

Supplemental Information

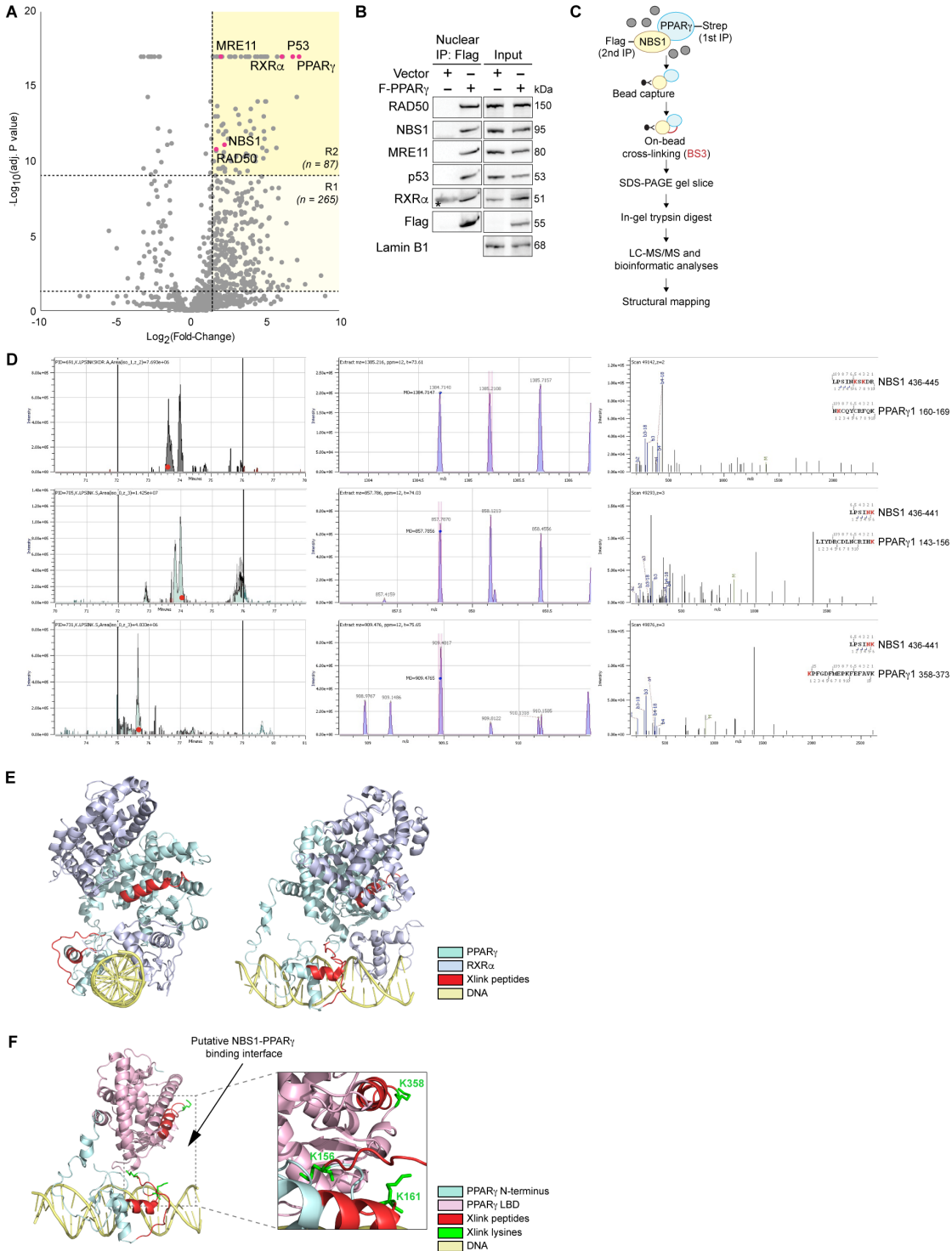
PPAR γ Interaction with UBR5/ATMIN

Promotes DNA Repair

to Maintain Endothelial Homeostasis

Caiyun G. Li, Cathal Mahon, Nathaly M. Sweeney, Erik Verschueren, Vivek Kantamani, Dan Li, Jan K. Hennigs, David P. Marciano, Isabel Diebold, Ossama Abu-Halawa, Matthew Elliott, Silin Sa, Feng Guo, Lingli Wang, Aiqin Cao, Christophe Guignabert, Julie Sollier, Nils P. Nickel, Mark Kaschwich, Karlene A. Cimprich, and Marlene Rabinovitch

