Supplementary Data

Figure S1. A cytoplasmic version of COMPASS complex.

A) SET1B was knocked down in HeLa cells. The endogenous SET1B protein levels were detected by immunofluorescence, n=4. B) HEK293T cells were transfected with Flag-SET1B. Immunofluorescence was performed with anti-Flag 24 hours after transfection, n=4. The endogenous protein of SET1B in HMLE cells (C) and MCF7 cells (D) was detected by immunofluorescence, n=4.

Figure S2. SET1B but not its SET-domain maintains cell viability

A) The mRNA level of MLL1, MLL2, MLL3, MLL4 and SET1A was determined by real-time PCR in shNONT and shSET1B cells. B) SET1A was knocked down in five cell lines including HEK293T, MDA-MB-231, BT549, T47D, and MCF7 cells. The protein level of SET1A was determined by western blotting, n=3. C) Cells in panel B were selected with puromycin for 48 hours, 4×10^4 living cells were seeded in 6-well plates and grown for one week before crystal violet staining (level panel), n=4. D, E) Genome browser track examples of H3K4me1 and H3K4me3 ChIP-seq in MDA-MB-231 cells transduced with sh*NONT*, sh*SET1B*#1, and sh*SET1B*#2 for the indicated genes. The x-axis indicates the chromosome position, and the y-axis represents normalized read density in reads per million (rpm). F) Design of CRIPSR-Cas9 targeting *SET1B* gene locus. G) RNA-seq was performed with SET-domain wild type and knockout cells. The representative tracks for SET1B locus was shown. Red arrow points to the deletion region. H) The cDNA sequencing result from SET-domain-WT and –KO cells were shown.

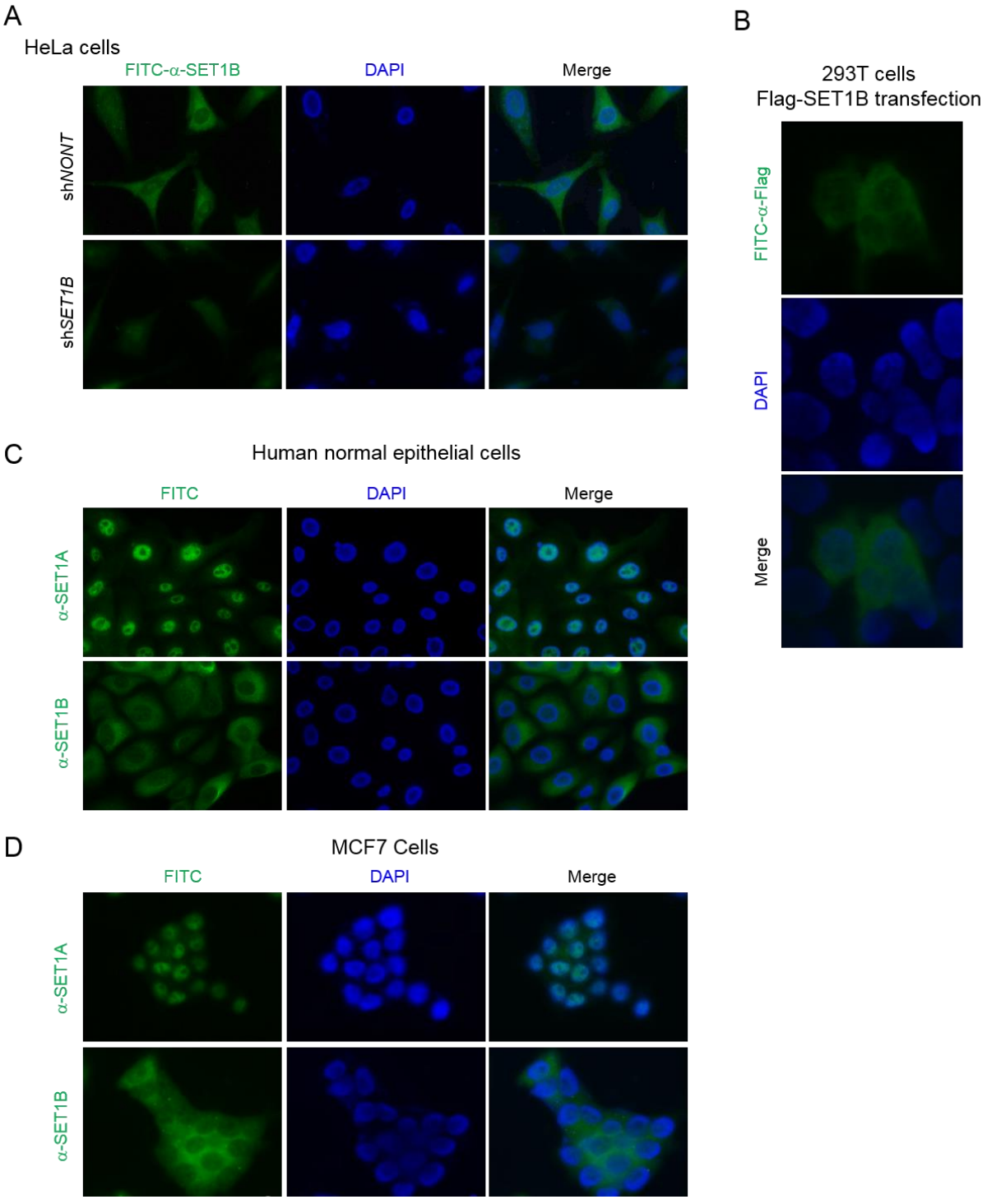
Figure S3. BOD1 is a cytoplasmic-specific subunit of SET1B/COMPASS.

