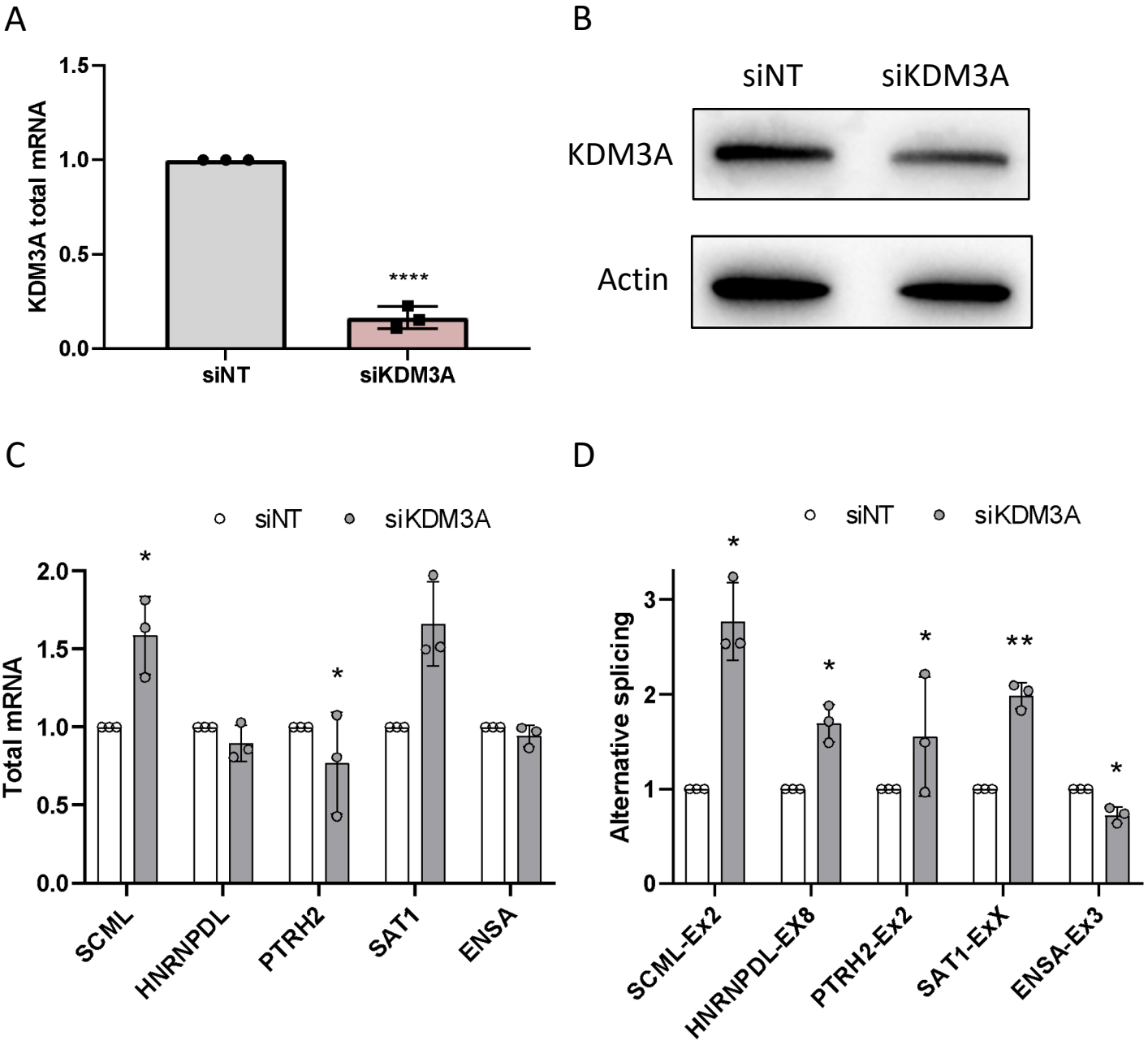
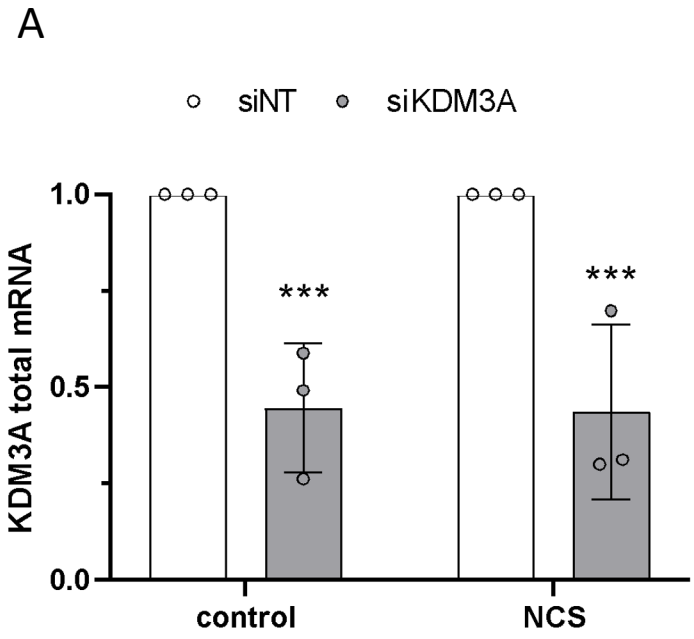
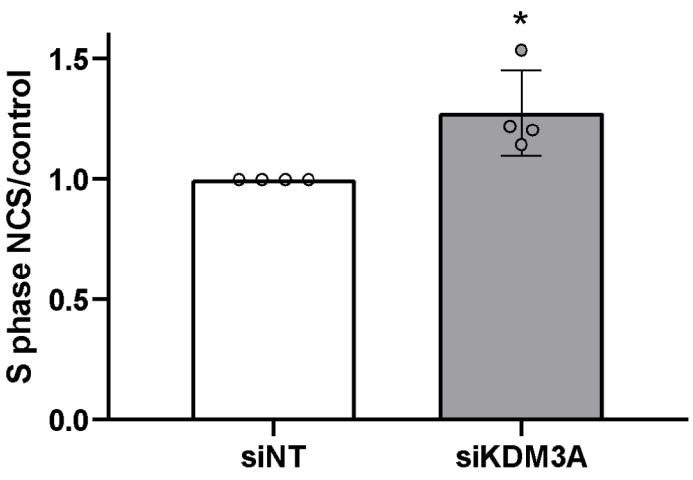
Supplementary Fig. S1

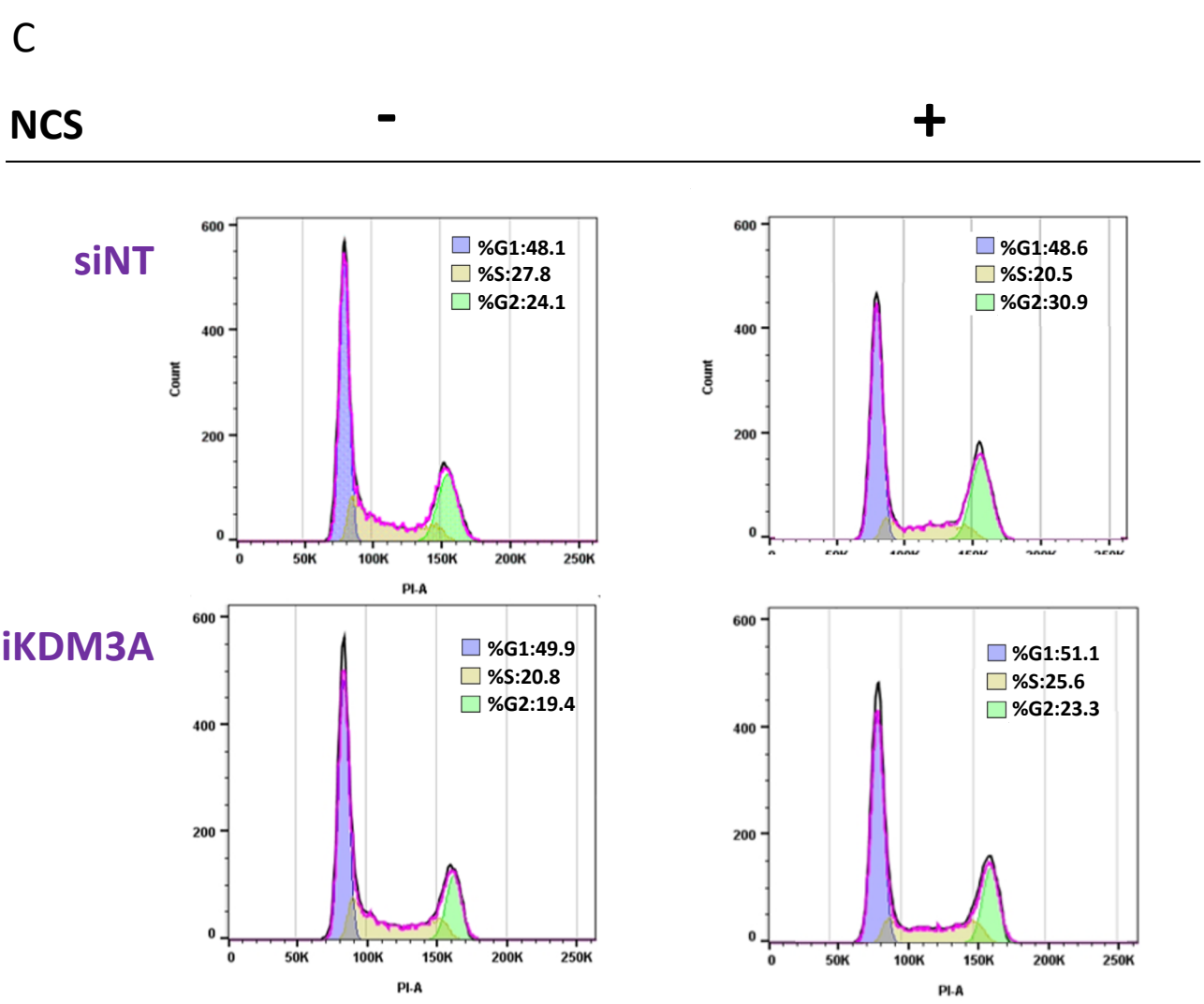



**Supplementary Figure S1.** **A-D**: MCF7 cells were transfected with non-targeting siRNA (siNT) and siKDM3A for 72 h. Total RNA was extracted and analyzed by real-time PCR for total mRNA amount of KDM3A (**A**). Immunoblotting was conducting using the indicated antibodies (**B**). Five genes (HNRNPDL, SAT1, SCML, PTRH2 and ENSA) were chosen for validation and total RNA was extracted and analyzed by real-time PCR for their total mRNA amount relative to *CycloA* reference gene (**C**); PSI of indicated genes was analyzed by real-time PCR relative to the total amount of that gene (**D**). Values represent averages of three independent experiments ±SD (* $p<0.05$; ** $p<0.01$; **** $p<0.0001$; Student's t test comparing to siNT).

