only



Davids & Letai, Cancer Cell 2013

ABT-199 as a new therapeutic strategy in human cancer

nature medicine

January 2013

ABT-199, a potent and selective BCL-2 inhibitor, achieves antitumor activity while sparing platelets

Andrew J Souers[1], Joel D Leverson[1], Erwin R Boghaert[1], Scott L Ackler[1], Nathaniel D Catron[1], Jun Chen[1], Brian D Dayton[1], Hong Ding[1], Sari H Enschede[1], Wayne J Fairbrother[2], David C S Huang[3,4], Sarah G Hymowitz[2], Sha Jin[1], Seong Lin Khaw[3,4], Peter J Kovar[1], Lloyd T Lam[1], Jackie Lee[2], Heather L Maecker[2], Kennan C Marsh[1], Kylie D Mason[3–5], Michael J Mitten[1], Paul M Nimmer[1], Anatol Oleksijew[1], Chang H Park[1], Cheol-Min Park[1,7], Darren C Phillips[1], Andrew W Roberts[3–5], Deepak Sampath[2], John F Seymour[4,6], Morey L Smith[1], Gerard M Sullivan[1], Stephen K Tahir[1], Chris Tse[1], Michael D Wendt[1], Yu Xiao[1], John C Xue[1], Haichao Zhang[1], Rod A Humerickhouse[1], Saul H Rosenberg[1] & Steven W Elmore[1]

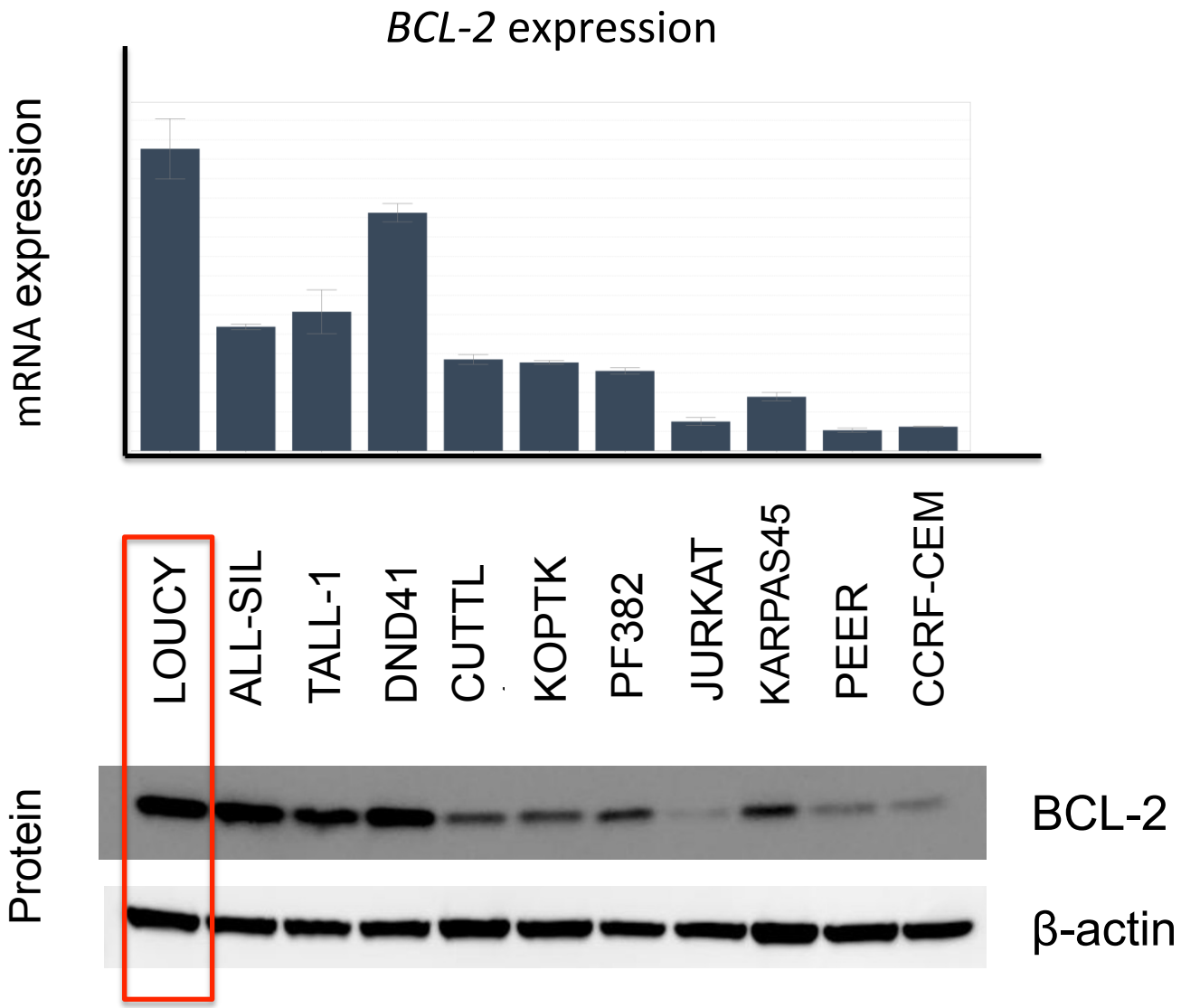
Clinical Phase I Trials in relapsed or treatment-resistant CLL

Tumor Lysis Syndrome

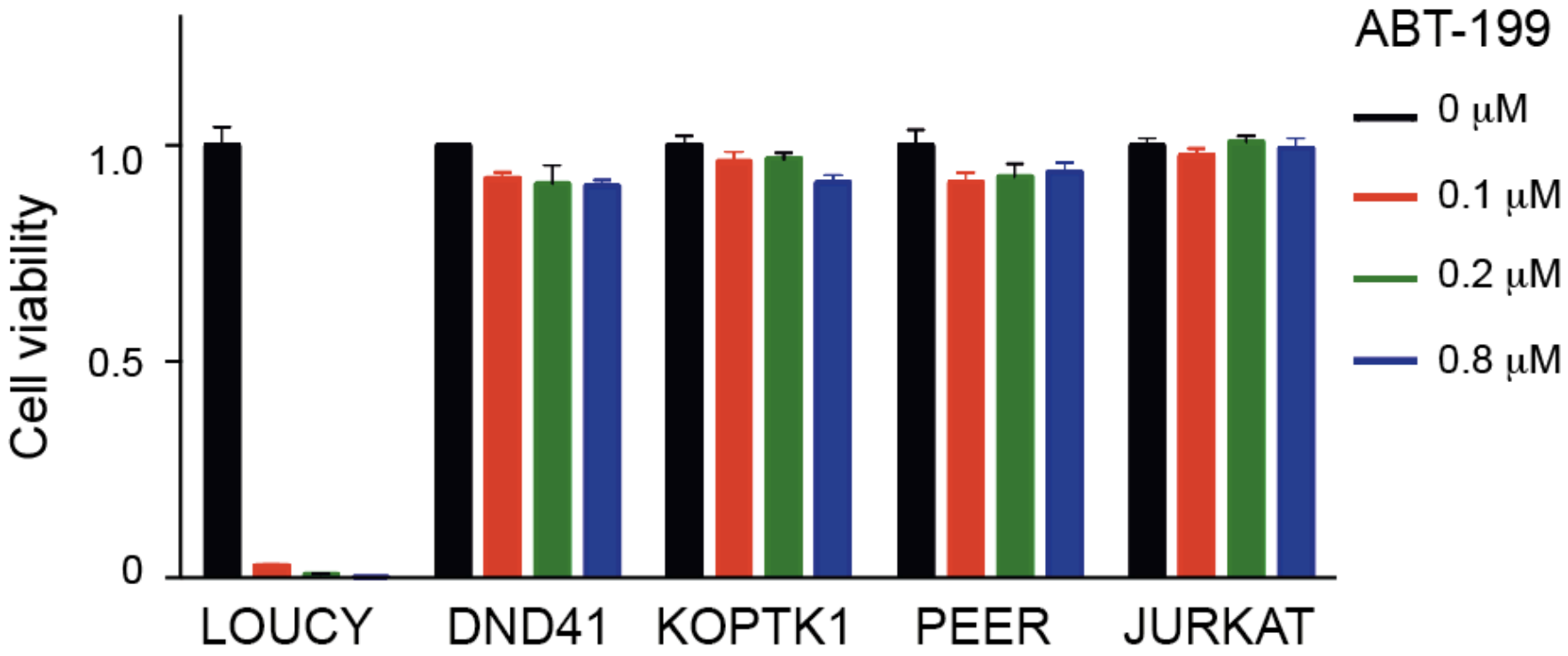
Reports on anti-tumoral effect in:

acute myeloid leukemia
multiple myeloma
estrogen receptor-positive breast cancer

ABT-199 expression in human T-ALL cell lines



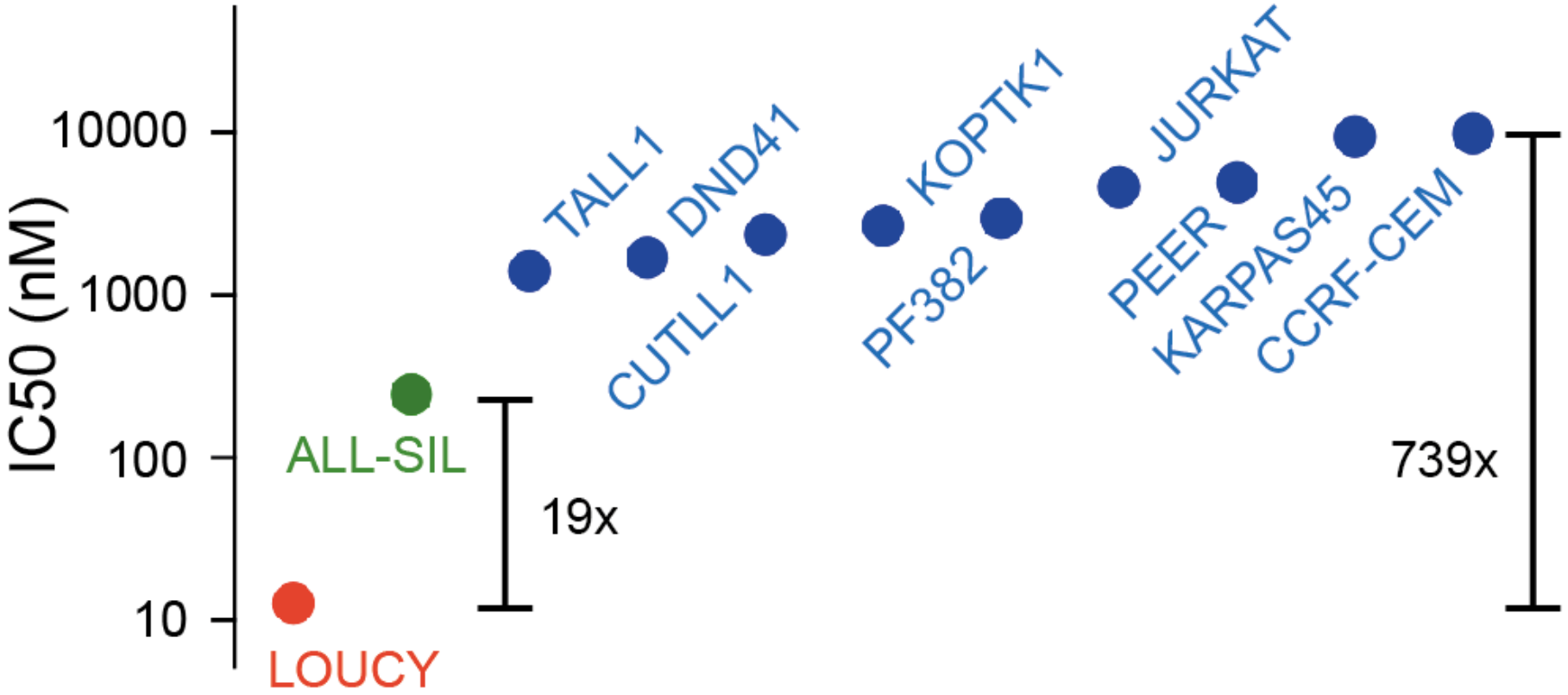
ABT-199 and cell viability in human T-ALL cell lines