1 **Supplementary Information**
 2 **Figure S1**



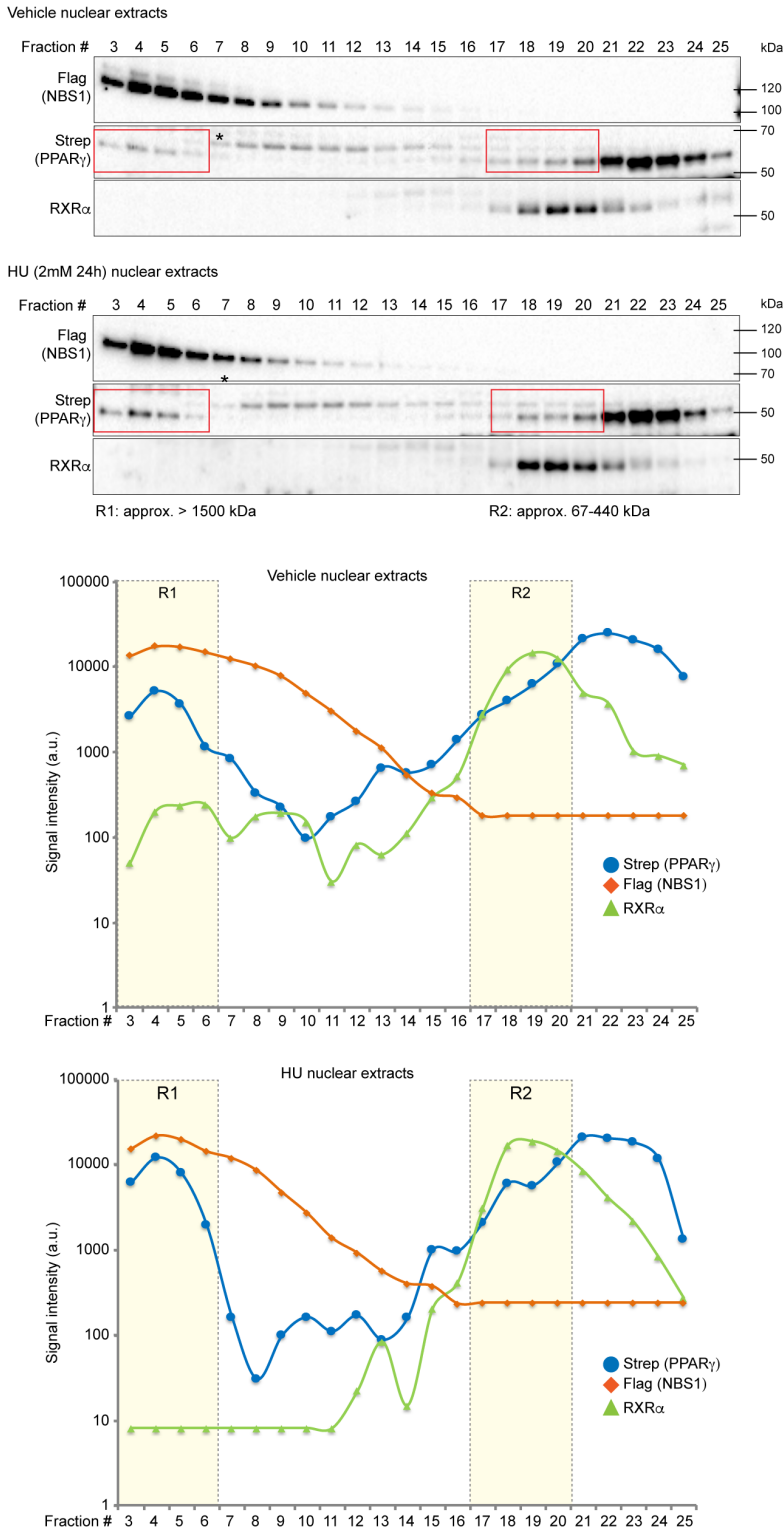
3 **Figure S1 PPAR γ interacts with the MRE11-RAD50-NBS1 (MRN) complex via NBS1 in 293T cells. – Related**
 4 **to Figure 1, Table S1-2.**

5 (A) AP-MS experiments yielded a volcano plot revealing 87 PPAR γ high confidence interactors (R2) out of the 352
 6 interactors (R1+R2). Four (in red) were validated in (C) Dotted lines represent cut-off at $\text{Log}_2(\text{Fold-Change}) > 1.5$
 7 and $\text{Log}_{10}(\text{adj. P value}) \leq 0.05$ and 1.0×10^{-9}). R1, region 1; R2, region 2, described in Results.
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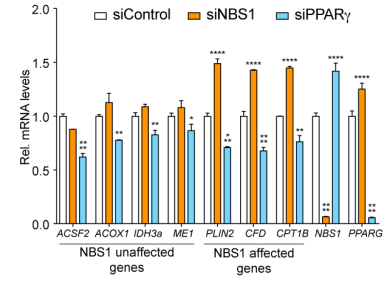
9 (B) Representative immunoblots of PPAR γ interactions in 293T nuclear extracts.
10 (C) Experimental set up of the BS3 crosslinking mass spectrometry (XL-MS) using tandem immunoprecipitation of
11 Flag-NBS1 and PPAR γ -Strep to determine NBS1 and PPAR γ binding interface.
12 (D) Raw MS/MS data of the identified crosslinked peptides from BS3 treated beads and trypsin-digested NBS1 and
13 PPAR γ . Amino acid positions of the peptides are as indicated. Analyses and methods are described in experimental
14 procedures.
15 (E) Structural mapping of the three PPAR γ peptides (red) identified in (D) to PPAR γ crystal structure (light green)
16 obtained from PDB:3DZU. This structure depicts PPAR γ and RXR α (blue) complex on DNA (yellow).
17 (F) Putative NBS1-PPAR γ binding interface as indicated by the locations of the three Xlink peptides (red). The
18 Xlink lysines (K) are labeled as green in inset. PPAR γ N-terminus is in light green, and its ligand-binding domain
19 (LBD) is in pink.
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26 **Figure S2**

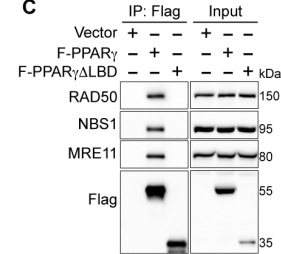
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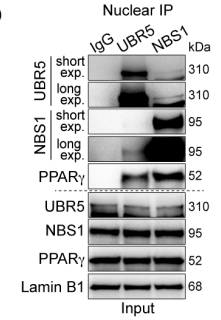
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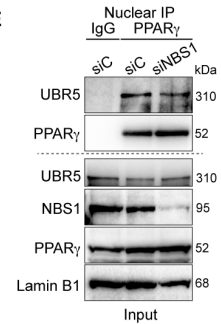
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Figure S2 Nuclear PPAR γ and NBS1 are in an independent cellular pool from PPAR γ and RXR α . – Related to Figure 1.