Supplementary Fig. S2



Supplementary Fig. S2, continued

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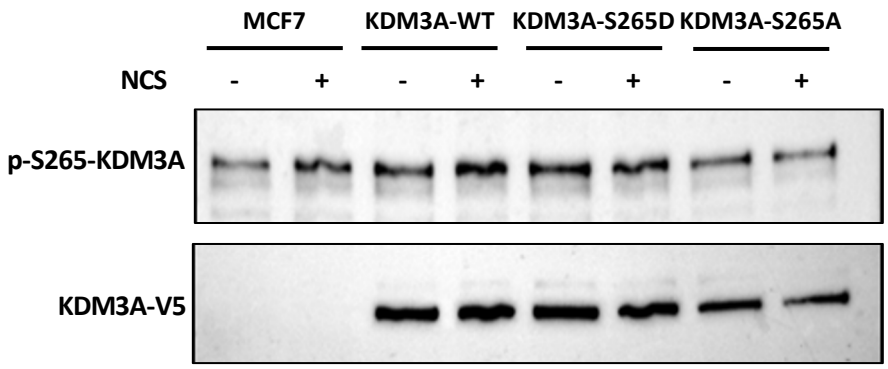


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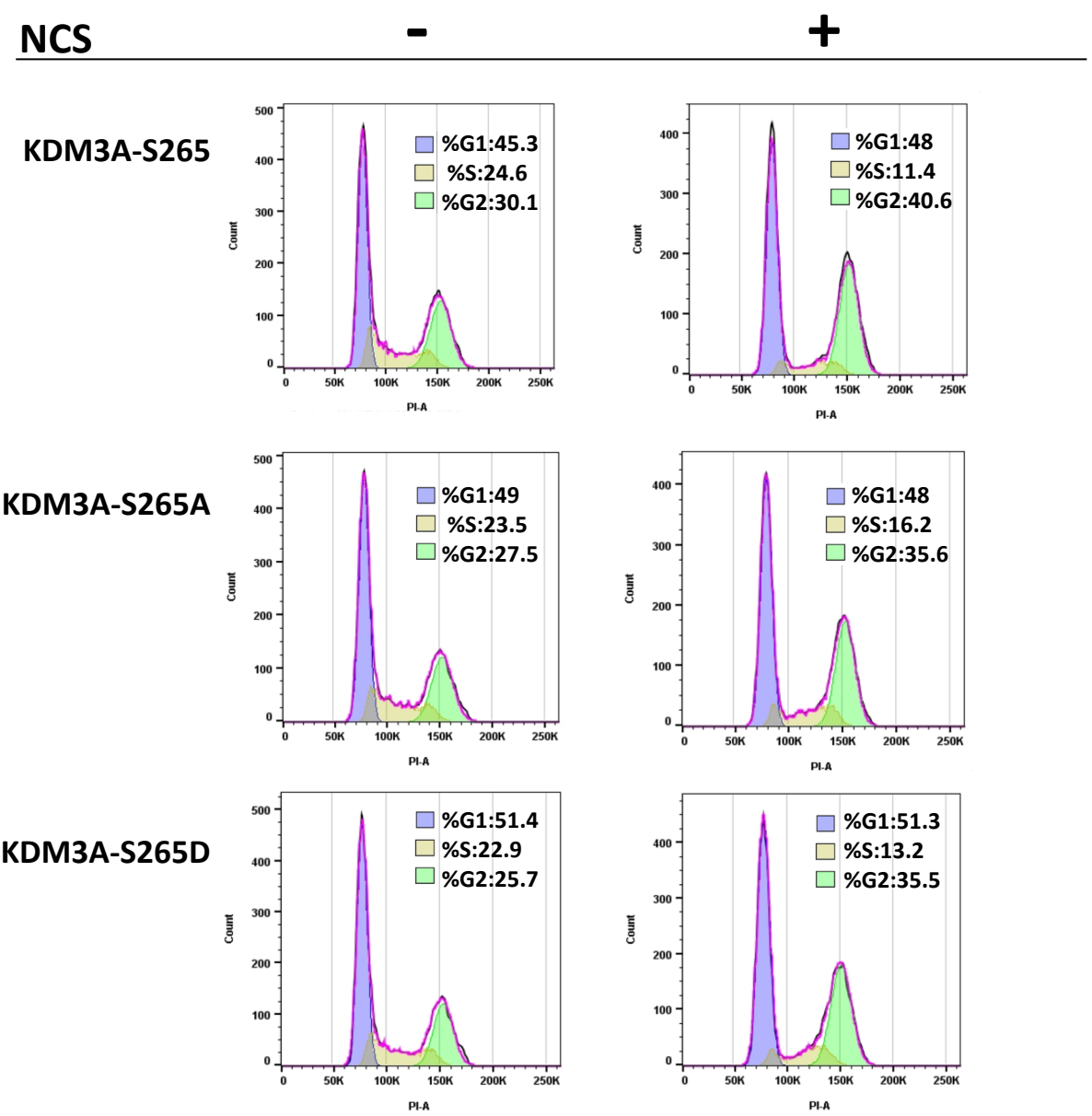
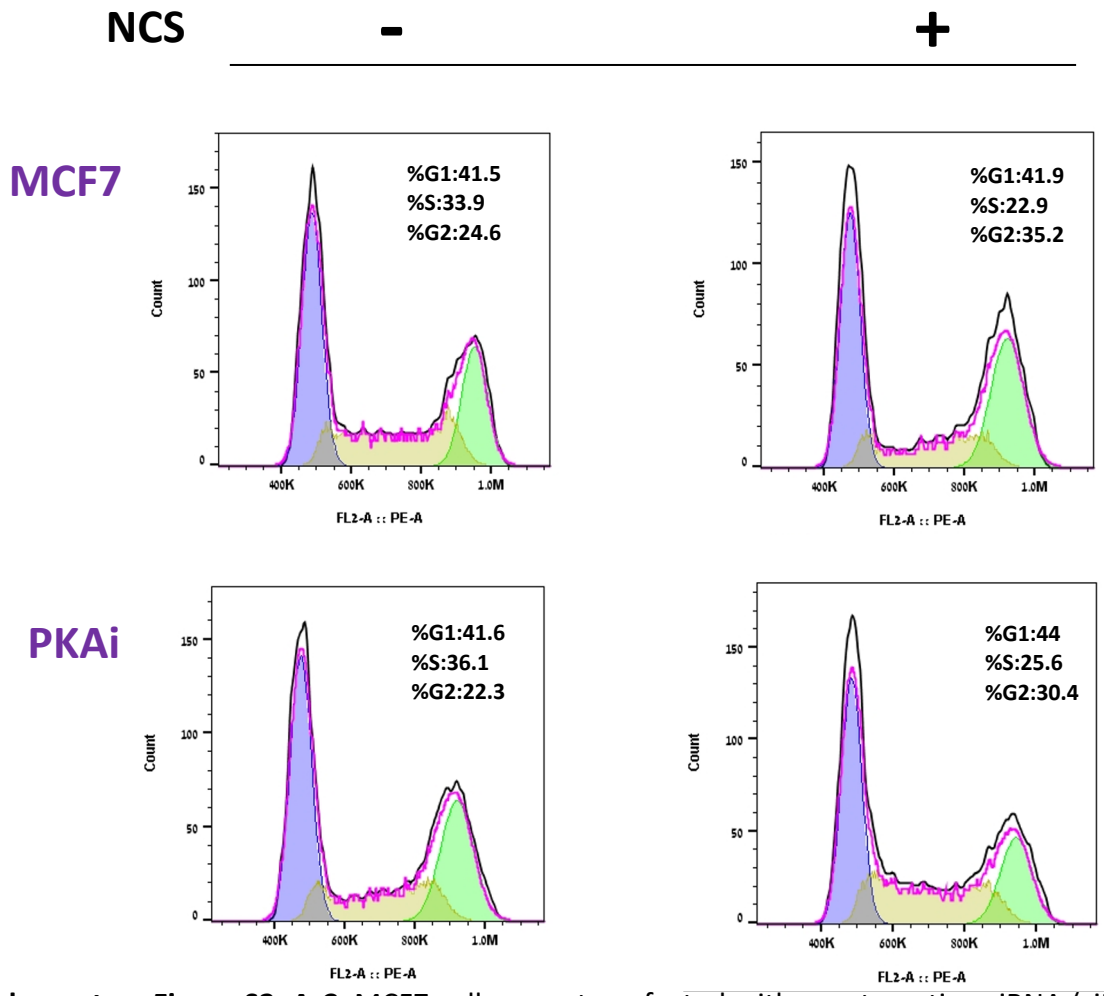