CellTiter-Glo® Luminescent Cell Viability

GloMax®-Multi Microplate Reader

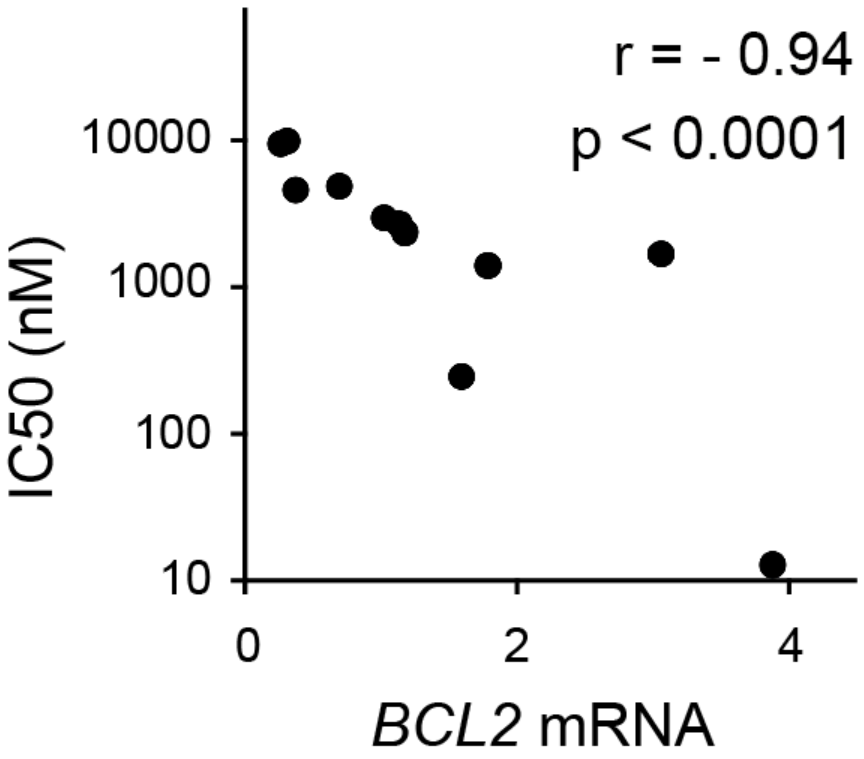
ABT-199 IC50 in human T-ALL cell lines

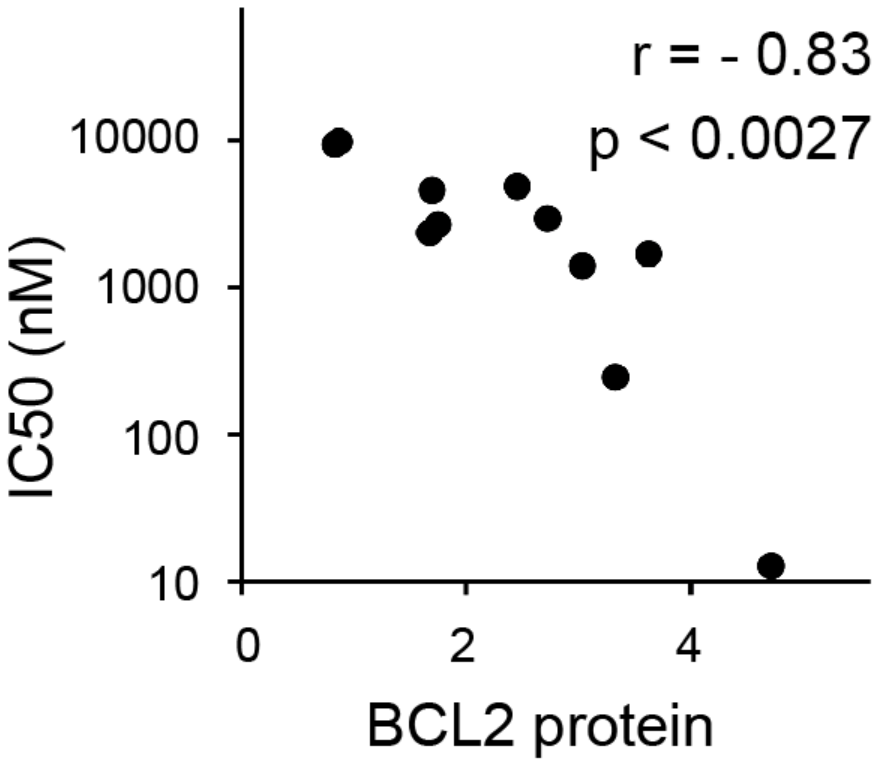


CellTiter-Glo® Luminescent Cell Viability

GloMax®-Multi Microplate Reader

Correlation ABT-199 IC50 and BCL2 expression

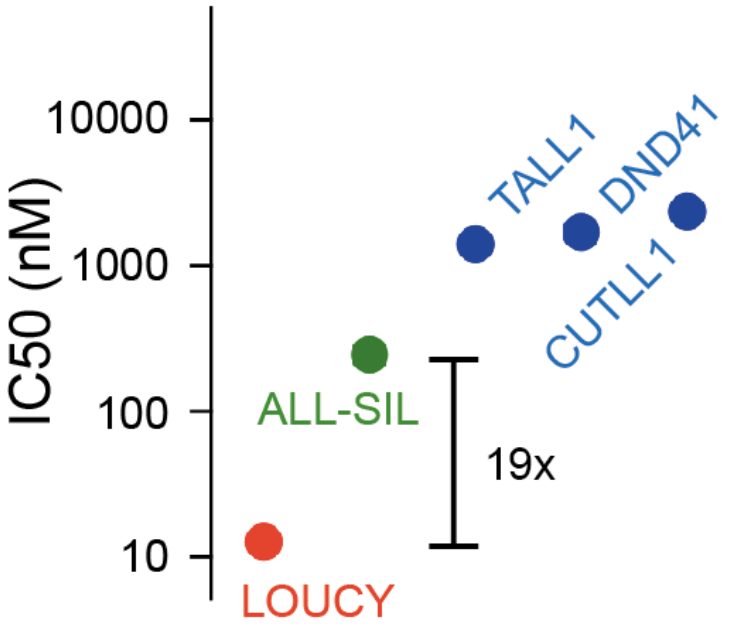


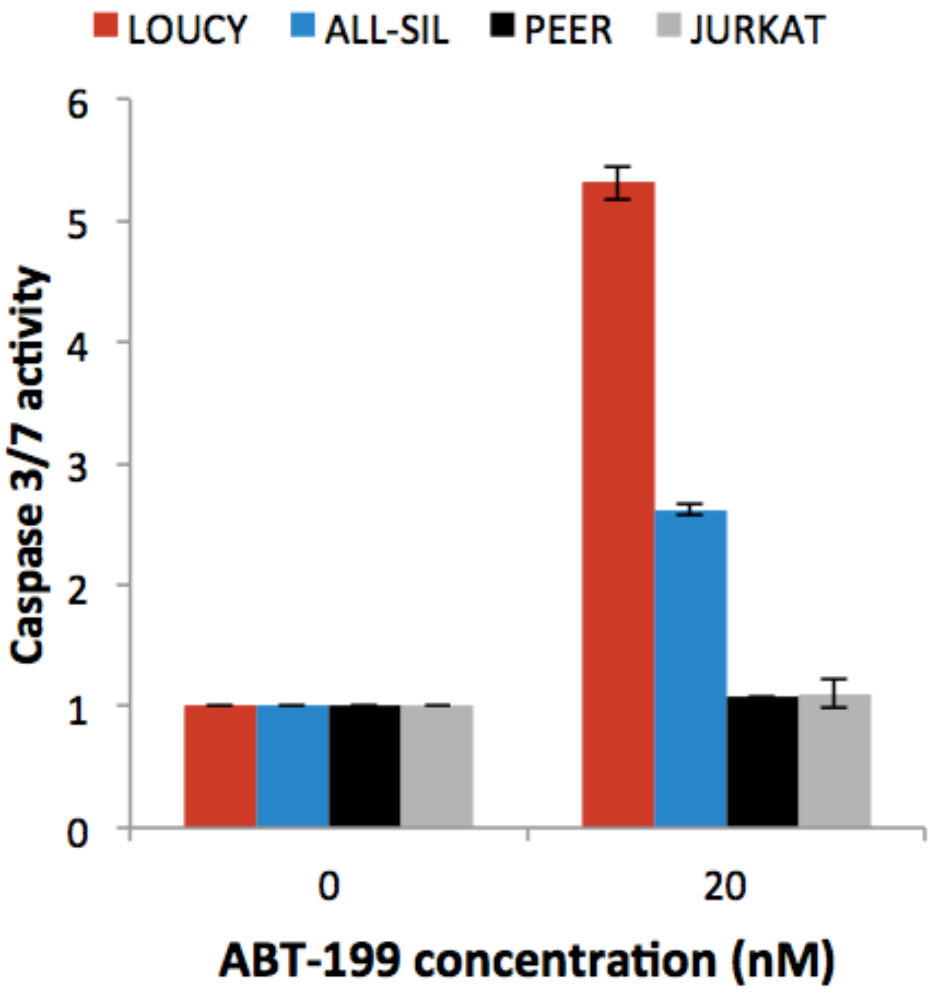


CellTiter-Glo® Luminescent Cell Viability

GloMax®-Multi Microplate Reader

ABT-199 induces apoptosis



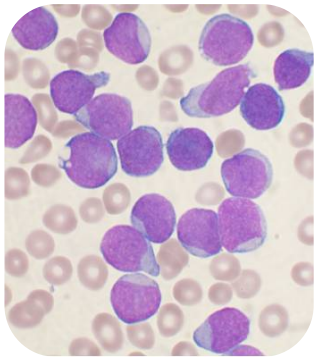


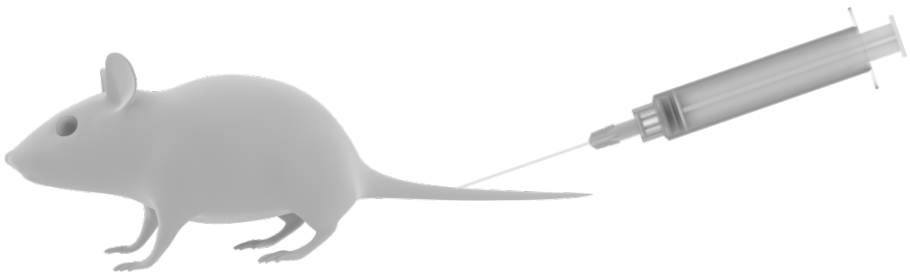
Caspase-Glo® 3/7 Apoptosis Assay

GloMax®-Multi Microplate Reader

In vivo xenograft model human T-ALL

Luciferase positive
LOUCY cells



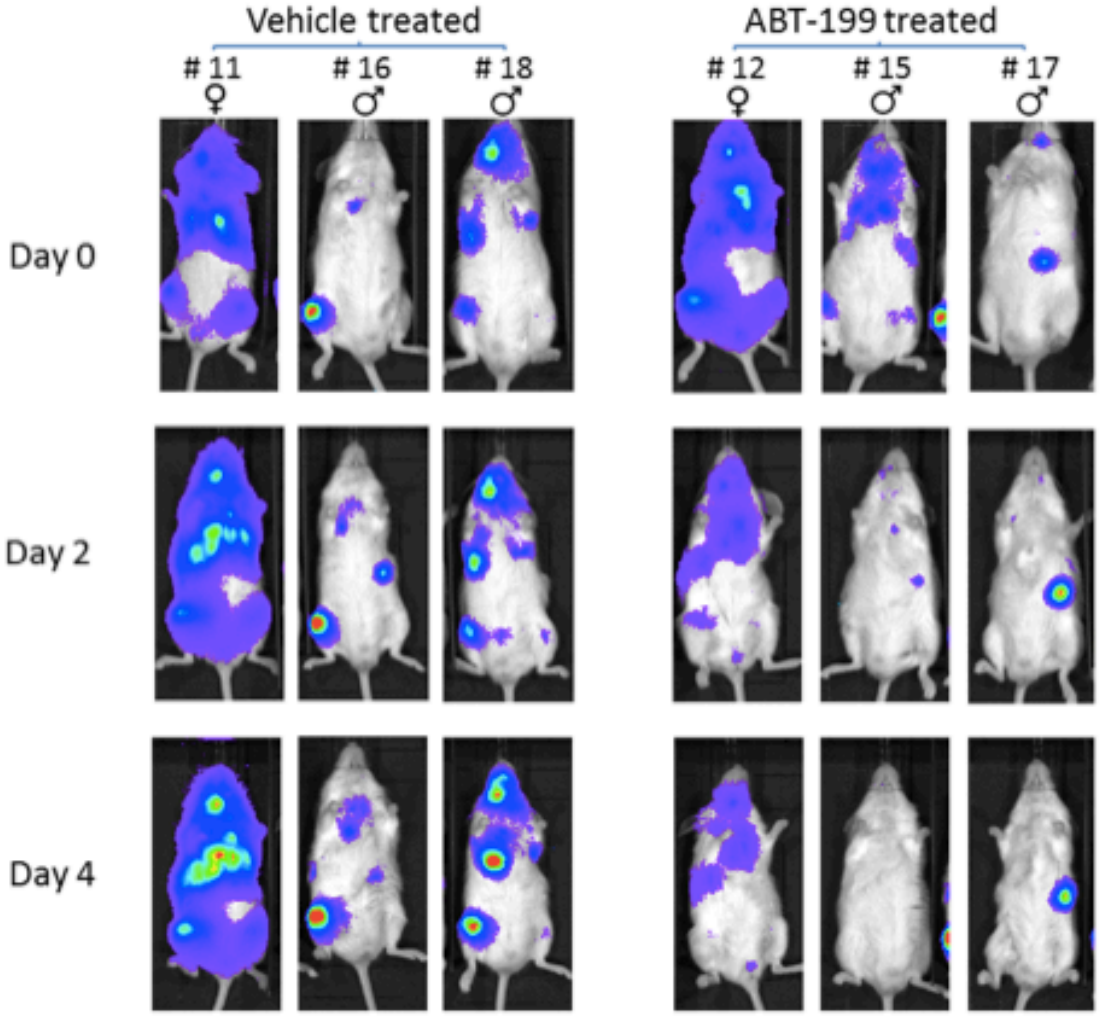


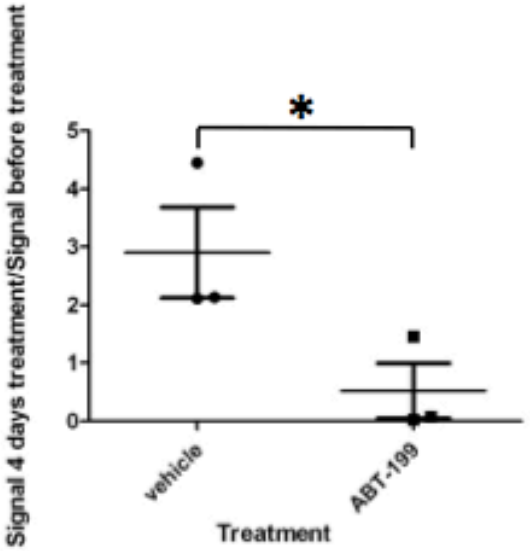
Engraftment of cells
(6 weeks)