30 (A) 293T nuclear extracts expressing PPAR γ -2xStrep and Flag-NBS1 with and without HU treatment were analyzed
31 by size-exclusion chromatography on a Superose 6 gel filtration column. Proteins eluted from fractions #3-25 were
32 further analyzed by immunoblots to detect PPAR γ (anti-Strep), NBS1 (ant-Flag) and RXR α (anti-RXR α). Graphs
33 indicate densitometry results of each protein. Region 1 and 2 (R1, R2) are highlighted in red (in immunoblots) and
34 yellow (in graphs) to indicate PPAR γ eluted fractions. Since PPAR γ -2xStrep is approximately (approx.) a 60 kDa
35 protein (analyzed by silver staining in Fig. 1e), PPAR γ eluted from fraction #21 (approx. 67 kDa) might be the
36 excess monomeric form (due to overexpression). * indicates non-specific band.

37 (B) mRNA expression of PPAR γ target genes with NBS1 or PPAR γ depletion. mRNA expression was normalized to
38 *β -actin* mRNA.

39 (C) Representative immunoblots of interactions between Flag-PPAR γ (F-PPAR γ) without the ligand binding domain
40 (Δ LBD) and MRN.

41 (E) Representative immunoblots of endogenous co-immunoprecipitation (co-IP) between nuclear UBR5, NBS1 and
42 PPAR γ using anti-UBR5 or anti-NBS1.

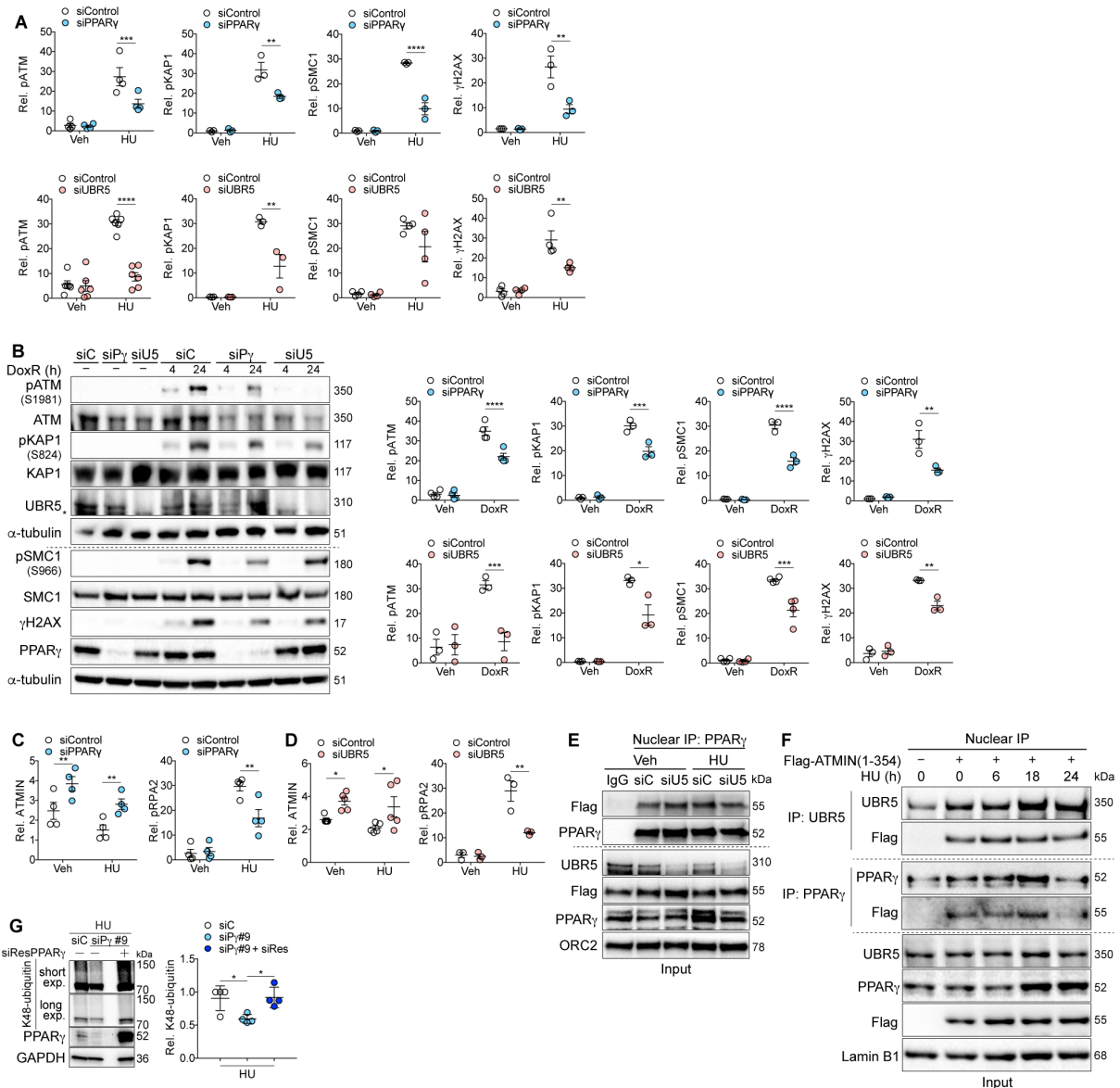
43 (F) Representative immunoblots of endogenous IP of PPAR γ with UBR5 with NBS1 depletion (siNBS1).