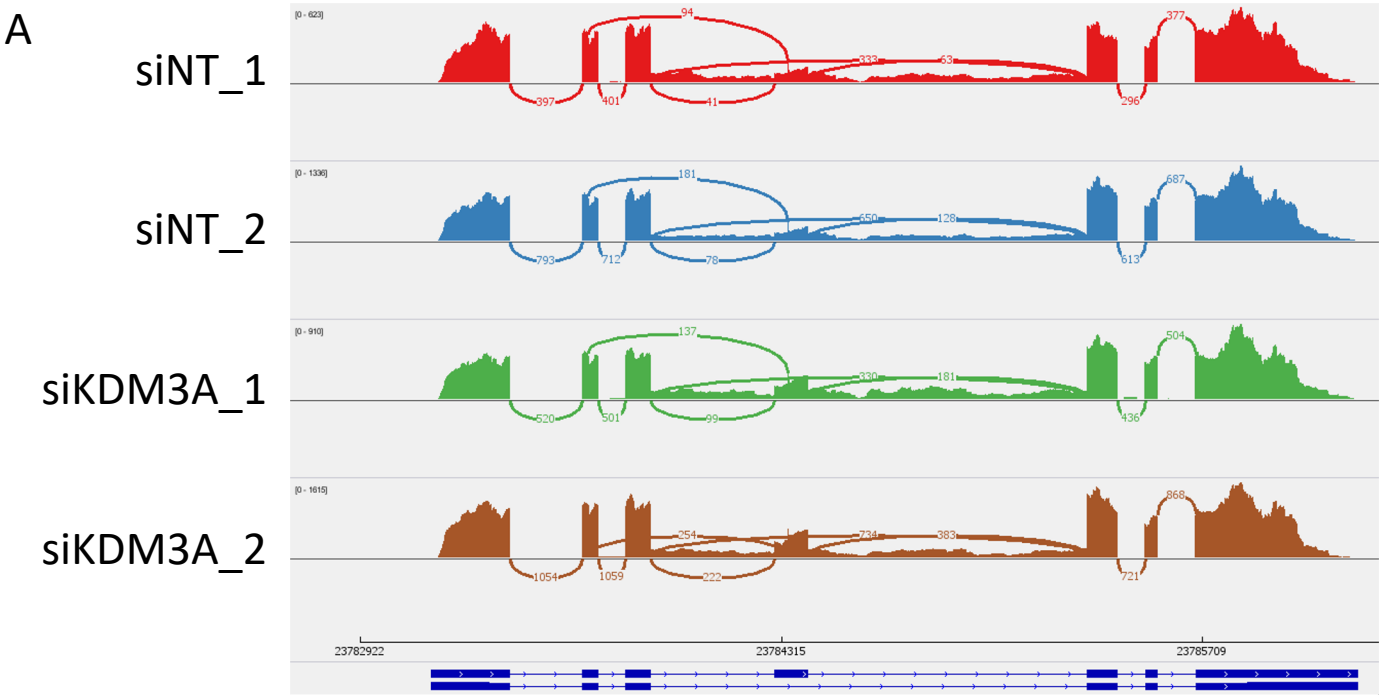
Supplementary Fig. S2, continued

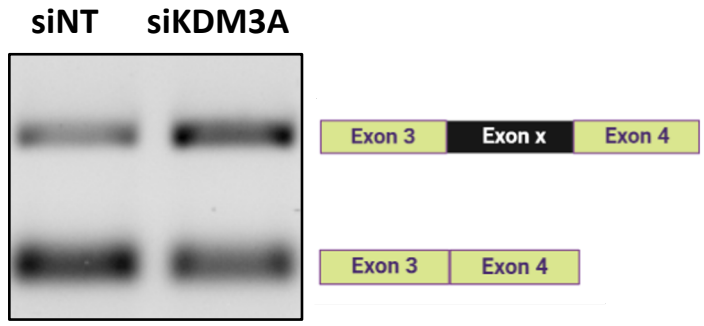
F

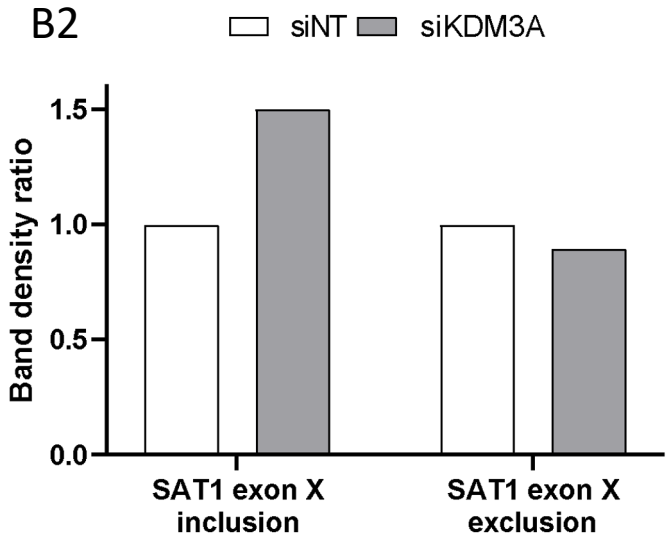


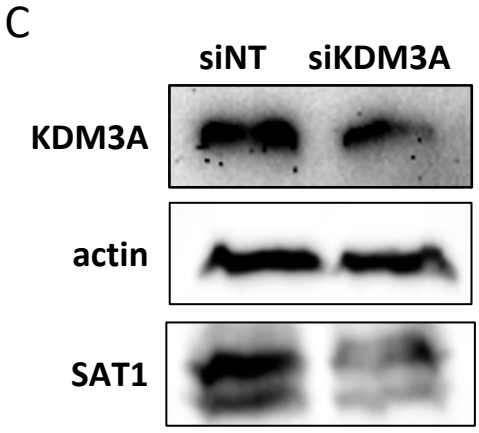

**Supplementary Figure S2**. **A-C**. MCF7 cells were transfected with non-targeting siRNA (siNT) and siKDM3A for 72 h and treated with 200 ng/ml NCS for 8 h. Total RNA was extracted and analyzed by real-time PCR for total mRNA amount of KDM3A relative to *CycloA* reference gene (**A**). Cells were analyzed using flow cytometry. The amount of cells accumulated in S-phase is shown as a fold-change relative to untreated cells (**B**). Values in **A** & **B** represent averages of three independent experiments ±SD (* $p<0.05$; *** $p<0.005$; Student's t test comparing to siNT), and cells were analyzed using flow cytometry, a population histogram for one representative experiment is shown (**C**). **D&E**. Protein was collected from MCF7 cells stably expressing either wild-type KDM3A or KDM3A mutated at serine 265 to alanine (S265A) or to aspartic acid (S265D). Immunoblotting was conducted with the indicated antibodies (**D**) and cells were treated with 200 ng/ml NCS for 8 h and analyzed using flow cytometry. A population histogram for one representative experiment is shown (**E**). **F.** MCF7 cells were treated with PKAi for 30 min followed by treatment with 200 ng/ml NCS for 8 h and analyzed using flow cytometry. A population histogram for one representative experiment is shown.