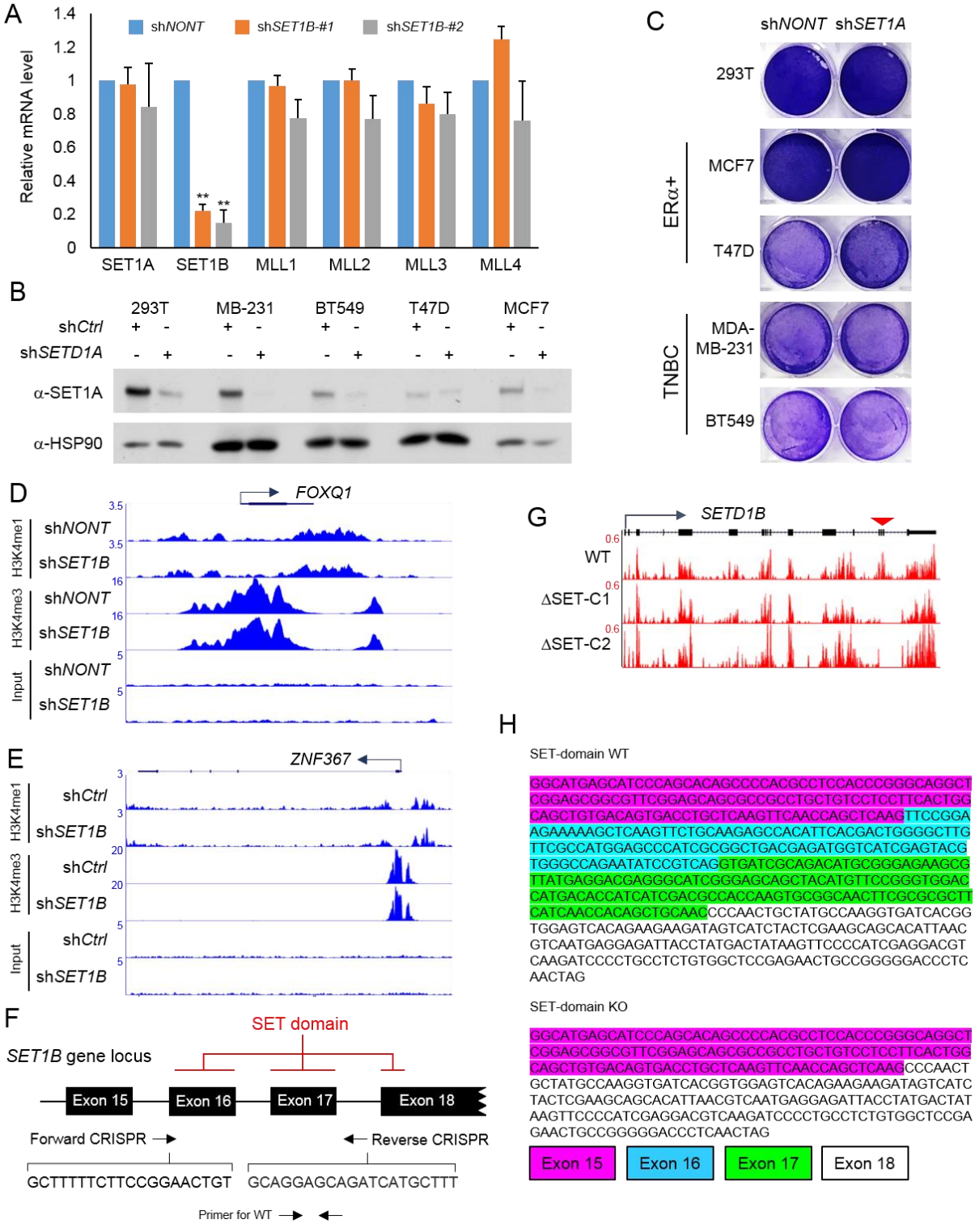
A) Peptide number of BOD1 and BOD1L co-purified with RBBP5 in Figure 3C are indicated in the table. B) HEK293T cells were transfected with plasmid expressing either GFP or Flag-tagged BOD1L. Nuclear extract was prepared from cells 48 hours after transfection and incubated with M2 beads. The interacting proteins of BOD1L were analyzed by mass spectrometry. Peptide number of BOD1L, SET1A, SET1B, and BOD1 are shown in the table. C) The protein levels of ASH2L, RBM15, and BOD1 were determined by Western blotting with purified cyto-SET1B COMPASS and nuc-SET1B COMPASS, n=3. D) The representative cell morphology of SET1B