Measure luminescence on
different time points

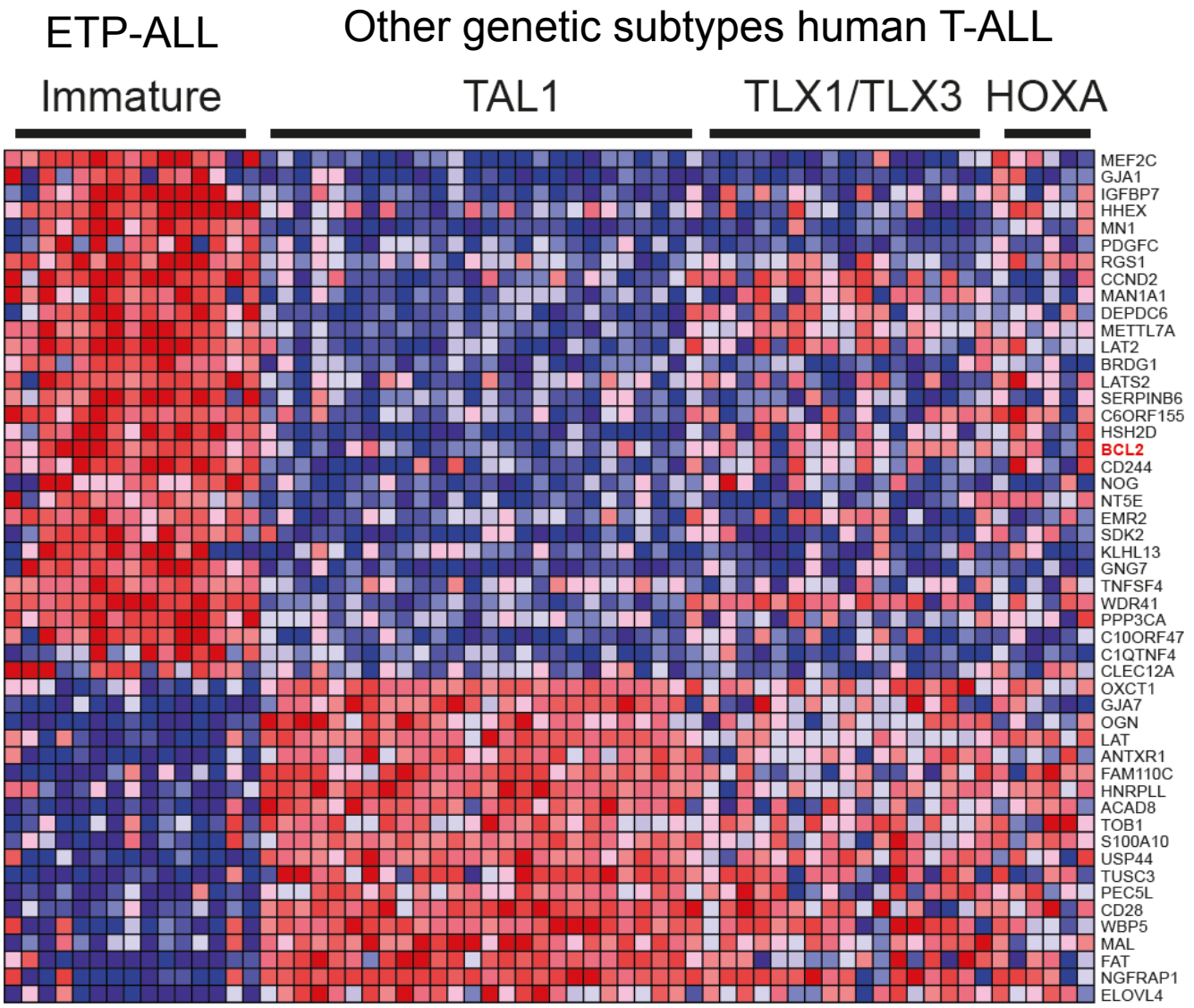
Oral gavage
100 mg ABT-199/kg
or vehicle

