

NUP214-ABL1-mediated cell proliferation in T-cell acute lymphoblastic leukemia is dependent on the LCK kinase and various interacting proteins

Kim De Keersmaecker,^{1,2*} Michaël Porcu,^{1,2*} Luk Cox,^{1,2} Tiziana Girardi,^{1,2} Roel Vandepoel,^{1,2} Joyce Op de Beeck,^{1,2} Olga Gielen,^{1,2} Nicole Mentens,^{1,2} Keiryn L. Bennett,³ and Oliver Hantschel⁴

¹Center for the Biology of Disease, VIB, Leuven, Belgium; ²Center for Human Genetics, KU Leuven, Leuven, Belgium; ³CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences, Vienna, Austria; and ⁴Swiss Institute for Experimental Cancer Research (ISREC), School of Life Sciences, École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland

**KDK and MP contributed equally to this manuscript.*

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Correspondence: oliver.hantschel@epfl.ch or kim.dekeersmaecker@cme.vib-kuleuven.be

SUPPLEMENTARY INFORMATION FOR:

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¹ Center for the Biology of Disease, VIB, Leuven, Belgium; ² Center for Human Genetics, KU Leuven, Leuven, Belgium; ³ CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences, Vienna, Austria; ⁴ Swiss Institute for Experimental Cancer Research (ISREC), School of Life Sciences, École polytechnique fédérale de Lausanne, Lausanne, Switzerland

* equal contribution

Corresponding authors:

Oliver Hantschel (oliver.hantschel@epfl.ch),

Kim De Keersmaecker (kim.dekeersmaecker@cme.vib-kuleuven.be)

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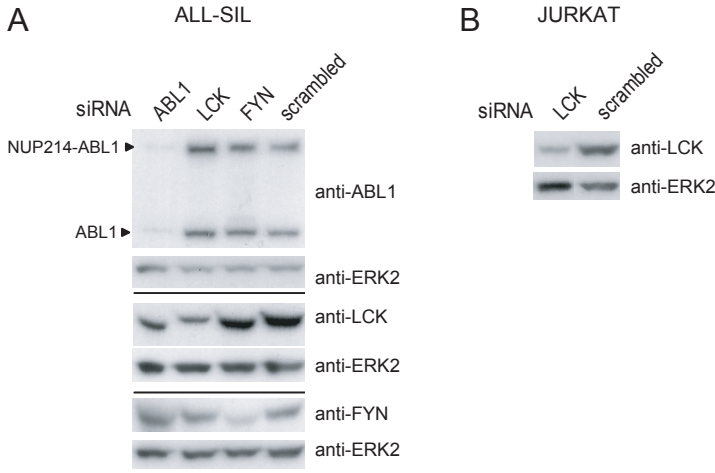
Supplementary Figure 1-4

Supplementary table 1

The following supplementary file is available as a separate .xls file:

Supplementary table 2

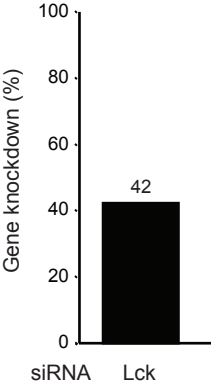
Supplementary Figure 1



Suppl. Figure 1. siRNA knock-down levels in human T-ALL cells.

Western blot analysis of ALL-SIL (A) and JURKAT (B) cells treated with indicated siRNAs. The western blots were probed with the antibodies indicated at the right side of each blot.

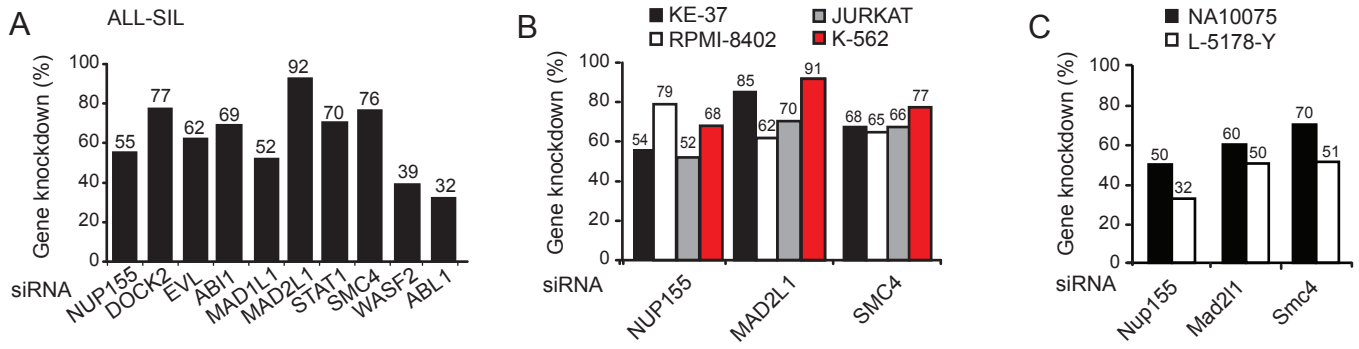
Supplementary Figure 2



Suppl. Figure 2. siRNA knock-down levels of Lck in mouse T-ALL cells.