or BOD1 knockdown cells are shown. The cytoskeleton of cells transfected with sh*NONT*, sh*SET1B*, or sh*BOD1* was detected by phalloidin staining, n=4. E) The growth rate of SET1B or BOD1 knocked down cells was determined by cell counting. n=4, Student's *t* test was used for statistical analysis. $**P < 0.01$; $*P < 0.05$. Error bars represent sd. F) MDA-MB-231 cells were infected with lentivirus expressing sh*NONT*, sh*SET1B*#1 and #2, and sh*BOD1*#1 and #2. Cells were selected with puromycin (2 ug/ml) 48 hours after virus infection. Total RNA was extracted from the cells subjected to RNA-seq, two replicates. The Venn diagram shows the common genes regulated by both SET1B and BOD1. G) Heat map analysis shows the genes significantly regulated by SET1B and BOD1.

Figure S4. Loss of SET1B or BOD1 induces the activation of ADIPOR1 signaling.
A) Heat map analysis shows the genes significantly regulated by SET1B in MDA-MB-231 cells. The genes involved in metabolic process are labeled. B) The representative RNA-seq tracks of *COQ7*, *SDC4* and *PRKAR2A* in MDA-MB-231 cells infected with sh*NONT*, sh*SET1B* virus are shown. C) The lipid droplet in sh*NONT* and sh*SET1B* cells were detected by Oil-O staining. D) Peptide number of HADHA/B co-purified with BOD1 in Figure 3E are indicated in the table. E) The protein level of HADHA was determined by western blot in sh*NONT*, sh*HADHA* and sh*SET1B* cells. F) Immunoprecipitation was performed using anti-SET1B antibody from either whole cell lysate. SET1B, RBBP5 and HADHA protein levels were detected in the immunoprecipitates by Western blot analysis. G) Western blot shows that the protein level of inducible GFP and GFP-AdipoR1 in cells. H) The representative RNA-seq track shows the AdipoR1 gene locus in AdipoR1-WT and –KO cells.