Supplementary Fig. S3

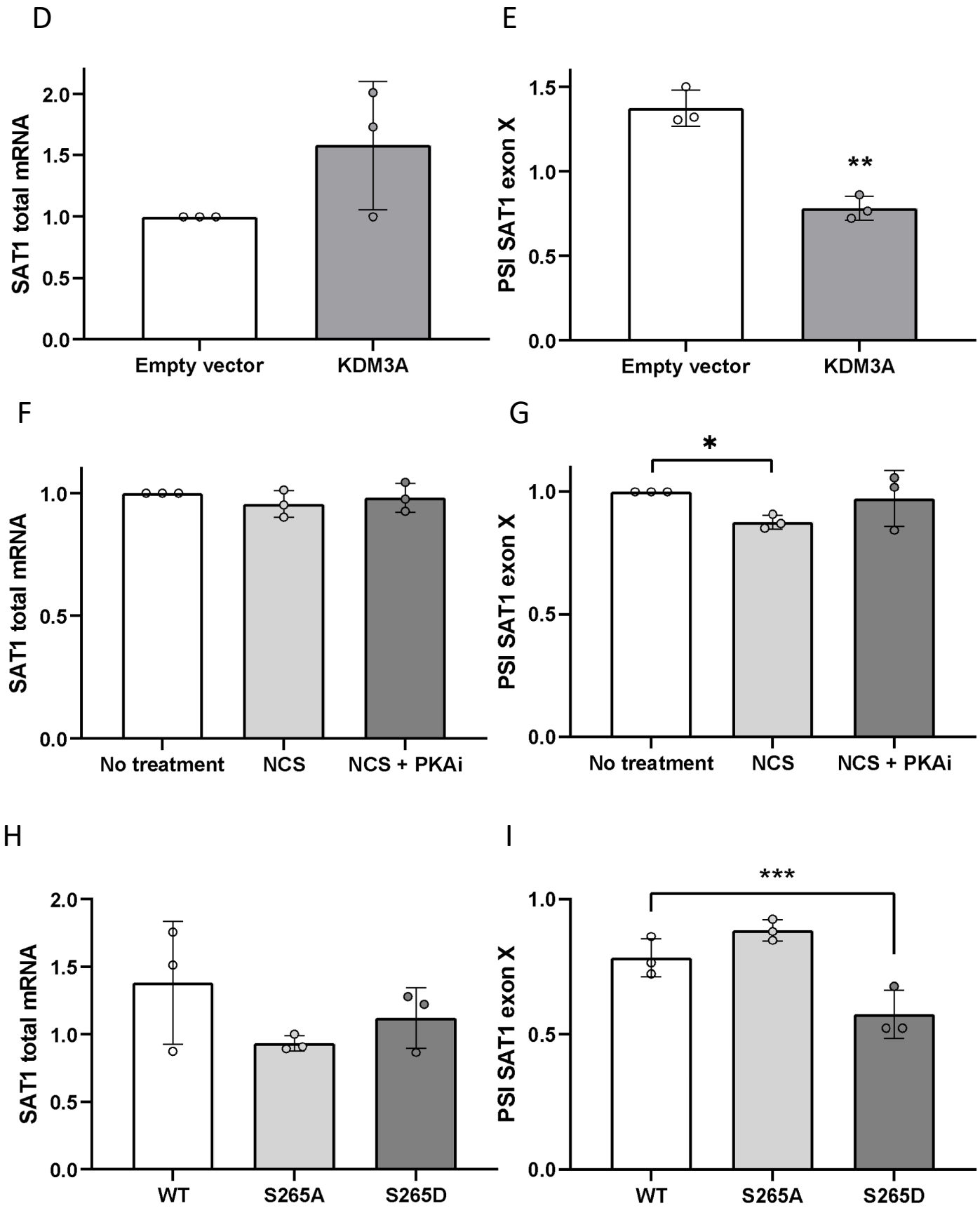


Supplementary Fig. S3



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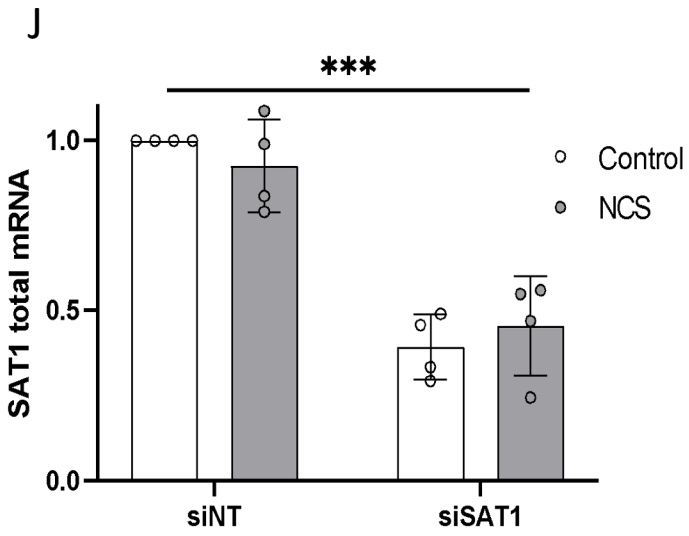
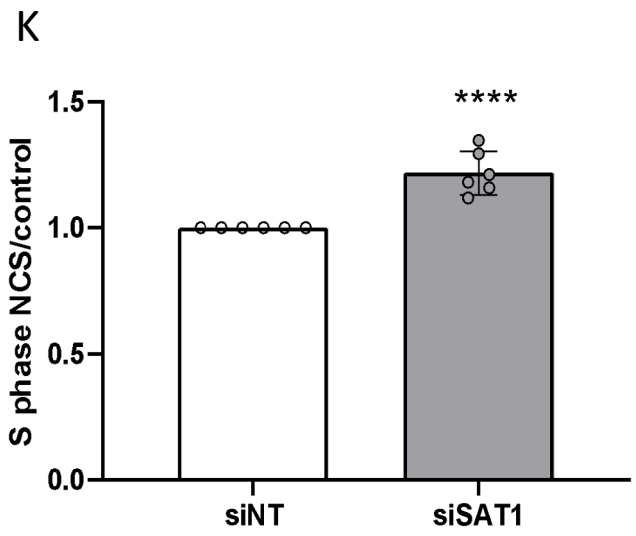
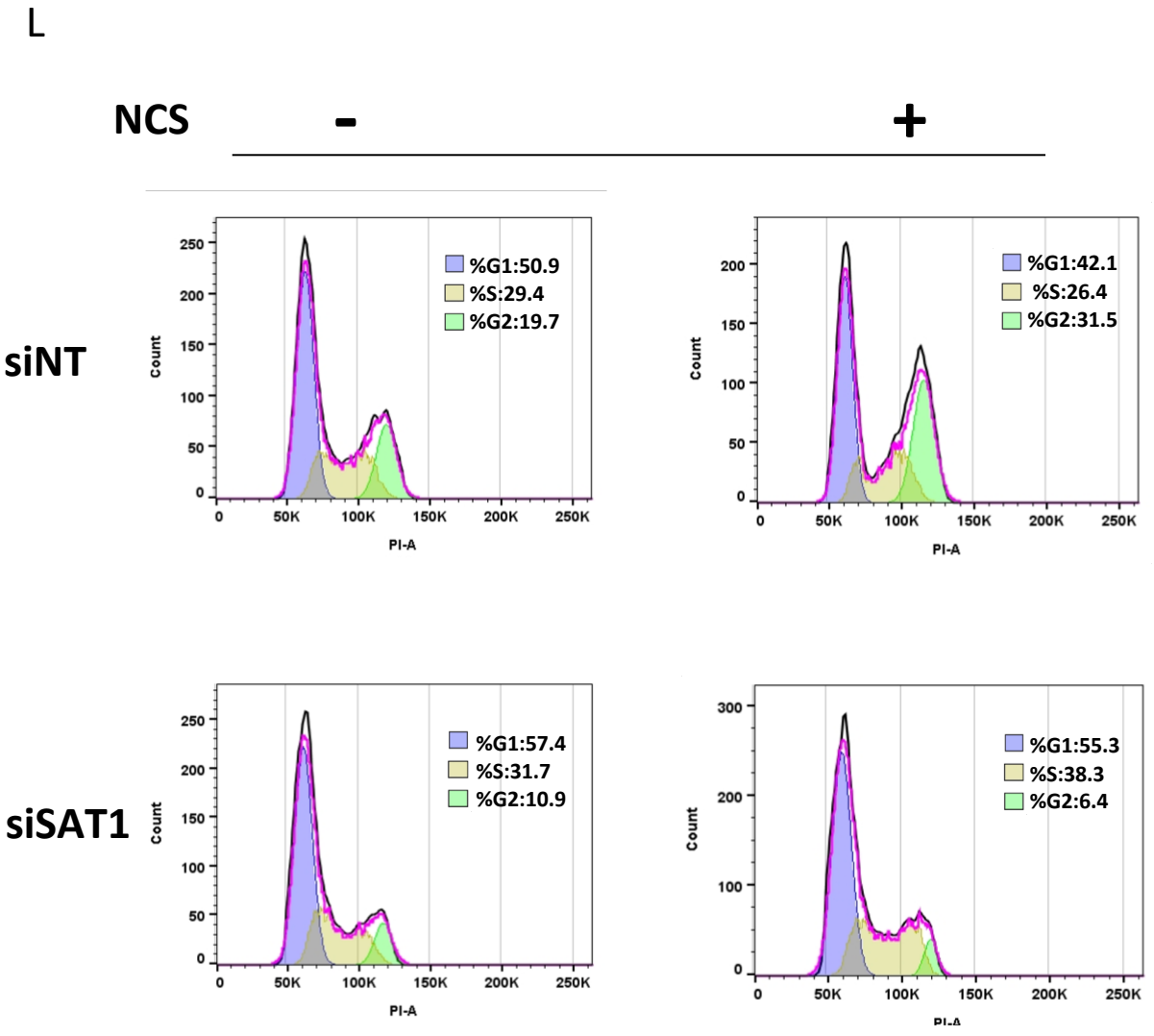
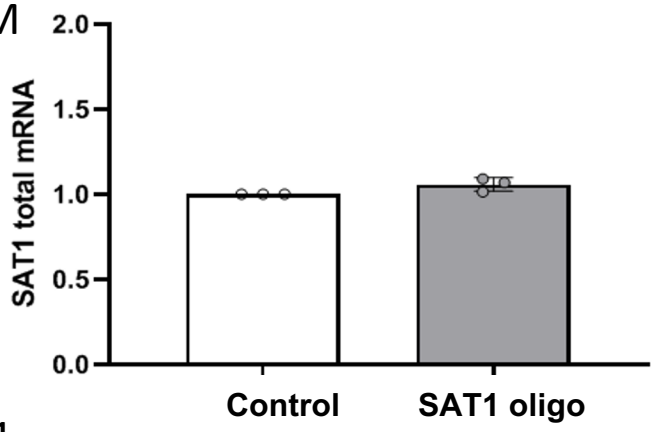


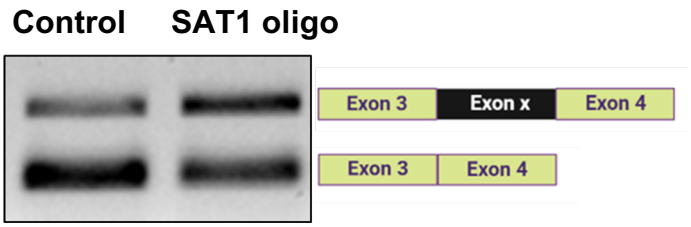



Supplementary Fig. S3

M




N1

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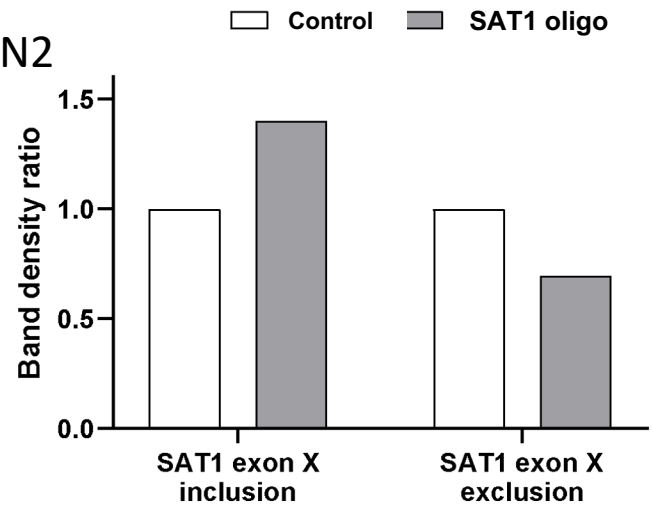


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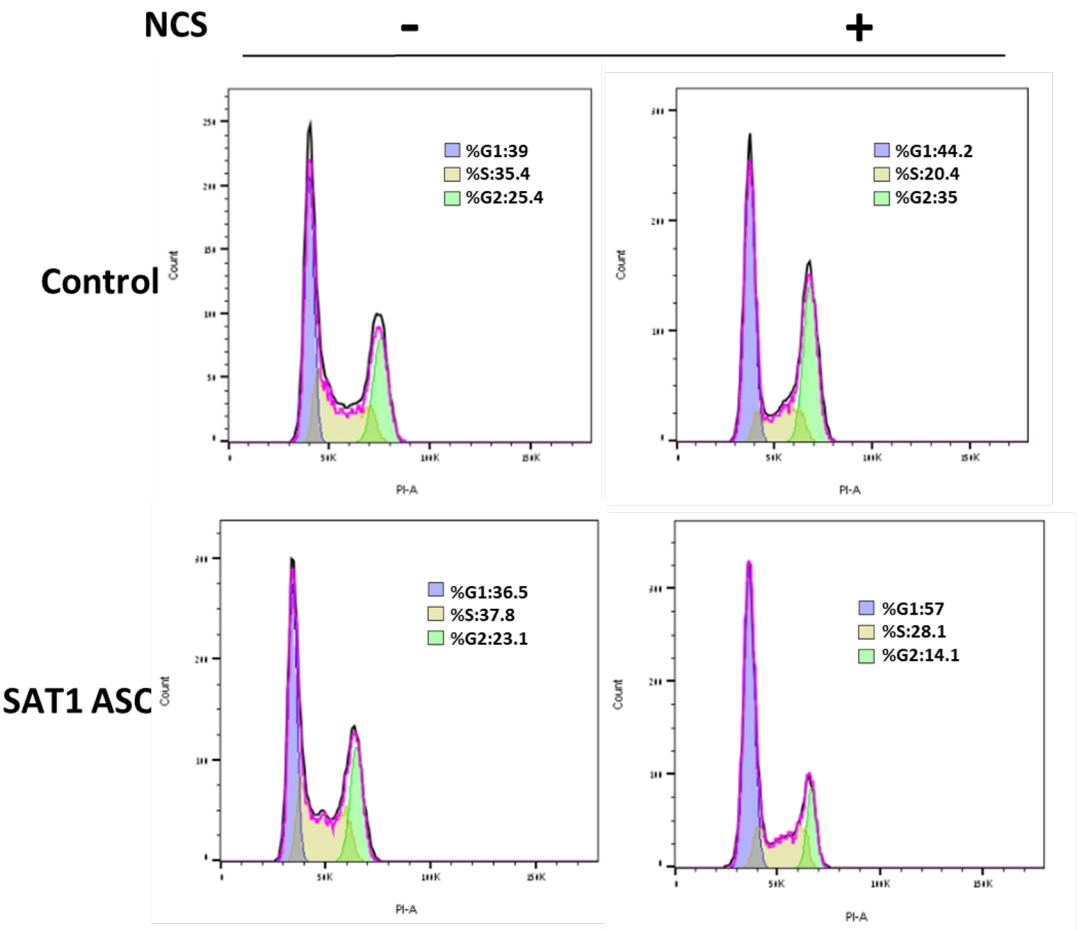