In vivo xenograft model human T-ALL

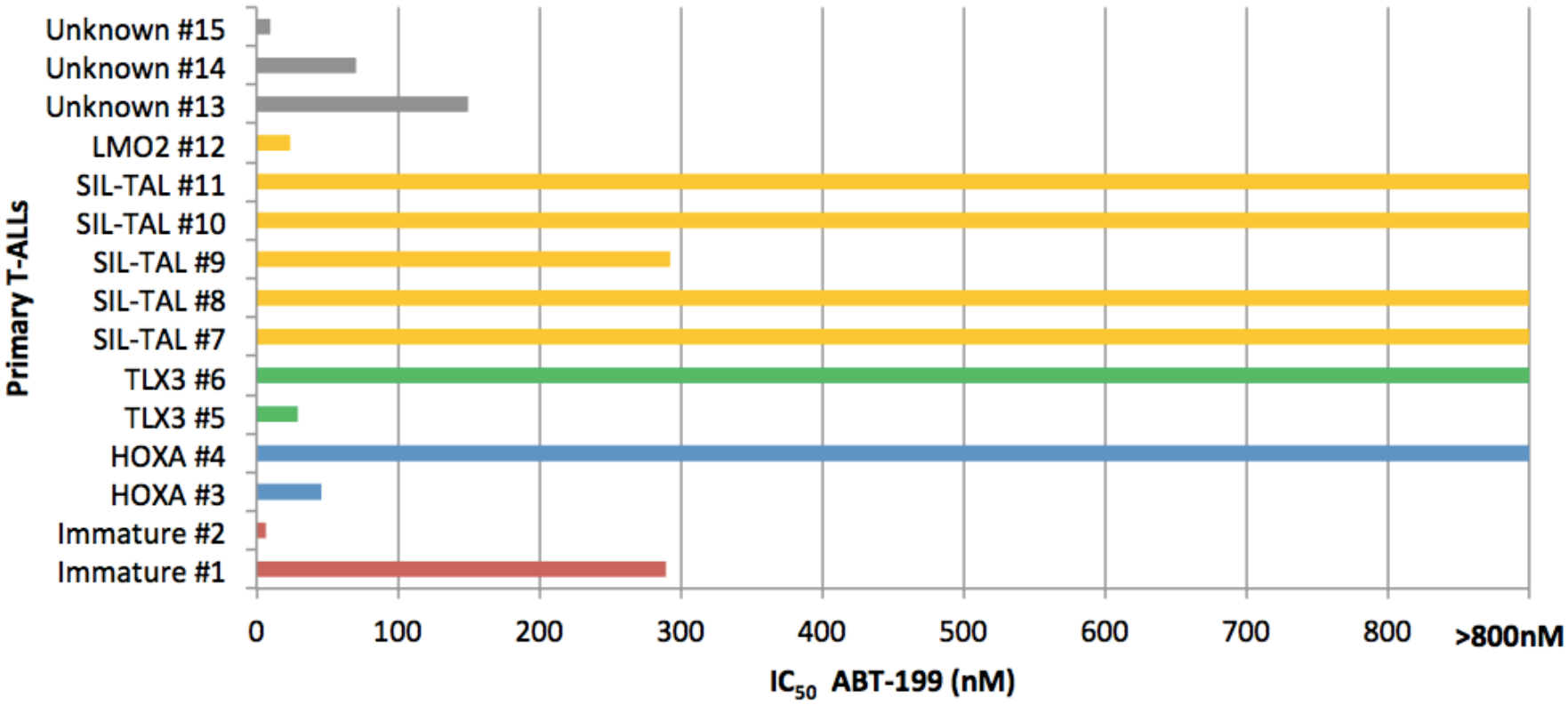




Genetic subtypes of human T-ALL



In vitro ABT-199 sensitivity primary T-ALL samples

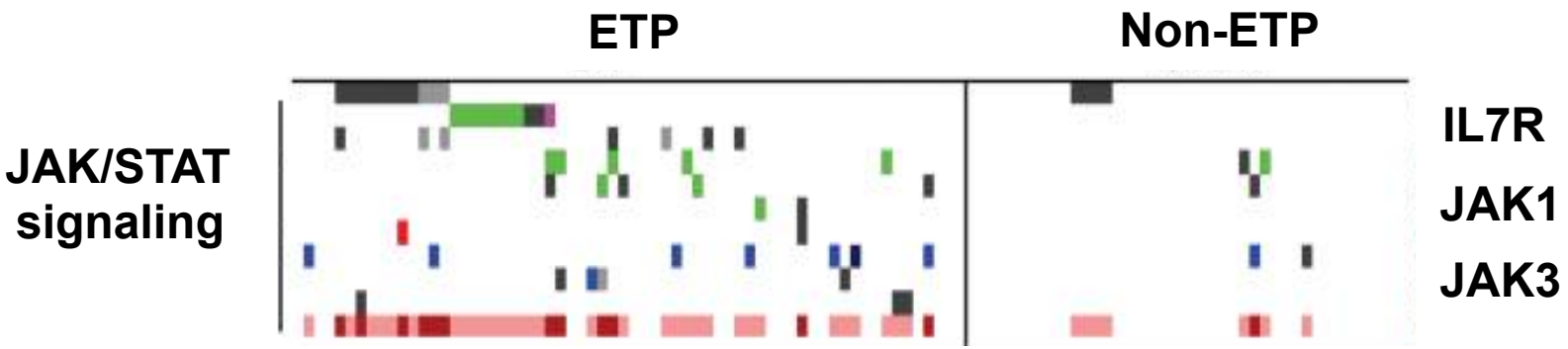


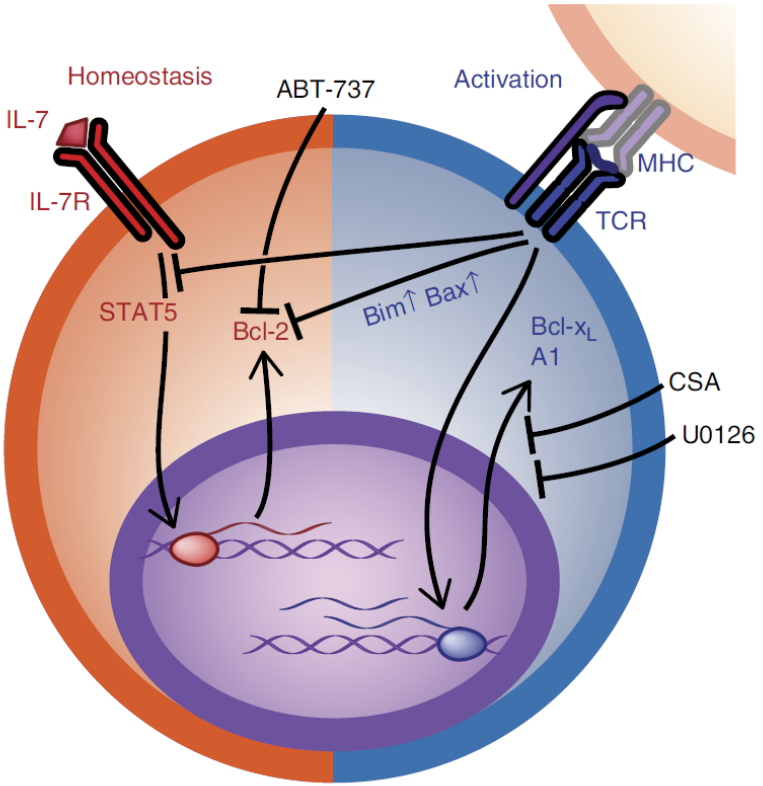
Hypothesis
The cell of origin and cooperative genetic defects define ABT-199 sensitivity in T-ALL

CellTiter-Glo® Luminescent Cell Viability

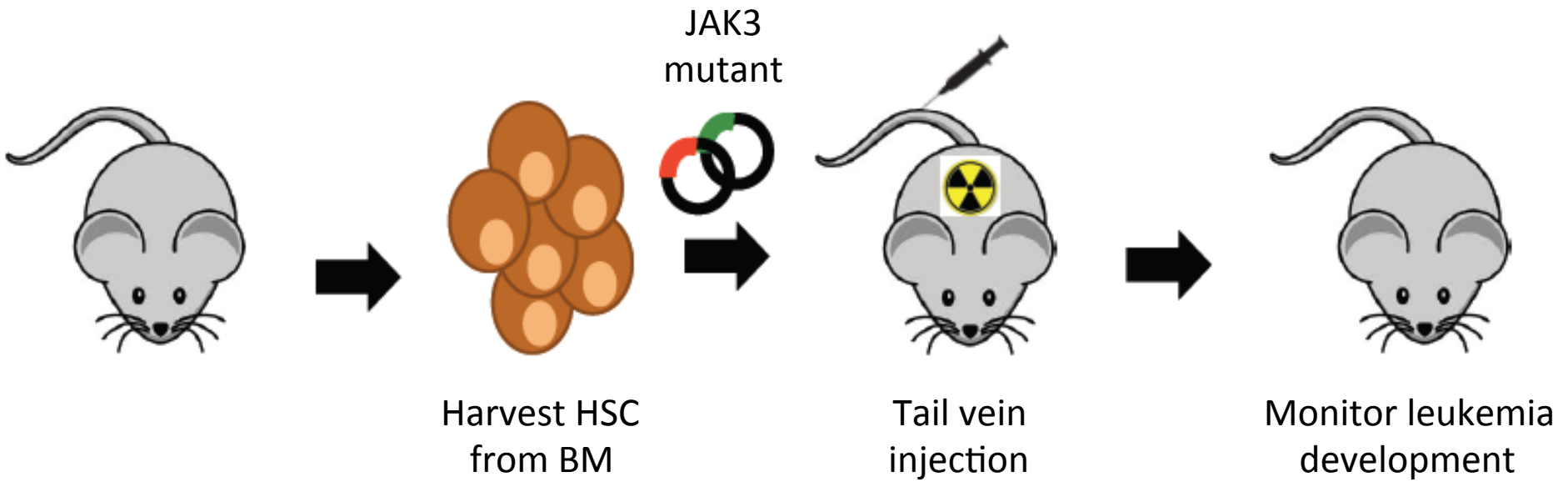
GloMax®-Multi Microplate Reader

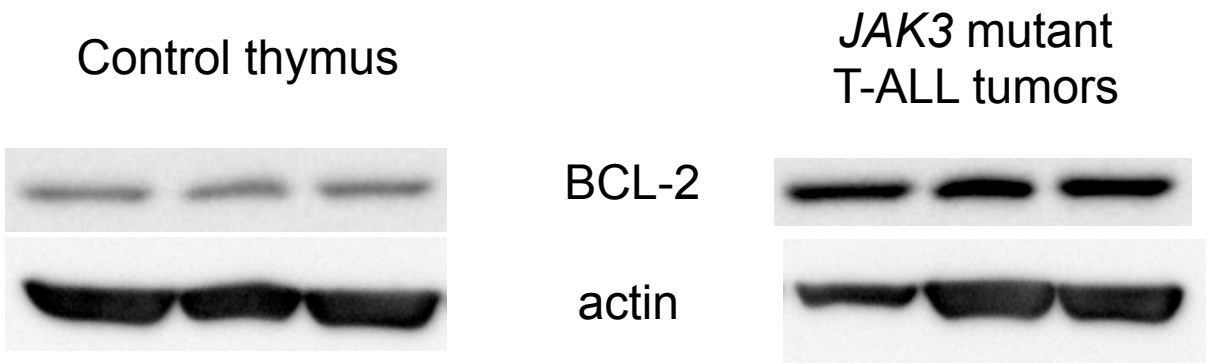
In vitro ABT-199 sensitivity human T-ALL





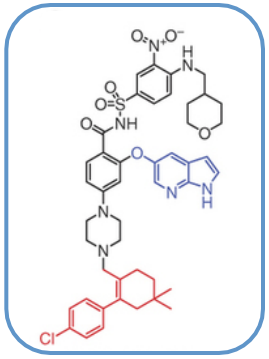
JAK3 mutant murine T-ALL



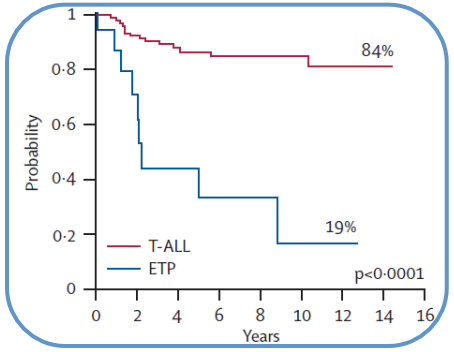


Prof. Jan Cools, KU Leuven

Conclusions



ABT-199 is a promising new drug for human cancer



Our pre-clinical data suggest ABT-199 as a promising new drug for specific subtypes of pediatric T-ALL