Wang_Supplemental_FigS1




A
shNONT
shSET1B-#1
shSET1B-#2
Relative mRNA level
SET1A
SET1B
MLL1
MLL2
MLL3
MLL4
B
293T
MB-231
BT549
T47D
MCF7
shCtrl
shSETD1A
α-SET1A
α-HSP90
C
shNONT shSET1A
293T
ERα+
MCF7
T47D
TNBC
MDA-MB-231
BT549
D
FOXQ1
H3K4me1
H3K4me3
Input
shNONT
shSET1B
E
ZNF367
shCtrl
shSET1B
F
SET domain
SET1B gene locus
Exon 15
Exon 16
Exon 17
Exon 18
Forward CRISPR
Reverse CRISPR
GCTTTTTCTTCCGGAACTGT
GCAGGAGCAGATCATGCTTT
Primer for WT
G
SETD1B
WT
ΔSET-C1
ΔSET-C2
H
SET-domain WT
GGCATGAGCATCCCAGCACAGCCCCACGCCTCCACCCGGGCAGGCT
CGGAGCGGCGTTCGGAGCAGCGCCGCCTGCTGTCCTCCTTCACTGG
CAGCTGTGACAGTGACCTGCTCAAGTTCAACCAGCTCAAGTTCCGGA
AGAAAAAGCTCAAGTTCTGCAAGAGCCACATTCACGACTGGGGCTTG
TTCGCCATGGAGCCCATCGCGGCTGACGAGATGGTCATCGAGTACG
TGGGCCAGAATATCCGTCAGGTGATCGCAGACATGCGGGAGAAGCG
TTATGAGGACGAGGGCATCGGGAGCAGCTACATGTTCCGGGTGGAC
CATGACACCATCATCGACGCCACCAAGTGCGGCAACTTCGCGCGCTT
CATCAACCACAGCTGCAAGCCCAACTGCTATGCCAAGGTGATCACGG
TGGAGTCACAGAAGAAGATAGTCATCTACTCGAAGCAGCACATTAAC
GTCAATGAGGAGATTACCTATGACTATAAGTTCCCCATCGAGGACGT
CAAGATCCCCTGCCTCTGTGGCTCCGAGAACTGCCGGGGGACCCTC
AACTAG
SET-domain KO
GGCATGAGCATCCCAGCACAGCCCCACGCCTCCACCCGGGCAGGCT
CGGAGCGGCGTTCGGAGCAGCGCCGCCTGCTGTCCTCCTTCACTGG
CAGCTGTGACAGTGACCTGCTCAAGTTCAACCAGCTCAAGCCCAACT
GCTATGCCAAGGTGATCACGGTGGAGTCACAGAAGAAGATAGTCATC
TACTCGAAGCAGCACATTAACGTCAATGAGGAGATTACCTATGACTAT
AAGTTCCCCATCGAGGACGTCAAGATCCCCTGCCTCTGTGGCTCCGA
GAACTGCCGGGGGACCCTCAACTAG
Exon 15
Exon 16
Exon 17
Exon 18

A

RBBP5 purification

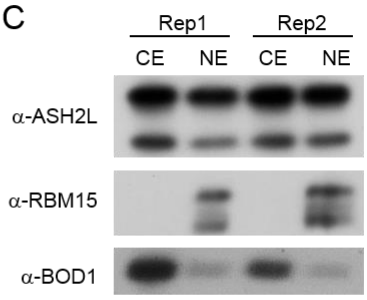
Spec count	Cytoplasm		Nucleus	
	GFP	RBBP5	GFP	RBBP5
BOD1	0	12	0	0
BOD1L	0	0	0	30