44 Error bars, mean \pm s.e.m. (B). siC, siControl. Two-way ANOVA test with Fisher's LSD test (B). *, P < 0.05, **, P <
45 0.01, ***, P < 0.001, ****, P < 0.0001.

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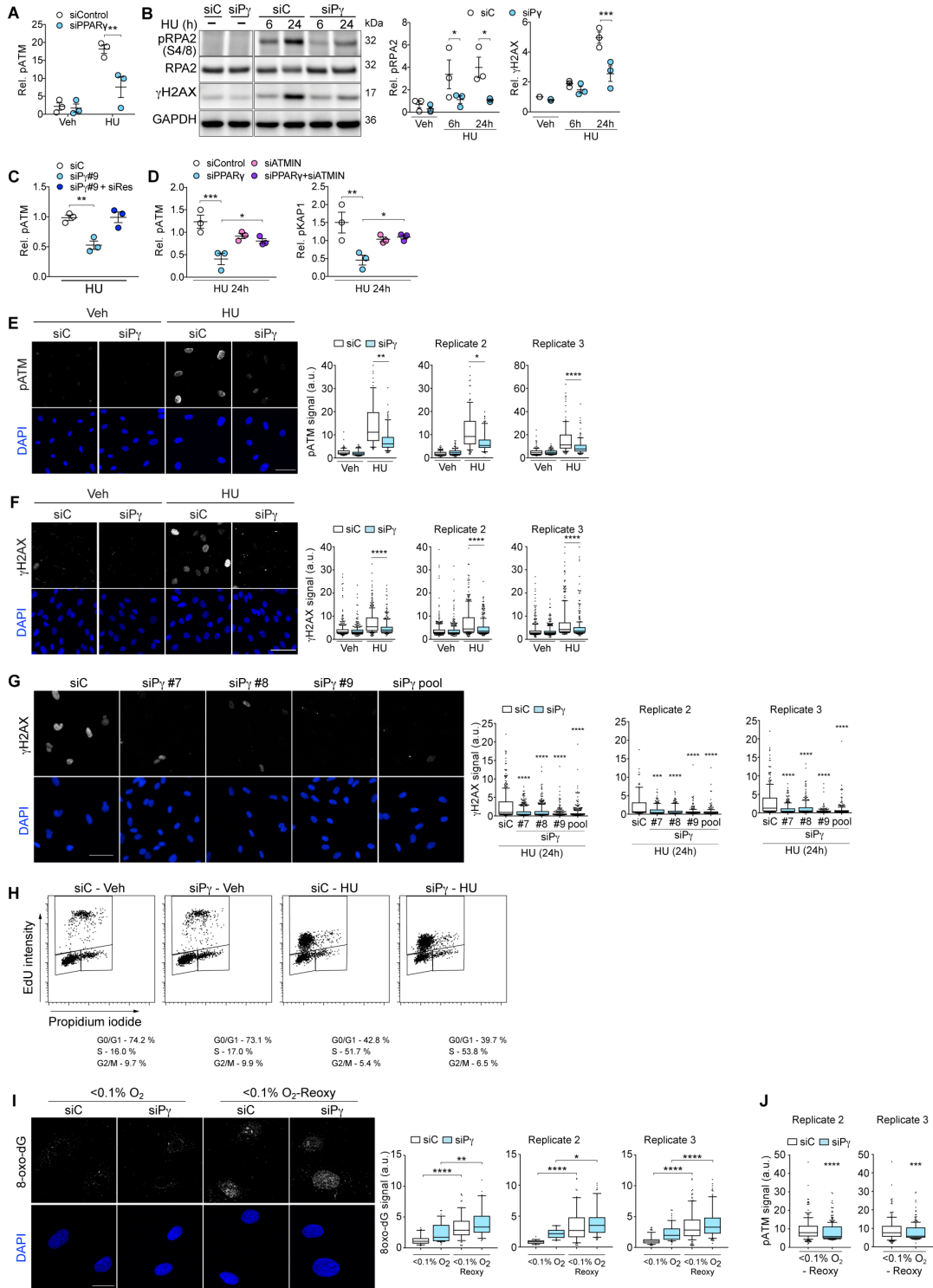
47 siC, siControl, siP γ , siPPAR γ , siU5, siUBR5. Error bars, mean \pm s.e.m. (B). siC, siControl. Two-way ANOVA test
48 with Fisher's LSD test (A-C). One-way ANOVA test with Fisher's LSD test (G). *, P < 0.05, **, P < 0.01, ***, P <
49 0.001, ****, P < 0.0001.