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Center for Medical Genetics

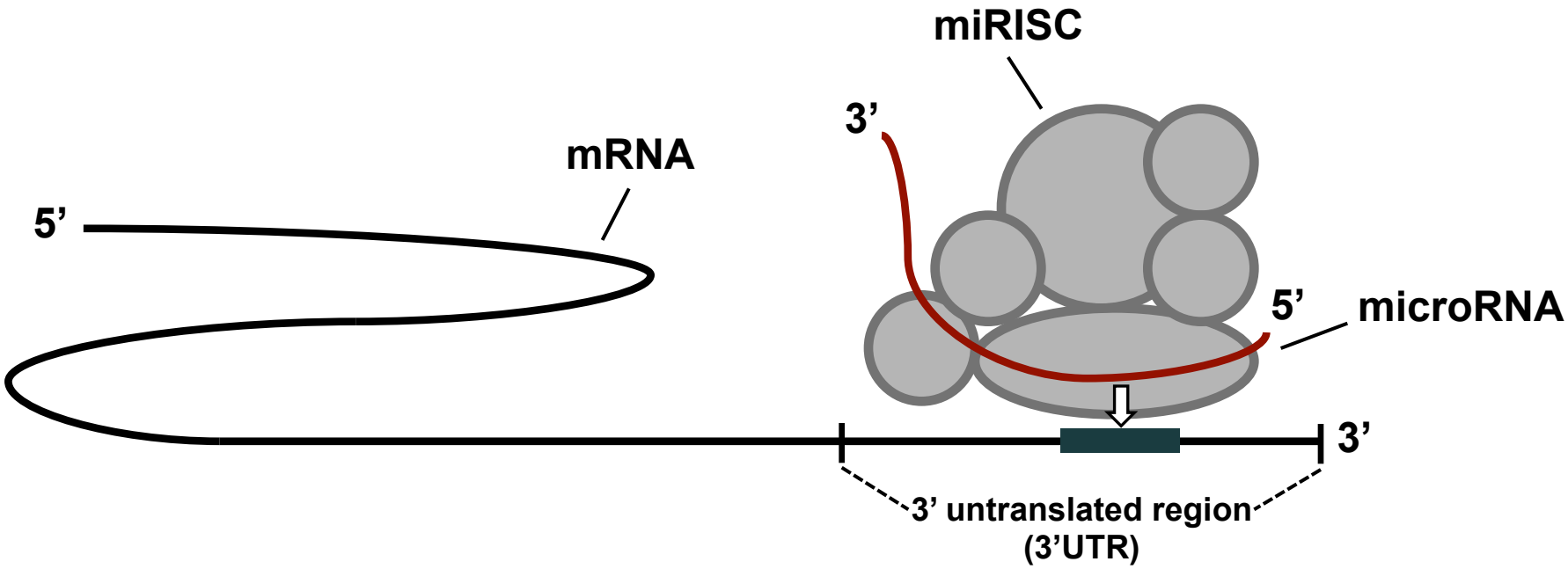
An unbiased high-throughput microRNA library screen identifies novel microRNA regulators of key oncogenes and tumor suppressor genes

Pieter Van Vlierberghe

Bioluminescent Cell-based Assay Seminar Day
Monday 31 march 2014
UCL De Duve institute
Brussels, Belgium

microRNAs

- small, non-coding RNA molecules
- post-transcriptional regulation of gene expression



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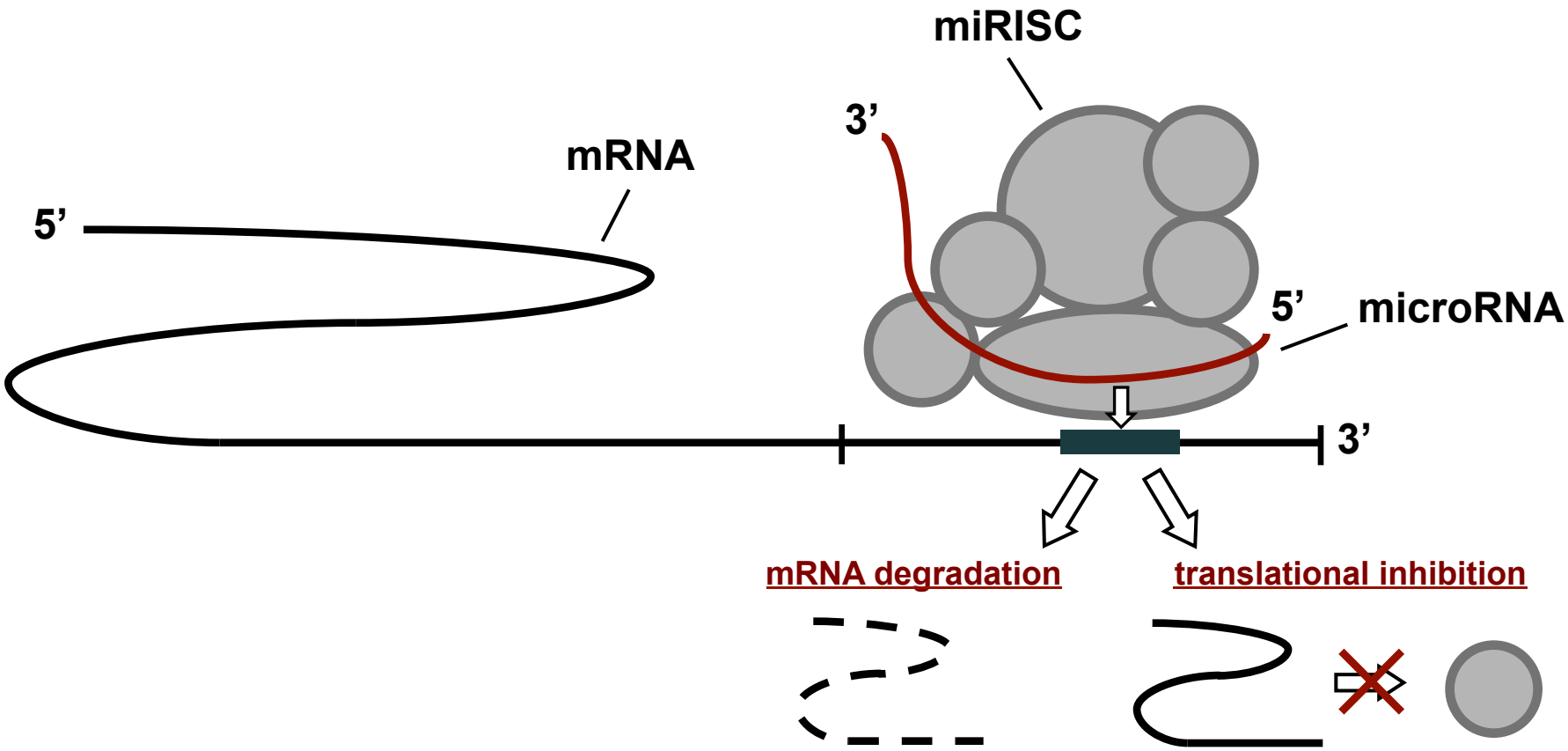

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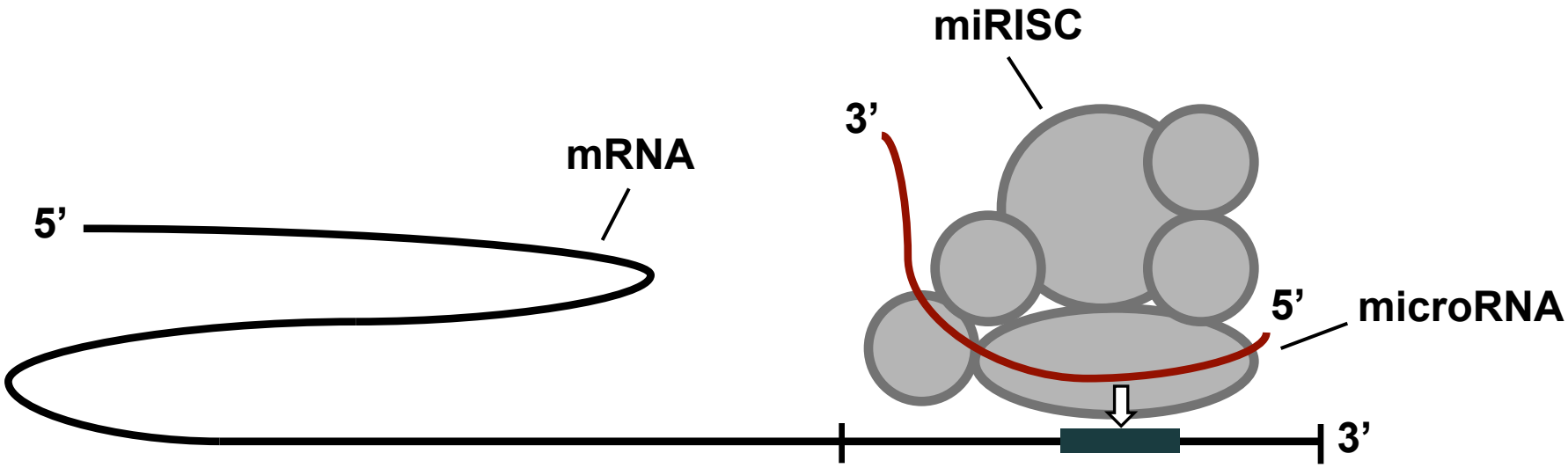
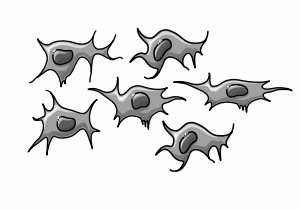


- microRNA function? microRNA targets!
- **microRNA target prediction algorithms!**