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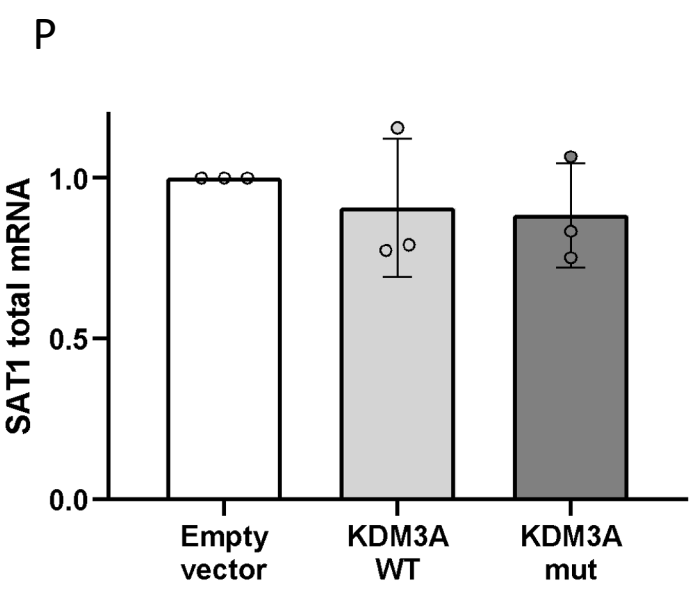
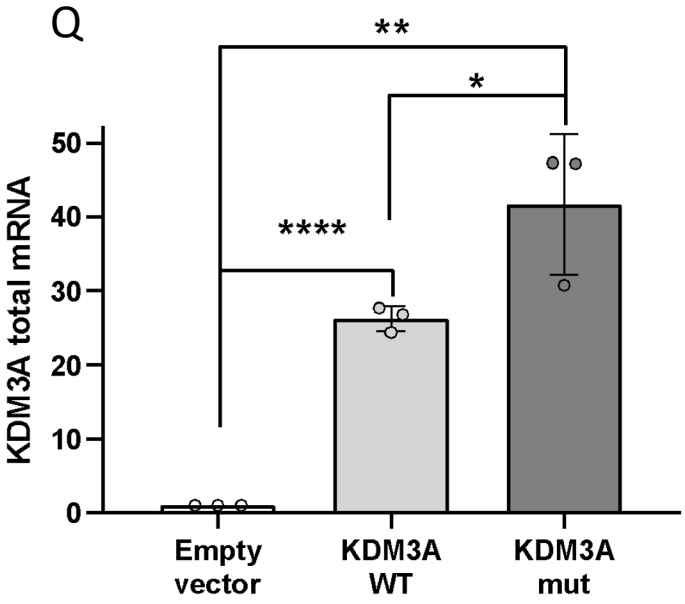
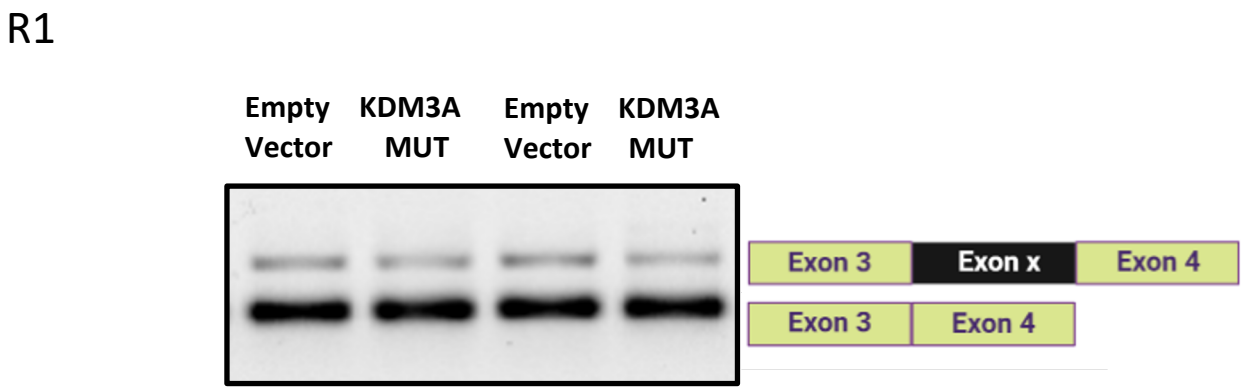
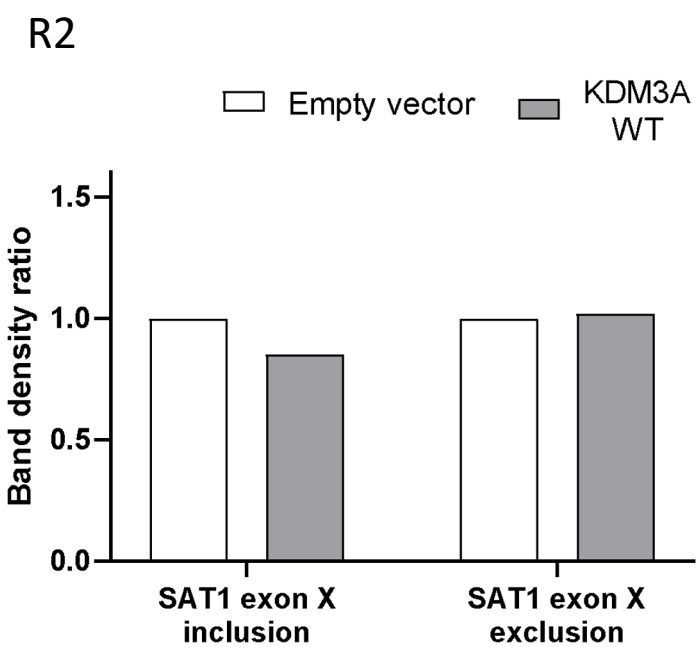
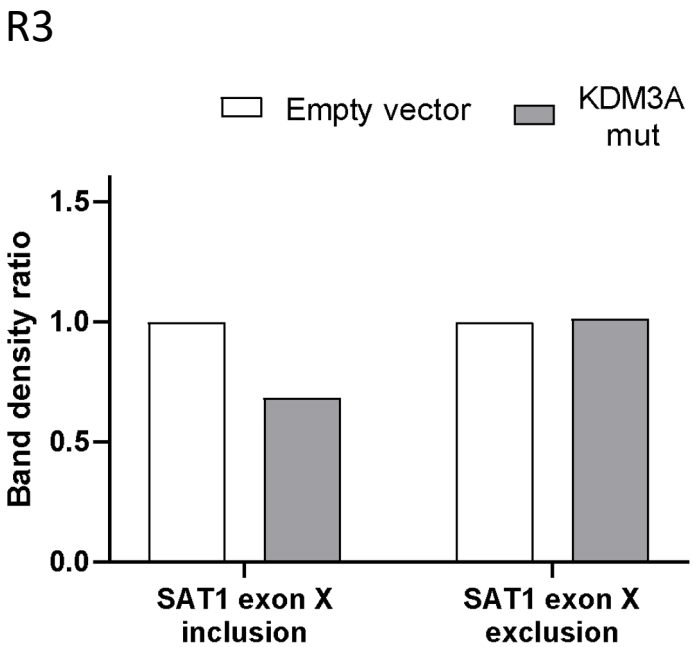


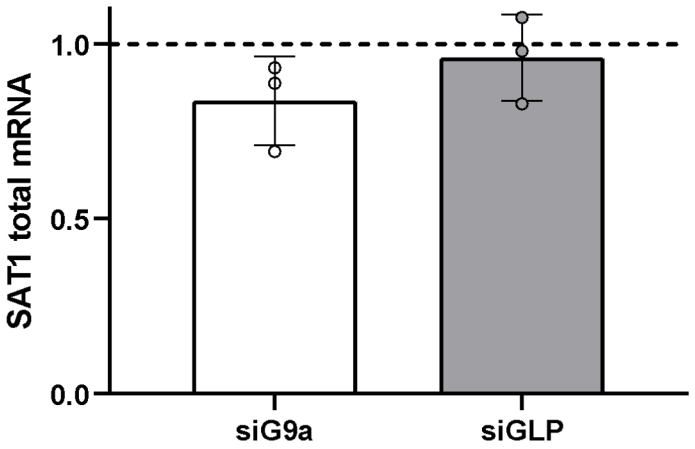



Supplementary Fig. S3, continued

S

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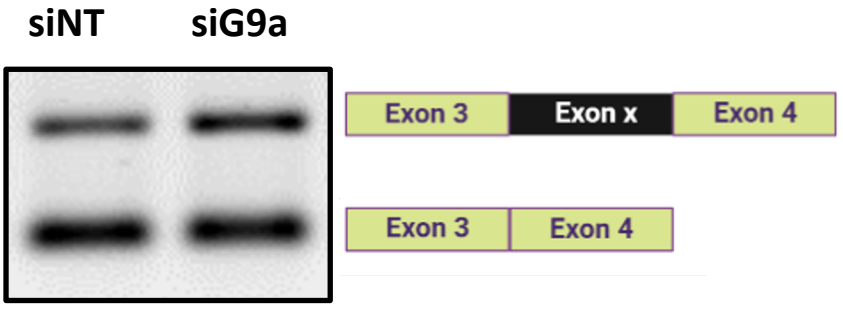