50 **Figure S3**



51
 52 **Figure S3 PPAR γ promotes ATM signaling by increasing ATMIN ubiquitination in 293T cells. – Related to**
 53 **Figure 2.**
 54 (A) Densitometry of HU-induced ATM signaling pathways, pATM, pKAP1, pSMC1, γ H2AX levels with PPAR γ or
 55 UBR5 depletion.
 56 (B) Representative immunoblots and densitometry data of DoxR-induced ATM signaling pathways, pATM, pKAP1,
 57 pSMC1, γ H2AX levels with PPAR γ or UBR5 depletions.
 58 (C) Densitometry data of HU-induced ATMIN and pRPA2 with PPAR γ depletion.
 59 (D) Densitometry data of HU-induced ATMIN and pRPA2 with UBR5 depletion.
 60 (E) Representative immunoblots of endogenous IP of nuclear PPAR γ with Flag-ATMIN (aa1-354) with UBR5
 61 depletion.
 62 (F) Representative immunoblots of endogenous IP of nuclear PPAR γ or UBR5 with Flag-ATMIN (aa1-354) with
 63 HU. The samples were harvested at the indicated time after HU treatment and lysates were separately incubated with
 64 anti-PPAR γ or anti-UBR5.
 65 (G) Representative immunoblots and densitometry data of restoration of K48-linked polyubiquitins with siRNA
 66 (siPPAR γ #9)-resistant PPAR γ overexpression.

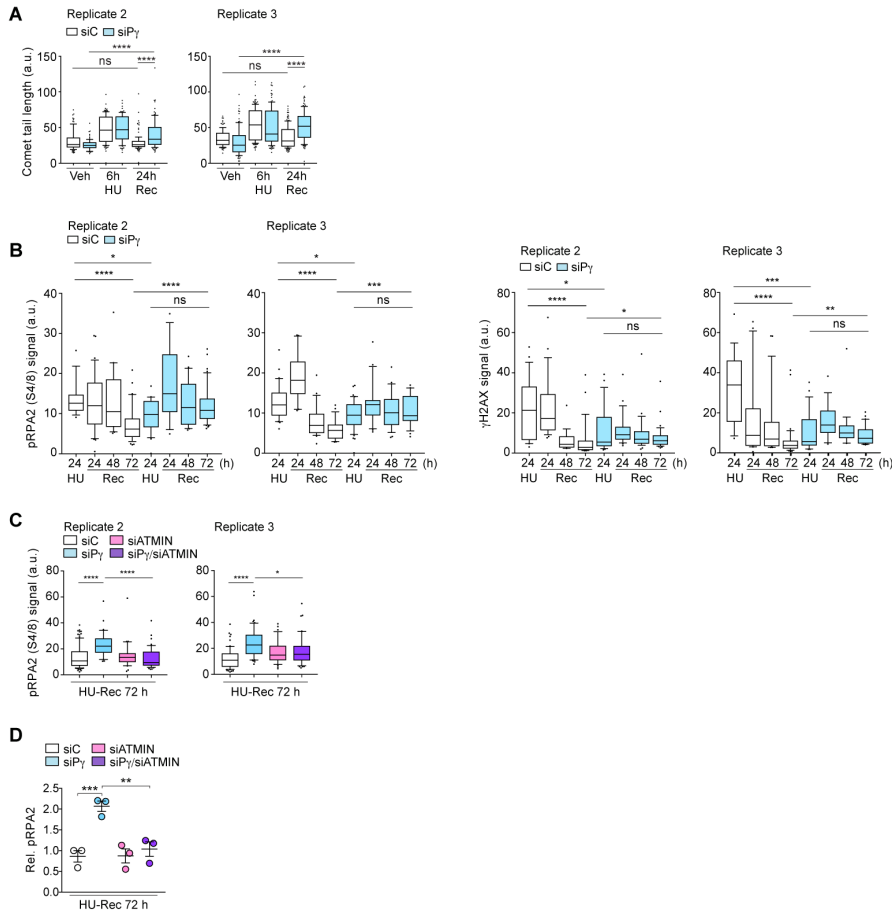
67 **Figure S4**



68
 69 **Fig. S4 PPAR γ depletion suppresses pATM and γ H2AX foci formation with various DNA damage stimuli and**
 70 **has no effects on cell cycle in primary endothelial cells. – Related to Figure 3.**
 71 (A) Densitometry data of HU-induced pATM with PPAR γ depletion in PAEC.
 72 (B) Representative immunoblots and densitometry data of HU-induced pRPA2 (S4/8) and γ H2AX protein levels
 73 with PPAR γ depletion in PAEC.