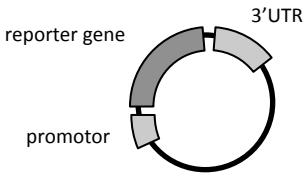
3’UTR microRNA library screen

3’UTR reporter assay

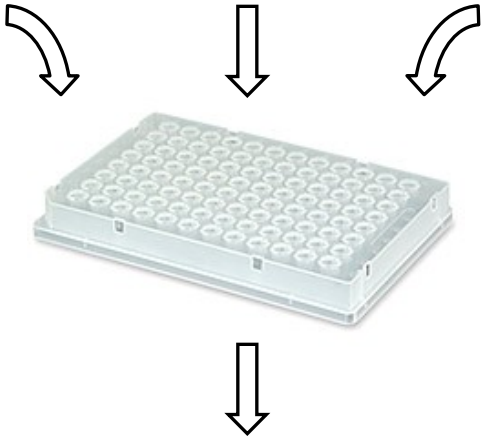
HEK-293T cells



reporter construct

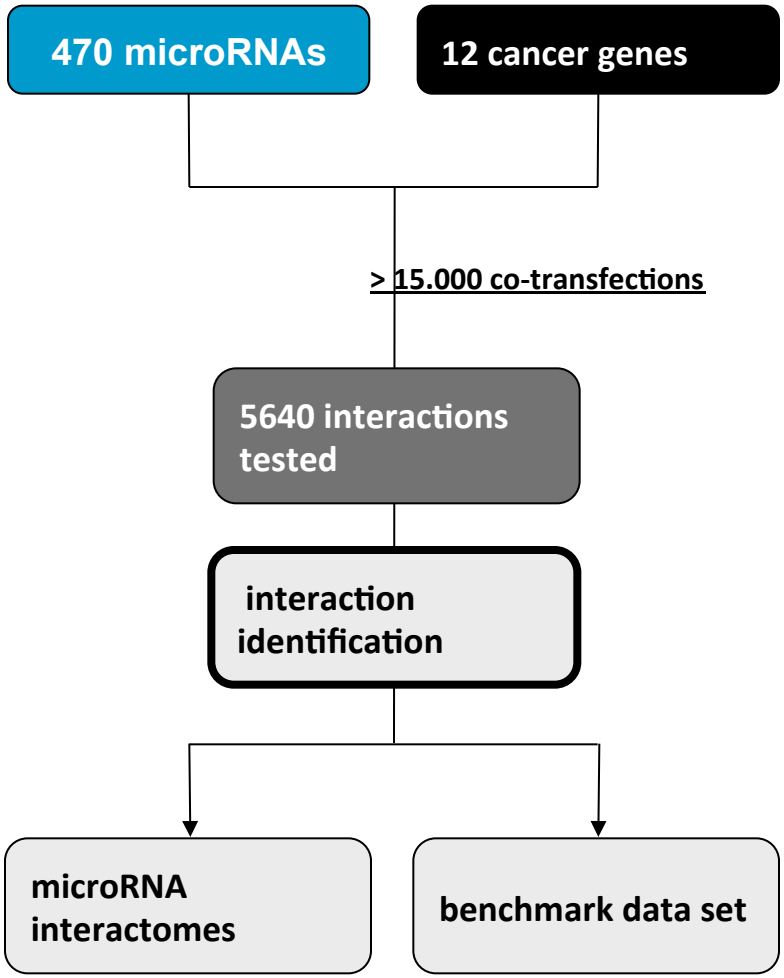


miRNA mimic



reporter gene activity

3’UTR microRNA library screen



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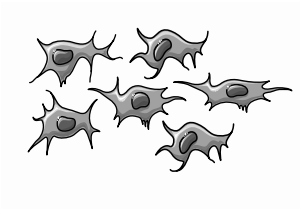
3’UTR microRNA library screen

3’UTR reporter assay

HEK-293T cells



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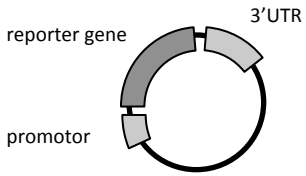
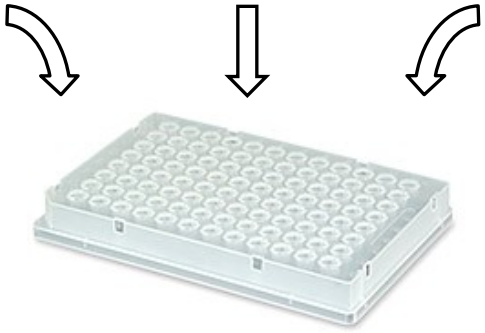


miRNA mimic

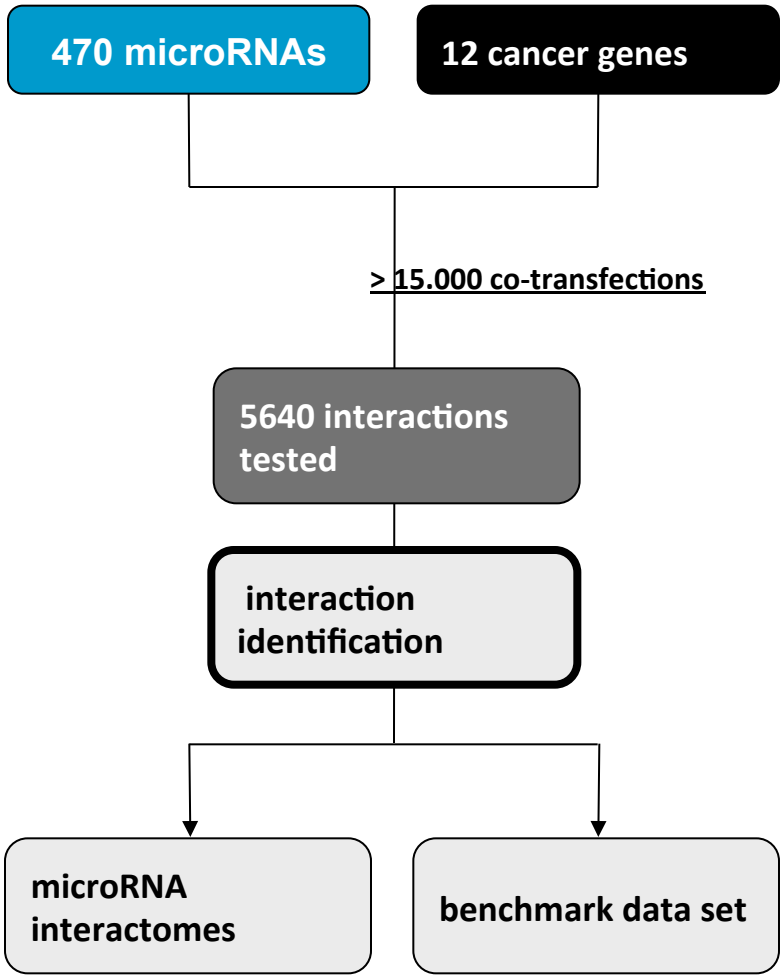


reporter gene activity

microRNA interaction score

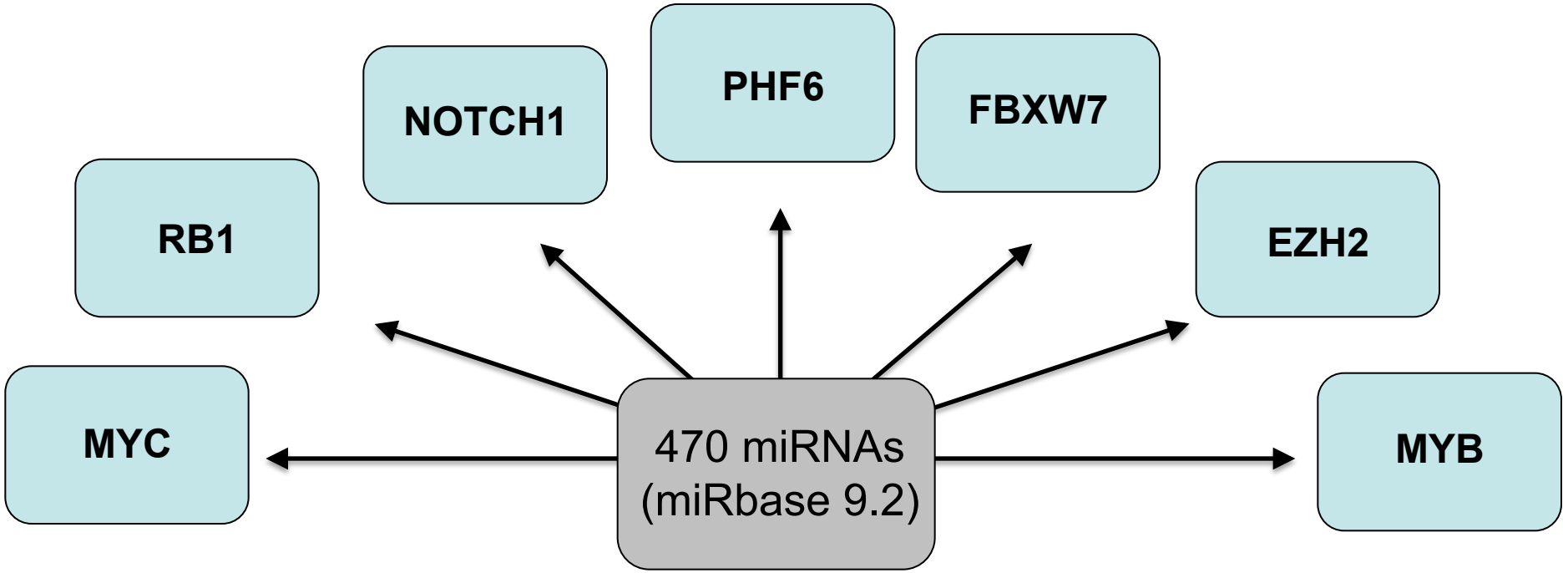
$$\Delta(\frac{r_n - median_{r_n}}{MAD}) \qquad r_n = \log\frac{r_{\exp}}{r_{norm}}$$