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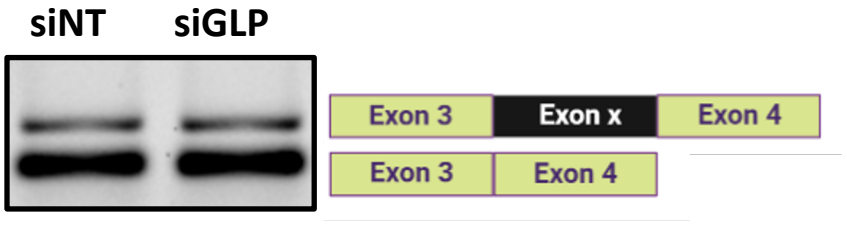
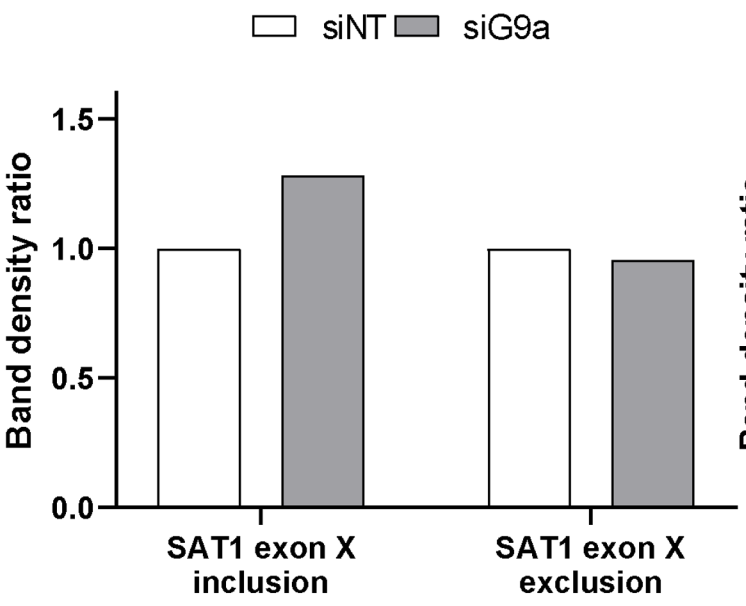


Supplementary Fig. S3, continued

W1

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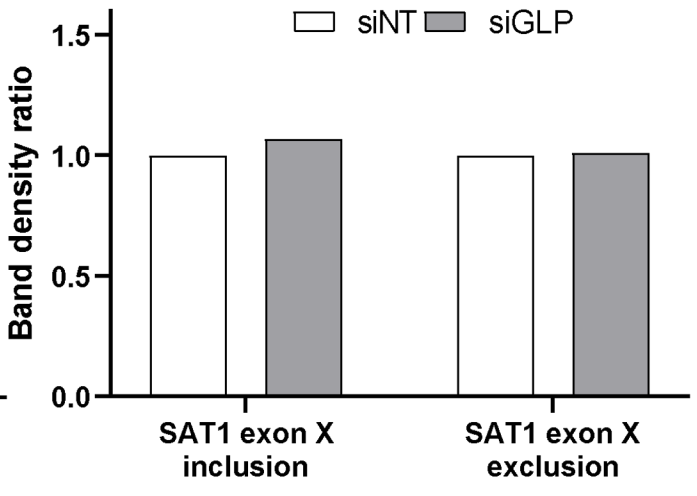


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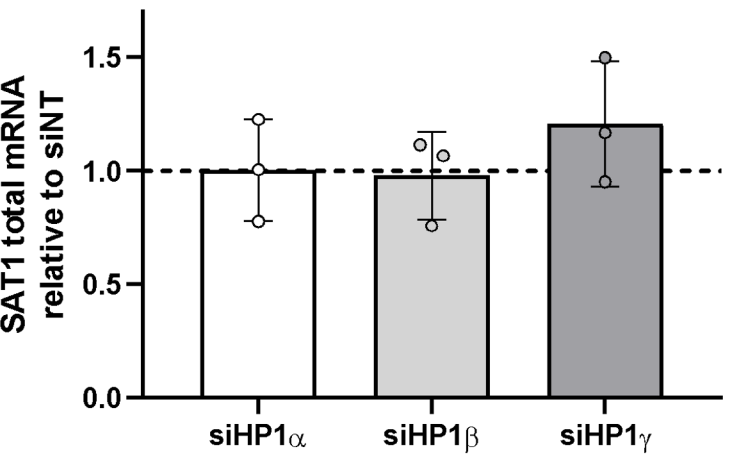


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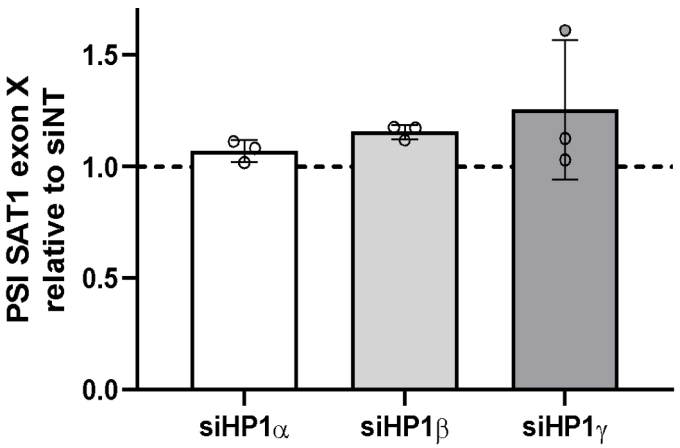
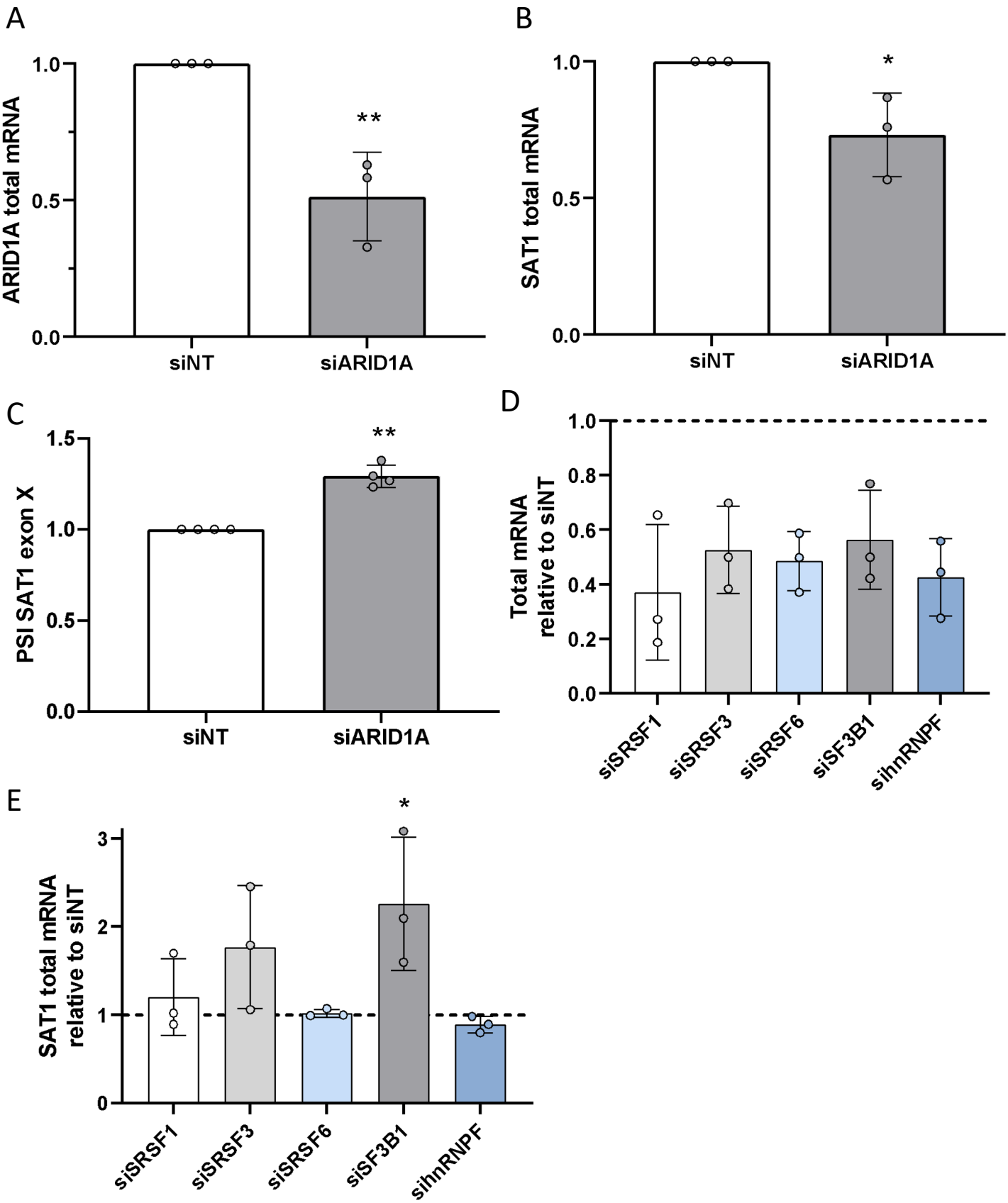