74 (C) Densitometry data of restoration of pATM with siRNA-resistant PPAR γ (siResPPAR γ) overexpression in
75 HUVEC.
76 (D) Densitometry data of HU-induced pATM and pKAP1 with PPAR γ or/and ATMIN depletion in PAEC.
77 (E) Confocal microscopy and triplicate quantitative data of effects of PPAR γ depletion on pATM foci formation
78 upon HU (24 hours) treatment in PAEC.
79 (F) Confocal microscopy and triplicate quantitative data of effects of PPAR γ depletion on γ H2AX foci formation
80 upon HU (24 hours) treatment in PAEC.
81 (G) Confocal microscopy and triplicate quantitative data of effects of PPAR γ depletion by multiple siRNAs on
82 γ H2AX foci formation upon HU (24 hours) treatment in PAEC. PPAR γ siRNAs (#7-9) are three out of the four
83 individual PPAR γ siRNAs used in the siP γ pool.
84 (H) Flow cytometry analysis of EdU incorporation shows cell cycle profiles of cells with PPAR γ depletion and with
85 HU (24 hours) treatment in PAEC.
86 (I) Confocal microscopy and triplicate quantitative data of 8-oxo-dG foci upon hypoxia (24 hours) (< 0.1% O₂) and
87 10 min reoxygenation (Reoxy) in PAEC.
88 (J) Replicate quantitative data of pATM foci with hypoxia-reoxygenation as described in (I).
89 siC, siControl, siP γ , siPPAR γ . Error bars, mean \pm s.e.m. The line in the box-and-whisker plots marks the median and
90 whiskers correspond to the 10th to 90th percentiles (E-G, I-J). Two-way ANOVA with Fisher's LSD test (A-D).
91 Kruskal-Wallis ANOVA test with Dunn's test (E-G, I). Unpaired t test (J). *, P < 0.05, **, P < 0.01, ***, P < 0.001;
92 ****, P < 0.0001. Scale bars, E-G, 50 μ m; I, 20 μ m.
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94 **Figure S5**



95
 96 **Fig. S5 PPAR γ depletion leads to persistent pRPA2 and γ H2AX foci after HU-induced damage which are**
 97 **resolved by ATMIN depletion in PAEC. – Related to Figure 4.**

98 (A) Replicate quantitative data of extended comet tail lengths in PPAR γ depleted cells after HU (6 hours) was
 99 removed (recovery for 24 hours).

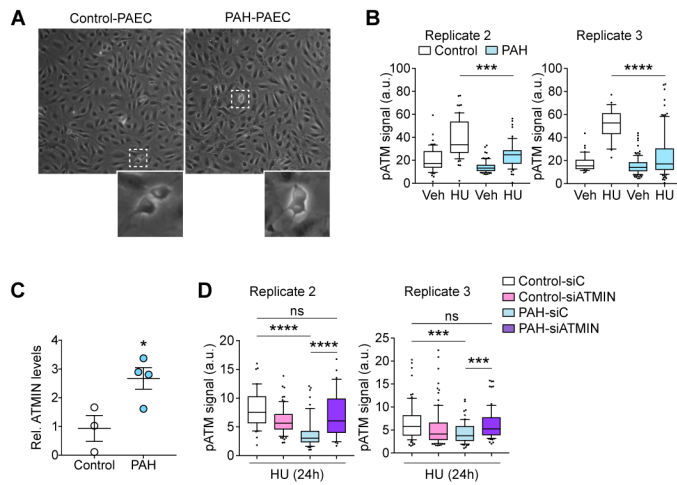
100 (B) Replicate quantitative data of pRPA2 and γ H2AX foci in PPAR γ depleted cells after HU (24 hours) was
 101 removed. Cells were fixed at various recovery time points as indicated.

102 (C) Replicate quantitative data of resolution of pRPA2 foci with ATMIN depletion in addition to PPAR γ depletion.

103 (D) Densitometry data show restoration of pRPA2 protein levels with ATMIN depletion in addition to PPAR γ
 104 depletion.

105 siC, siControl, siP γ , siPPAR γ . Error bars, mean \pm s.e.m. The line in the box-and-whisker plots marks the median and
 106 whiskers correspond to the 10th to 90th percentiles (A-C). Kruskal–Wallis ANOVA test with Dunn’s test (A-C).

107 Two-way ANOVA with Fisher’s LSD test (D). *, P < 0.05, **, P < 0.01, ***, P < 0.001; ****, P < 0.0001, ns, not
 108 significant.



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 111 **Fig. S6 PPAR γ and ATMIN axis is impaired in PAEC from PAH patients. – Related to Figure 5.**
 112 (A) Phase-contrast microscopy shows healthy primary PAEC cultures established from explanted lungs of control
 113 (unused donors) and PAH patient. Inserts indicate mitotic cells from the dotted square.
 114 (B) Replicate quantitative data of HU-induced pATM levels in control and PAH-PAEC.
 115 (C) Densitometry data of ATMIN protein levels in control and PAH-PAEC.
 116 (D) Replicate quantitative data of pATM levels in control and PAH-PAEC with ATMIN depletion.
 117 siC, siControl, siP γ , siPPAR γ . Error bars, mean \pm s.e.m. The line in the box-and-whisker plots marks the median and
 118 whiskers correspond to the 10th to 90th percentiles (B,D). Kruskal–Wallis ANOVA test with Dunn’s test (B,D).
 119 Unpaired t test (C). *, P < 0.05, ***, P < 0.001; ****, P < 0.0001, ns, not significant.
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Table S1 High-confidence PPAR γ interactors identified from AP-MS. – Related to Figure 1; Figure S1.