B

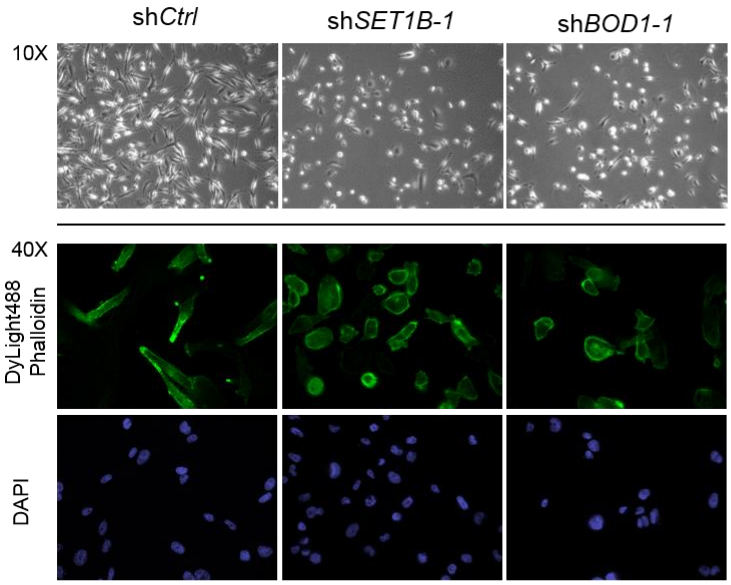
BOD1L purification

Spec count	GFP	Flag-BOD1L
BOD1L	13	548
SET1A	1	71
SET1B	0	2
BOD1	0	0

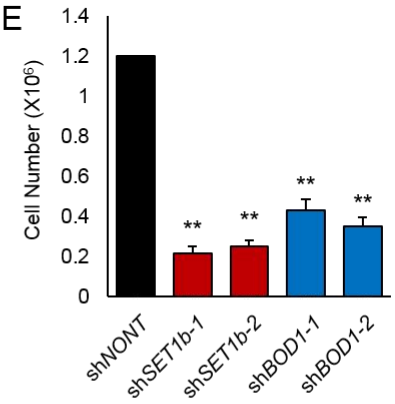
C



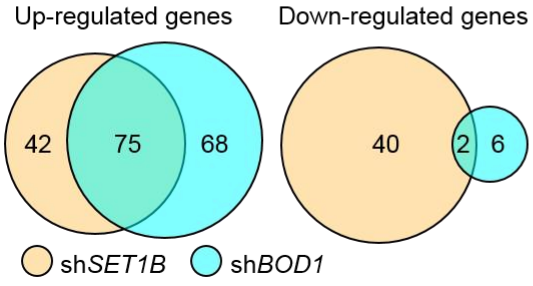
D



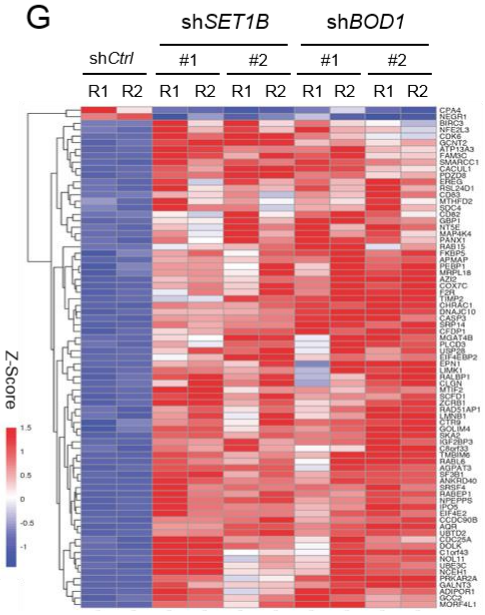
E

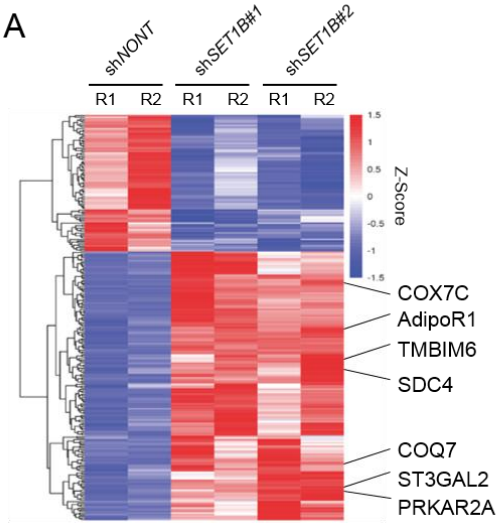


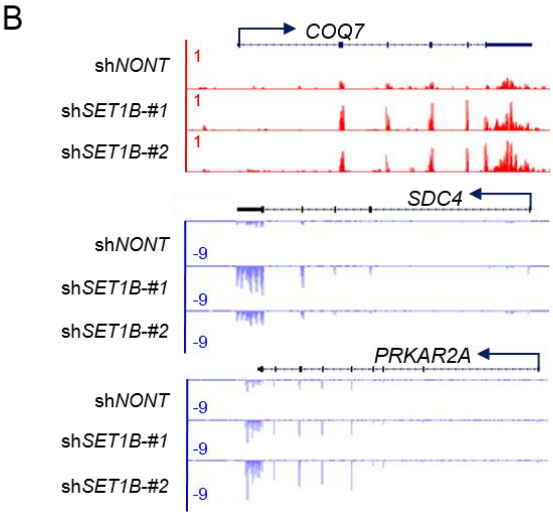
F

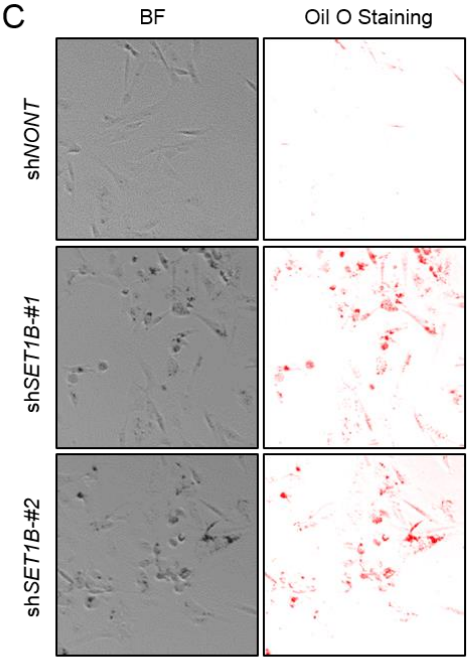


G









D

BOD1 purification

	GFP	BOD1
HADHA	0	18
HADHB	0	15