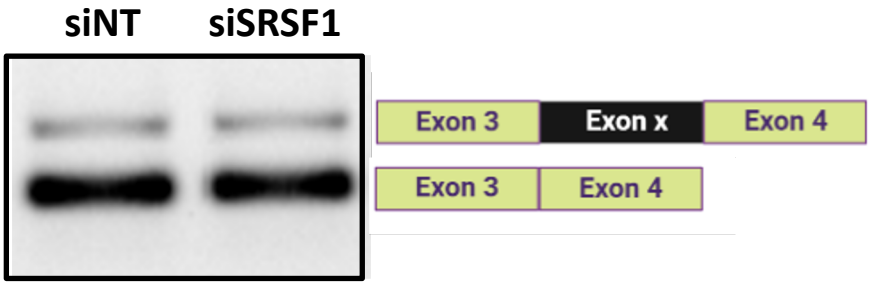
**Supplementary Figure S3**: **A**. RPKM for each alignment track are shown as sashimi plots (Katz *et al.* 2015). Arcs denote splice junctions, quantified in spanning reads, as specified near each exon junction of *SAT1* gene. **B1-C**. MCF7 cells were transfected with non-targeting siRNA (siNT) and siKDM3A for 72 h. Total RNA was extracted and analyzed by semi-quantitative PCR. Semi-quantitative PCR results for SAT1 isoforms (ExX in, ExX out), in siNT and siKDM3A (**B1**). Bar graph showing the volume of each band of SAT1 (ExX in, ExX out) for siKDM3A relative to siNT (**B2**). Immunoblotting was conducted using the indicated antibodies (**C**). **D & E**. SAT1 alternative splicing in MCF7 cells stably expressing either the empty vector or wild-type KDM3A. Total RNA was extracted and analyzed by real-time PCR for KDM3A relative to *CycloA* reference gene (**D**) and for SAT1 exon X inclusion. PSI was calculated by SAT1 exon X relative to SAT1 total mRNA amount (**E**). **F & G**. MCF7 cells were treated with PKAi for 30 minutes following by treatment with 200 ng/ml NCS for 1 h. Total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene (**F**) and for SAT1 exon X inclusion. PSI was calculated by SAT1 exon X relative to SAT1 total mRNA amount (**G**). **H & I**. SAT1 alternative splicing in MCF7 cells stably expressing either wild-type KDM3A or KDM3A mutated at serine 265 to alanine (S265A) or to aspartic acid (S265D). Total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene (**H**) and for SAT1 exon X inclusion. PSI was calculated by SAT1 exon X relative to SAT1 total mRNA amount (**I**). **J-L** MCF7 cells were transfected with non-targeting siRNA (siNT) and siSAT1 for 72 h, total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene (**J**). The amount of cells accumulated in S-phase is shown as a fold-change relative to untreated cells (**K**). Population histogram for one experiment is presented (**L**). **M–O.** MCF7 cells were mock transfected or transfected with 1 μM SAT1 oligo for 72 h, total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene (**M**). Semi-quantitative PCR results for SAT1 isoforms (ExX in, ExX out), in mock transfection SAT1 oligo (**N1**). Bar graph showing the volume of each band of SAT1 (ExX in, ExX out) for mock transfection SAT1 oligo (**N2**). Cells were treatmented with 200 ng/ml NCS for 8 h and analyzed using flow cytometry. A population histogram for one representative experiment is shown (**O**). **P-R3**. MCF7 cells stably expressing either the empty vector, wild-type KDM3A or KDM3A mutated demethylase (mut). Total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene (**P**) and real-time PCR for KDM3A relative to *CycloA* reference gene (**Q**). Semi-quantitative PCR results for SAT1 isoforms (ExX in, ExX out), in empty vector and wild-type KDM3A or KDM3A mutated demethylase (mut)(**R1**). Bar graph showing the volume of each band of SAT1 (ExX in, ExX out) for wild type KDM3A or KDM3A mut relative to empty vector (**R2&R3**). **S-W2**. MCF7 cells were transfected with non-targeting siRNA (siNT) or siG9a, siGLP for 72 h. Total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene**(S).** Semi-quantitative PCR results for SAT1 isoforms (ExX in, ExX out), in siNT and siG9a or siGLP (**T1& T2**). Bar graph showing the volume of each band of SAT1 (ExX in, ExX out) for siG9a or siGLP relative to siNT (**W1&W2**). **X & Y**. MCF7 cells were transfected with non-targeting siRNA (siNT) or siHP1α, β, γ for 72 h, total RNA was extracted and analyzed by real-time PCR for SAT1 relative to *CycloA* reference gene (**X**) and for SAT1 exon X inclusion. PSI was calculated by SAT1 exon X relative to SAT1 total mRNA amount (**Y**).