Uniprot ID	Log2FC	Adj. P value	Gene name	Protein name
P37231	7.32	0	PPARG	Peroxisome proliferator-activated receptor gamma
P04637	6.9	0	TP53	Cellular tumor antigen p53
P11142	6.22	0	HSPA8	Heat shock cognate 71 kDa protein
O95816	6.22	0	BAG2	BAG family molecular chaperone regulator 2
P19793	6.17	0	RXRA	Retinoic acid receptor RXR-alpha
P23396	5.86	0	RPS3	40S ribosomal protein S3
P52272	5.18	0	HNRNPM	Heterogeneous nuclear ribonucleoprotein M
P31943	5.08	0	HNRNPH1	Heterogeneous nuclear ribonucleoprotein H, N-terminally processed
P49411	4.97	0	TUFM	Elongation factor Tu, mitochondrial
P49368	4.82	0	CCT3	T-complex protein 1 subunit gamma
Q92841	4.74	0	DDX17	Probable ATP-dependent RNA helicase DDX17
P08107	4.53	0	HSPA1B	Heat shock 70 kDa protein 1A/1B
P17987	4.38	0	TCP1	T-complex protein 1 subunit alpha
P55072	3.98	0	VCP	Transitional endoplasmic reticulum ATPase
Q9UL15	3.91	0	BAG5	BAG family molecular chaperone regulator 5
Q08211	3.87	0	DHX9	ATP-dependent RNA helicase A
P18669	3.74	0	PGAM1	Phosphoglycerate mutase 1
P48047	3.43	0	ATP5O	ATP synthase subunit O, mitochondrial
P52209	3.43	0	PGD	6-phosphogluconate dehydrogenase, decarboxylating
Q9Y265	3.05	0	RUVBL1	RuvB-like 1
Q9Y230	3.03	0	RUVBL2	RuvB-like 2
P38646	3	0	HSPA9	Stress-70 protein, mitochondrial
P11021	2.74	0	HSPA5	78 kDa glucose-regulated protein
P13796	2.65	0	LCP1	Plastin-2
P50990	2.65	0	CCT8	T-complex protein 1 subunit theta
O14830	2.16	0	PPEF2	Serine/threonine-protein phosphatase with EF-hands 2
P47929	2.11	0	LGALS7	Galectin-7
P49959	2.08	0	MRE11A	Meiotic recombination 11 MRE11A
Q02880	2	0	TOP2B	DNA topoisomerase 2-beta
Q9H0A0	1.83	0	NAT10	N-acetyltransferase 10
Q9Y490	1.79	0	TLN1	Talin-1
P15927	1.58	0	RPA2	Replication protein A 32 kDa subunit
P49916	1.55	0	LIG3	DNA ligase 3
Q9UBQ0	1.54	0	VPS29	Vacuolar protein sorting-associated protein 29
P62241	7.17	5.57E-15	RPS8	40S ribosomal protein S8
Q86U86	5.1	1.02E-14	PBRM1	Protein polybromo-1
Q7Z2W4	5.06	1.02E-14	ZC3HAV1	Zinc finger CCCH-type antiviral protein 1
Q92945	4.55	1.02E-14	KHSRP	Far upstream element-binding protein 2
Q6Y7W6	1.83	2.01E-14	GIGYF2	PERQ amino acid-rich with GYF domain-containing protein 2
P33993	4.09	2.95E-14	MCM7	DNA replication licensing factor MCM7
P17066	3.06	3.37E-14	HSPA6	Heat shock 70 kDa protein 6
Q13263	3.26	8.51E-14	TRIM28	Transcription intermediary factor 1-beta
Q9H583	1.86	1.02E-13	HEATR1	HEAT repeat-containing protein 1, N-terminally processed
P42704	1.71	1.05E-13	LRPPRC	Leucine-rich PPR motif-containing protein, mitochondrial
P62906	4.96	1.07E-13	RPL10A	60S ribosomal protein L10a
Q14151	4.04	2.02E-13	SAFB2	Scaffold attachment factor B2
Q15393	3.32	3.40E-13	SF3B3	Splicing factor 3B subunit 3
P78316	2.24	3.40E-13	NOP14	Nucleolar protein 14
P62424	2.14	3.78E-13	RPL7A	60S ribosomal protein L7a