Lck knock-down levels were measured by qRT-PCR in mouse L-5178-Y cells treated with control scrambled siRNA, or Lck siRNA. Indicated levels are relative to scrambled control siRNA treated cells (0% knock-down).

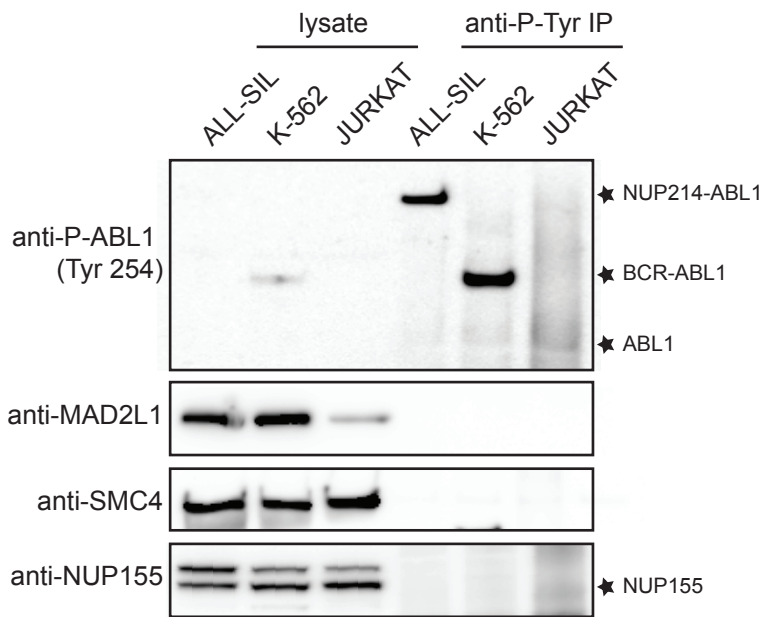
Supplementary Figure 3



Suppl. Figure 3. siRNA knock-down levels in human and mouse T-ALL cells.

Knock-down levels of indicated genes were measured by qRT-PCR in cells treated with the corresponding siRNAs. (A) ALL-SIL cells. (B) KE-37, JURKAT, RPMI-8402 and K-562 cells. (C) NA10075 and L-5178-Y cells. Indicated levels are relative to scrambled control siRNA treated cells (0% knock-down).

Supplementary Figure 4



Suppl. Figure 4. No detectable tyrosine phosphorylation of MAD2L1, SMC4 and NUP155 .

Immunoprecipitation (IP) using a pan-phospho-tyrosine (anti-P-Tyr) antibody on ALL-SIL, K562 and JURKAT cell lysates pulled down phosphorylated NUP214-ABL1 (in ALL-SIL) and BCR-ABL1 (in K-562) as detected on the western blot with the anti-phospho-ABL1 (Tyr 245) antibody. NUP214-ABL1 interaction partners MAD2L1, SMC4 and NUP155 were not detectable in the IP samples indicating these proteins may not be phosphorylated.

Supplementary table 1. siRNA sequences used in this study

Target Gene	Species	Type	Sequence
<i>ABL1</i>	Human	Invitrogen Stealth	GGAAUGGUGUGAAGCCCAAACCAAA
<i>LCK</i>	Human	Invitrogen Stealth	GCAUUCAUUGAAGAGCGGAAUUUAUA
<i>FYN</i>	Human	Invitrogen Stealth	CCCUGUACGGGAGGUUCACAAUCA
<i>NUP155</i>	Human	Invitrogen Stealth	CCGAUGGUGAAUUUCUUCAUGAAUU
<i>DOCK2</i>	Human	Invitrogen Stealth	CGACAUGAUGCUGUGUGAAUAUCA
<i>EVL</i>	Human	Invitrogen Stealth	GCAGCAGCGUCAGGAAUCUCUAGAA
<i>ABI1</i>	Human	Invitrogen Stealth	ACUGGGACGGAAUACUCCUUAUAAA
<i>MAD1L1</i>	Human	Invitrogen Stealth	GAAGACCUUUCAGAUUCGUGGUUG
<i>MAD2L1</i>	Human	Invitrogen Stealth	GCCACUGUUGGAAGUUUCUUGUUCA
<i>STAT1</i>	Human	Invitrogen Stealth	GCAAGCGUAAUCUUCAGGAUAAUUU
<i>SMC4</i>	Human	Invitrogen Stealth	CAGGGUGAAGUUGAACAAAUUGCUA
<i>WASF2</i>	Human	Invitrogen Stealth	CCCAUCUUUCCCACCUCACCCUGAU
<i>Lck</i>	Mouse	IDT Screening DsiRNA	GUAUUUCUGUUCUUCGAUGAACGCCAU
<i>Mad2L1</i>	Mouse	Ambion Silencer Select	UUGUAAAUGAGCGUAGACGga
<i>Nup155</i>	Mouse	Ambion Silencer Select	AUUCGAAGAAAAGUAAAUGCaa
<i>Smc4</i>	Mouse	Ambion Silencer Select	UGCUGUAUUAUGUCGACUGag
Scrambled	Human	Invitrogen Stealth	Available on request: #12935100
Scrambled	Mouse	IDT	CGUUAUUCGCGUAUAAUACGCGUat
Scrambled	Mouse	Ambion Silencer Select	Available on request #4390847