Supplementary Fig. S4



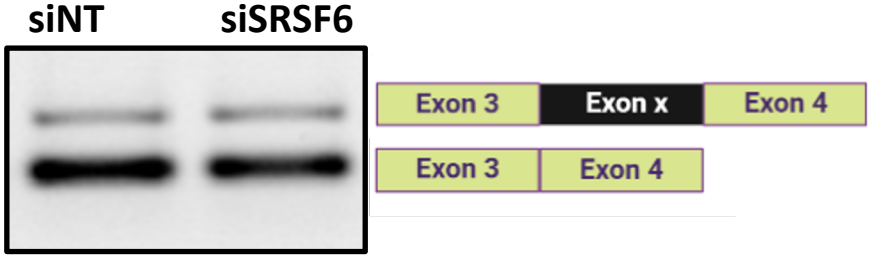
Supplementary Fig. S4

F1



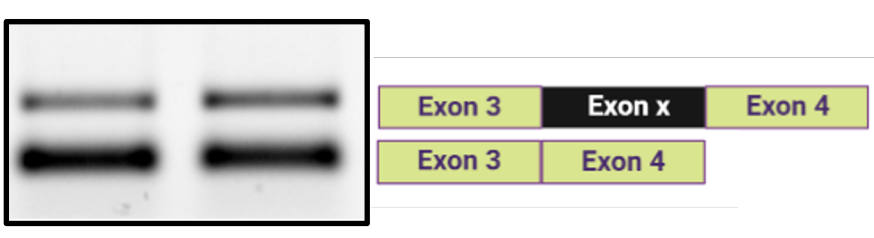

F2


F3

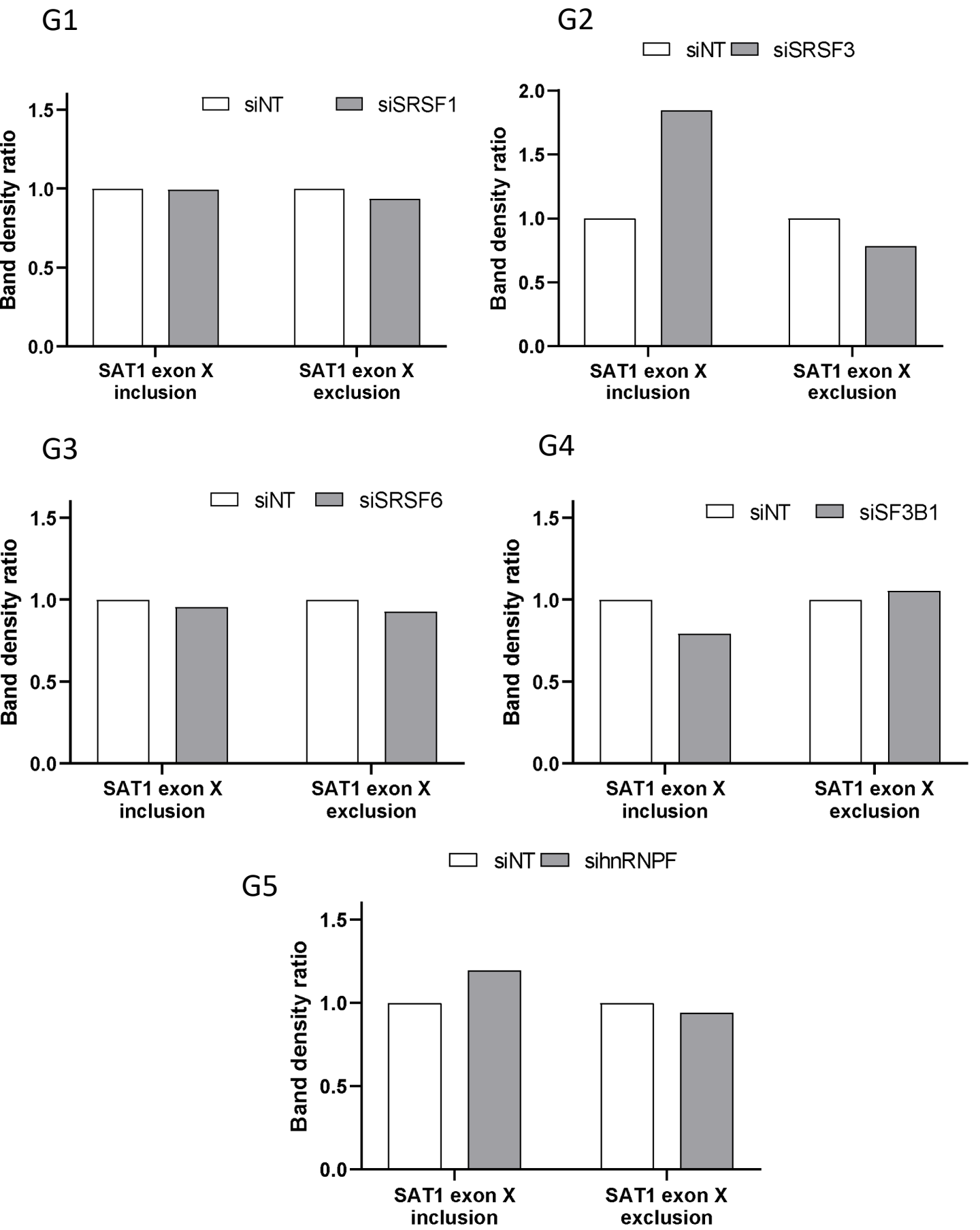



F4

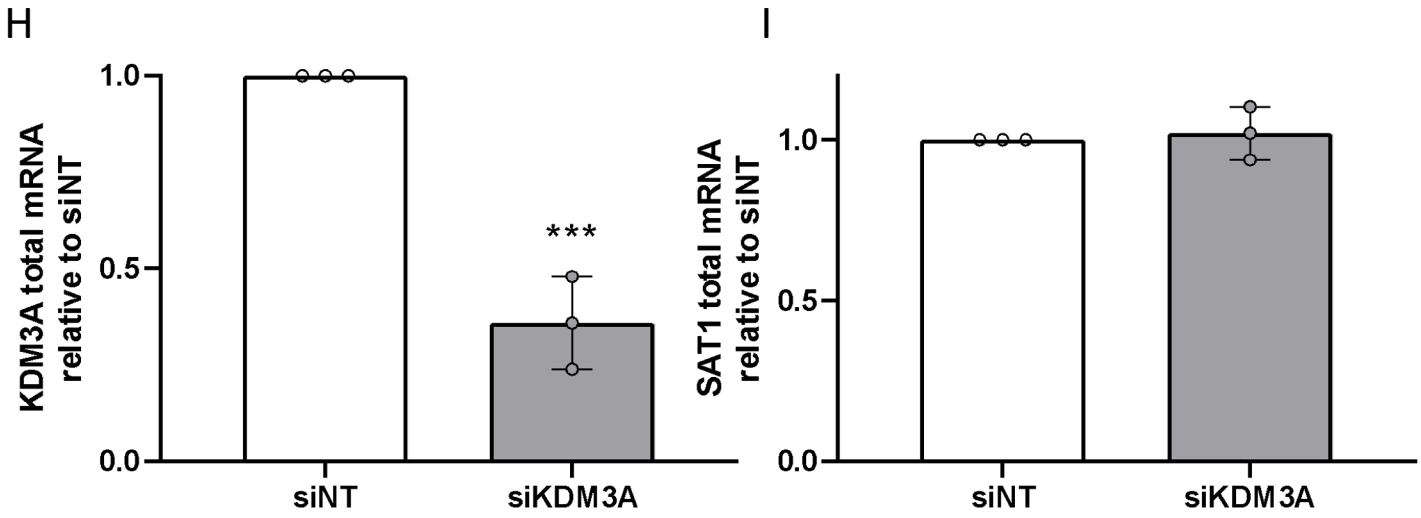

F5



Supplementary Fig. S4



Supplementary Fig. S4




**<u>Supplementary Figure S4</u>**: **A-C**. MCF7 cells were transfected with non-targeting siRNA (siNT) or siARID1A for 72 h. Total RNA was extracted and analyzed by real-time PCR for ARID1A (**A**) and for SAT1 (**B**) relative to *CycloA* reference gene and SAT1 exon X inclusion (**C**). PSI was calculated by SAT1 exon X relative to SAT1 total mRNA amount. Values in **A-C** represent averages of three independent experiments ±SD (* $p<0.05$, ** $p<0.01$; Student's t test comparing to siNT). **D-G5.** MCF7 cells were transfected with non-targeting siRNA (siNT) or siSF3B1, siSRSF1/3/6 and sihnRNPF for 72 h. Total RNA was extracted and analyzed by real-time PCR for each gene relative to *CycloA* reference gene (**D**) and SAT1 relative to *CycloA* reference gene (**E**). Values in **D** & **E** represent averages of three independent experiments ±SD (* $p<0.05$; Student's t test comparing to siNT). Semi-quantitative PCR results for SAT1 isoforms (ExX in, ExX out), in siNT or siSF3B1, siSRSF1/3/6 and sihnRNPF (**F1-F5**). Bar graph showing the volume of each band of SAT1 (ExX in, ExX out) for siSF3B1, siSRSF1/3/6 and sihnRNPF relative to siNT (**G1-G5**). **H&I**. MCF7 cells were transfected with non-targeting siRNA (siNT) or siKDM3A for 72 h. Total RNA was extracted and analyzed by real-time PCR for KDM3A (**H**) and for SAT1 (**I**) relative to *CycloA* reference gene. Values in H&I represent averages of three independent experiments ±SD (*** $p<0.001$; Student's t test comparing to siNT).

| Supplementary table S1. Primer used in this study | | |
|---|---|---|
| **Primer name** | **Description** | **Sequence** |
| F-CycloA | Endogenous control for qPCR | GTCAACCCCACCGTGTTCTT |
| R-CycloA | | CTGCTGTCTTTGGGACCTTGT |
| F-hTBP | | CGCCAGCTTCGGAGAGTTC |
| R-hTBP | | ACAACCAAGATTCACTGTGGATACA |
| F-KDM3A | Total KDM3A mRNA | TGAGCCACACAGACAGGTTG |
| R-KDM3A | | CAGCACCCTGTTGGCAATTC |
| F-G9a | Total G9a mRNA | CTGTCAGAGGAGTTAGGTTCTGC |
| R-G9a | | CTTGCTGTCGGAGTCCACG |
| F-SAT1 | Total SAT1 mRNA | TGTTTATCCGTCACTCGCCG |
| R-SAT1 | | TTTAGCCAGCTCCTTGATCAGC |
| F-SAT1-ExX | SAT1 isoform | TCCGTGTACATGGATGGGC |
| R-SAT1-Ex4 | | ATGGCAAAACCAACAATGCT |
| F-SRSF1 | Total SRSF1 mRNA | AGTGGTTGTCTCTGGACTGC |
| R-SRSF1 | | ACCACTGTATCCAATTCTGGTCAA |
| F-SRSF3 | Total SRSF3 mRNA | CTGATGCAGTCCGAGAGCT |
| R-SRSF3 | | ACCATTCGACAGTTCCACTC |
| F-SRSF6 | Total SRSF6 mRNA | CCACGCACAAGCCATAGGCGAT |
| R-SRSF6 | | CGACCTGGAACGGGAGCGACT |
| F-SF3B1 | Total SF3B1 mRNA | TGCAGATGGCTTCTATTCTGCTGC |
| R-SF3B1 | | GGGTTCCTGGGTTGCTAATCCGG |
| F-hnRNPF | Total hnRNPF mRNA | CGCCTCCGTGGACATTTCAT |
| R-hnRNPF | | GGAAGTTCTGCACGTCCTCA |
| F-HP1-gamma | Total HP1-gamma mRNA | AGTGGTCCCAAAGGGGTACT |
| R-HP1-gamma | | AGAGCTATTACGTTCGCGGC |
| F-HP1-alpha | Total HP1-alpha mRNA | TCCTGTTAGCCCAGAGGGAA |
| R- HP1-alpha | | TGTGCTCCTCAGAAAAGCCT |
| F-HP1-Beta | Total HP1-Beta mRNA | GCA AACCAAAGAAGAAGAAAGA |
| R-HP1-Beta | | ATAATCCGCTCCGGCTCCAAA |
| F-ARID1A | Total ARID1A mRNA | GAGATTGGTGGATTGACTCAG |
| R-ARID1A | | GTTGCAAGTTCCCGCCATT |
| F-ENSA-Ex3 | ENSA isoform | GCTTCTCTGTGCGGATGAAATG |
| R-ENSA-Ex4 | | GTGATCACCAGTCACCAGGTTC |
| F-ENSA | Total ENSA mRNA | TACAACATGGCCAAAGCCAAG |
| R-ENSA | | TGGTGACGAGCGAGGACTT |
| F-APTX-Ex2 | APTX isoform | GTGATGATGCGGGTGTGCTGGTT |
| R-APTX-Ex3 | | CTGCTTTACCTTGACATATCCCT |
| F-APTX | Total APTX mRNA | GAAGCGGTGAGGCACAGAT |
| R-APTX | | CACCCGCATCATCACTCTCC |
| F-RDM1-Ex5 | RDM1 isoform | CAGAAGCTTGCTATTCAGAAGGC |
| R-RDM1-Ex6 | | CCGTGTAGTTCTTCTTCGCATC |
| F-RDM1 | Total RDM1 mRNA | CCCACAGCATTCTCTGTTCACA |
| R-RDM1 | | CATTTGGAACTGTTCAGGGCAAGG |
| F-HUS1-Ex1 | HUS1 isoform | CTGTCTGAACCACTTCACAC |
| R-HUS1-Ex2 | | GTTCCAGCTCACACCACATGC |
| F-HUS1 | Total HUS1 mRNA | GCATGTGGTGTGAGCTGGAAC |
| R-HUS1 | | TCAAGGCTCGAGATAAGTTTTCC |

| | | |
|---|---|---|
| F-FIGNL1-Ex2 | FIGNL1 isoform | CTGCAGAAGCCAACTGTCTAGG |
| R-FIGNL1-Ex3 | | ATCACACAGCCAGTTATGAATGG |
| F-FIGNL1 | Total FIGNL1 mRNA | GCATTCAGTATGCATGGGC |
| R-FIGNL1 | | AGCTTGCATCATCTTCTGTACAC |
| F-TDRD1-Ex23 | TDRD1 isoform | GGGACTGTCGATGTAGCTGATA |
| R-TDRD1-Ex24 | | TTCTGTAACTCTGTGCAGCAGC |
| F-TDRD1 | Total TDRD1 mRNA | GATTGGAGGCGAGGGAAGTG |
| R-TDRD1 | | CATCGAGGCCTGGAAAGAGG |
| F-USP28-Ex3 | USP28 isoform | GTAATGGTGACATTACTCAGGCA |
| R-USP28-Ex4 | | CTGTTAAGATCTCTTCCATCAGC |
| F-USP28 | Total USP28 mRNA | TCTGAGGAACAGCAGCAAGA |
| R-USP28 | | GCTAGCTGGAATGCGTCCTC |
| F-STEAP-Ex2 | STEAP isoform | GTTCAAGCGATTCTCCTGCTTC |
| R-STEAP-Ex3 | | TACCCAGGATGCCCACTTTGG |
| F-STEAP | Total STEAP mRNA | CAGATCGGGCTGCTCAGCTTCTT |
| R-STEAP | | CACTCCCAGGGAGAGGTAGATC |
| F-SCML-Ex1 | SCML isoform | CGTGACAGGCTCTGTCACTAA |
| R-SCML-Ex2 | | CACATCGATTTCACTGGAGCTG |
| F-SCML | Total SCML mRNA | AACTGACCATGCTTCTGCAGCA |
| R-SCML | | AAGGGTGCTTAGTGATGCTTCC |
| F-HNRNPDL-Ex7 | HNRNPDL isoform | CCAGCCATACTAAAGGAGAAC |
| R-HNRNPDL-Ex8 | | CACAGGACAGCGTTTTCTGGT |
| F-HNRNPDL | Total HNRNPDL mRNA | CGAGCAAGAATCAGCAGGATGA |
| R-HNRNPDL | | GCTTGTATCCCAGCTCAAGCC |