3’UTR microRNA library screen



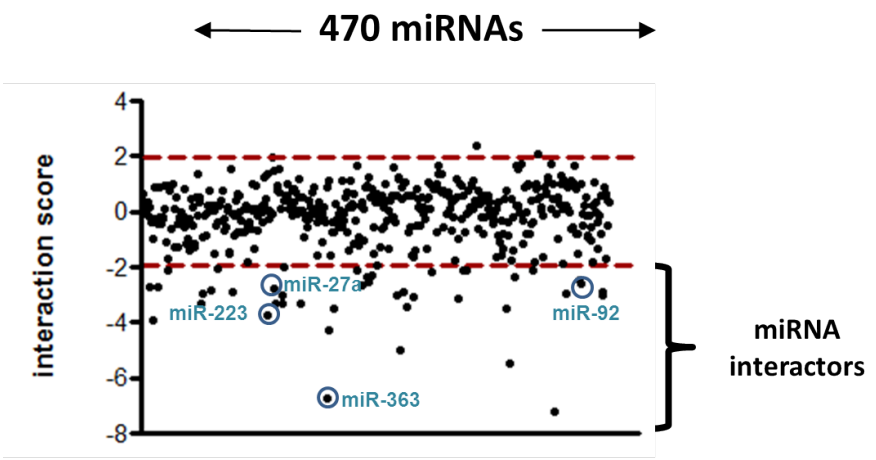
Center for Medical Genetics

3’UTR library screen



Cancer gene microRNA interactomes

miRNA-FBXW7 interactome

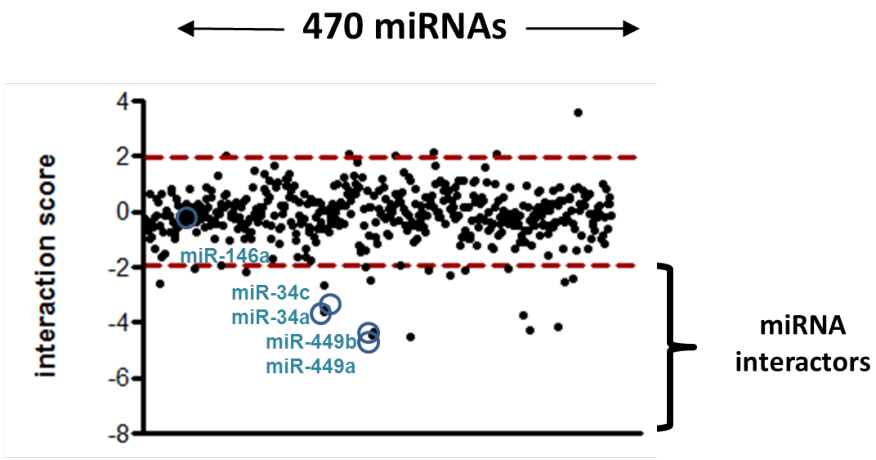


44 interactions identified

4 confirmed (already described in literature)

40 novel interactions (not yet described)

miRNA-NOTCH1 interactome



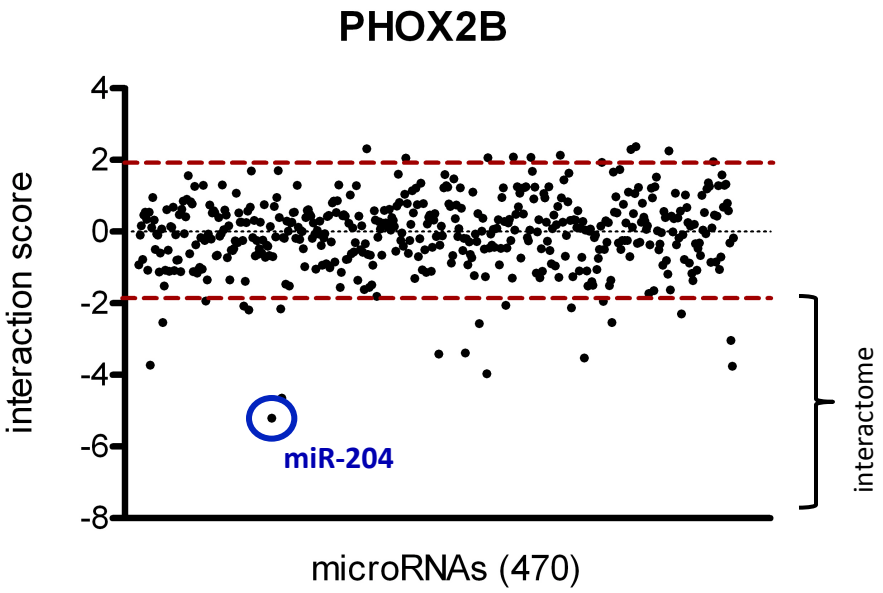
21 interactions identied

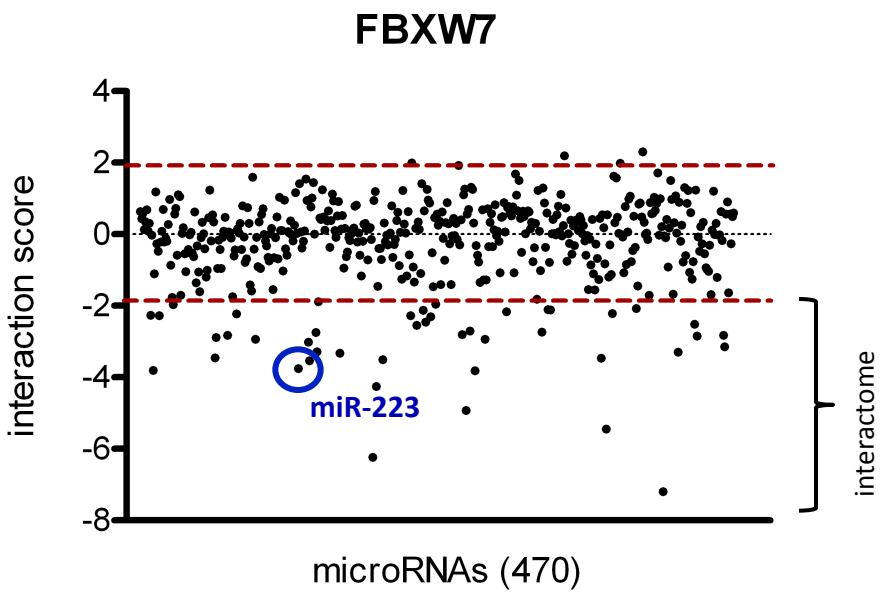
4 confirmed (already described in literature)

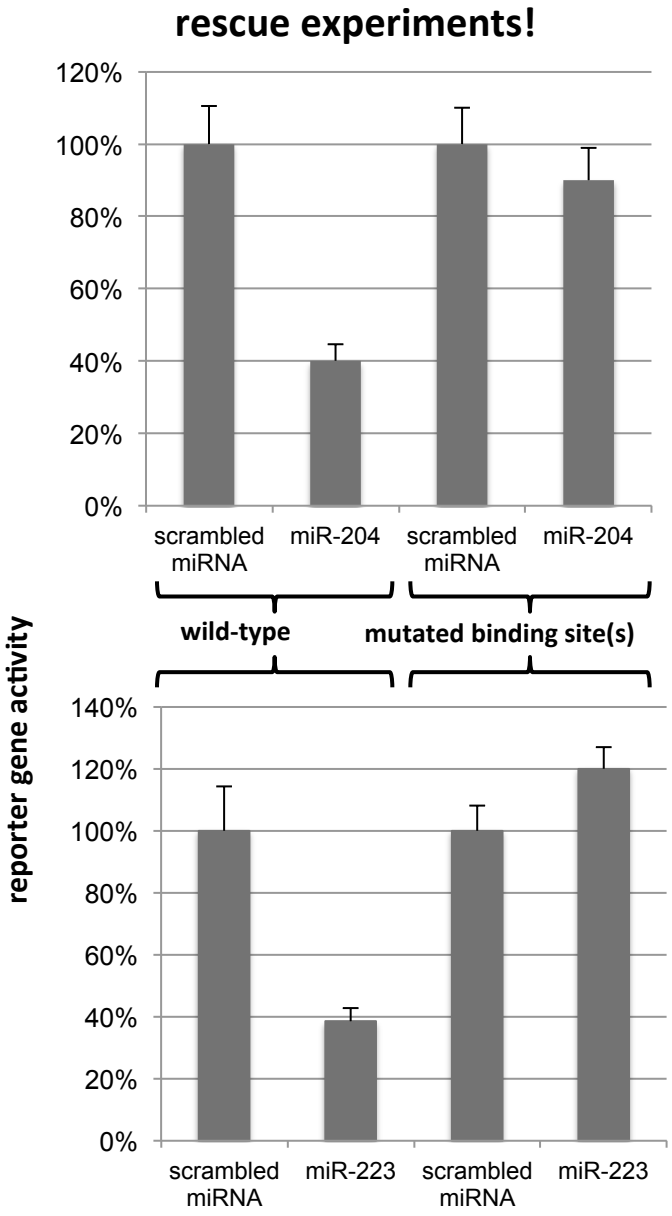
16 novel interactions (not yet described)

Center for Medical Genetics

Cancer gene microRNA interactomes







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