P32969	5.6	6.05E-13	RPL9P7	60S ribosomal protein L9
P78347	4.3	6.89E-13	GTF2I	General transcription factor II-I
Q99615	4.96	9.02E-13	DNAJC7	DnaJ homolog subfamily C member 7
Q15029	3.75	9.05E-13	EFTUD2	116 kDa U5 small nuclear ribonucleoprotein component
Q9NVP1	2.43	9.05E-13	DDX18	ATP-dependent RNA helicase DDX18
O75368	1.91	1.10E-12	SH3BGRL	SH3 domain-binding glutamic acid-rich-like protein
O95786	2.4	1.11E-12	DDX58	Probable ATP-dependent RNA helicase DDX58
P28702	4.1	1.50E-12	RXRβ	Retinoic acid receptor RXR-beta
P61978	2.25	1.66E-12	HNRNPK	Heterogeneous nuclear ribonucleoprotein K
Q96EY1	5.01	2.90E-12	DNAJA3	DnaJ homolog subfamily A member 3, mitochondrial
Q8N1F7	2.57	3.12E-12	NUP93	Nuclear pore complex protein Nup93
O60934	2.32	8.83E-12	NBS1	Nibrin
O00231	4.86	1.18E-11	PSMD11	26S proteasome non-ATPase regulatory subunit 11
Q12906	5.8	1.22E-11	ILF3	Interleukin enhancer-binding factor 3
Q92878	1.77	1.47E-11	RAD50	DNA repair protein RAD50
O60264	1.76	2.00E-11	SMARCA5	SWI/SNF-related matrix-associated actin-dependent regulator of chromatin subfamily A member 5
O15042	5.15	2.02E-11	U2SURP	U2 snRNP-associated SURP motif-containing protein
O76021	2.28	2.53E-11	RSL1D1	Ribosomal L1 domain-containing protein 1
P62191	3.77	3.35E-11	PSMC1	26S protease regulatory subunit 4
P46087	3.91	3.67E-11	NOP2	Probable 28S rRNA (cytosine(4447)-C(5))-methyltransferase
Q9Y2R4	3.68	4.88E-11	DDX52	Probable ATP-dependent RNA helicase DDX52
Q9BVJ6	4.41	4.97E-11	UTP14A	U3 small nucleolar RNA-associated protein 14 homolog A
O15144	2.26	5.08E-11	ARPC2	Actin-related protein 2/3 complex subunit 2
Q12905	3.1	5.40E-11	ILF2	Interleukin enhancer-binding factor 2
P62701	4.93	5.77E-11	RPS4X	40S ribosomal protein S4, X isoform
Q9BZE4	2.7	7.08E-11	GTPBP4	Nucleolar GTP-binding protein 1
O75367	2.76	9.21E-11	H2AFY	Core histone macro-H2A.1
P15924	1.62	9.21E-11	DSP	Desmoplakin
Q9Y2X3	4.4	9.27E-11	NOP58	Nucleolar protein 58
Q8WVV4	1.54	1.01E-10	POF1B	Protein POF1B
P17931	2.23	2.46E-10	LGALS3	Galectin-3
Q9BVP2	2.07	3.14E-10	GNL3	Guanine nucleotide-binding protein-like 3
P26038	2.36	3.16E-10	MSN	Moesin
P06702	1.91	3.16E-10	S100A9	Protein S100-A9
P07900	3.08	3.80E-10	HSP90AA1	Heat shock protein HSP 90-alpha
P31689	3.87	4.49E-10	DNAJA1	DnaJ homolog subfamily A member 1
O15511	2.34	5.01E-10	ARPC5	Actin-related protein 2/3 complex subunit 5
O15160	3.33	5.70E-10	POLR1C	DNA-directed RNA polymerases I and III subunit RPAC1
O43818	1.72	9.22E-10	RRP9	U3 small nucleolar RNA-interacting protein 2

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123 Proteins co-purified with Flag-PPAR γ from 293T nuclear extracts were detected by mass spectrometry and analyzed
124 as described in Experimental Procedures. Proteins in red were validated in Fig. 1c. Log₂FC (Fold-Change) indicates
125 the fold enrichment of proteins immunoprecipitated from Flag-PPAR γ expressing cells as compared to Flag-vector
126 expressing cells. The adjusted (adj.) P values are indicated and the data was obtained from four independent
127 experiments. Proteins are listed by their adj. P values.

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Table S2
Biological functions associated with the high-confidence PPAR γ interactome. – Related to Figure 1; Figure S1.

Uniprot ID	Gene name	Biological function name
O15144	ARPC2	Actin cytoskeleton organization
O15511	ARPC5	Actin cytoskeleton organization
P47929	LGALS7	Cell-cell interaction
Q8WVV4	POF1B	Cell-cell interaction
Q9Y490	TLN1	Cell-cell interaction
P48047	ATP5O	Cellular metabolism
O95816	BAG2	Cellular metabolism
Q9UL15	BAG5	Cellular metabolism
P18669	PGAM1	Cellular metabolism
P52209	PGD	Cellular metabolism
P37231	PPARG	Cellular metabolism
P19793	RXRA	Cellular metabolism
P28702	RXRB	Cellular metabolism
P49411	TUFM	Cellular metabolism
Q9UBQ0	VPS29	Cellular metabolism
Q08211	DHX9	DNA damage response and replication
P31689	DNAJA1	DNA damage response and replication
Q96EY1	DNAJA3	DNA damage response and replication
O75367	H2AFY	DNA damage response and replication
P07900	HSP90AA1	DNA damage response and replication
P08107	HSPA1B	DNA damage response and replication
P11142	HSPA8	DNA damage response and replication
Q92945	KHSRP	DNA damage response and replication
P49916	LIG3	DNA damage response and replication
P42704	LRPPRC	DNA damage response and replication
P33993	MCM7	DNA damage response and replication
P49959	MRE11A	DNA damage response and replication
O60934	NBN	DNA damage response and replication
Q8N1F7	NUP93	DNA damage response and replication
Q86U86	PBRM1	DNA damage response and replication
P62191	PSMC1	DNA damage response and replication
O00231	PSMD11	DNA damage response and replication
Q92878	RAD50	DNA damage response and replication
P15927	RPA2	DNA damage response and replication
Q9Y265	RUVBL1	DNA damage response and replication
Q9Y230	RUVBL2	DNA damage response and replication
Q14151	SAFB2	DNA damage response and replication
O60264	SMARCA5	DNA damage response and replication
Q02880	TOP2B	DNA damage response and replication
P04637	TP53	DNA damage response and replication
Q13263	TRIM28	DNA damage response and replication
P55072	VCP	DNA damage response and replication
O95786	DDX58	Innate immune response
P13796	LCPI	Innate immune response
P17931	LGALS3	Innate immune response
P06702	S100A9	Innate immune response
Q7Z2W4	ZC3HAV1	Innate immune response
Q92841	DDX17	mRNA metabolic process and RNA processing
Q9NVP1	DDX18	mRNA metabolic process and RNA processing
Q9Y2R4	DDX52	mRNA metabolic process and RNA processing

Q15029	EFTUD2	mRNA metabolic process and RNA processing
Q6Y7W6	GIGYF2	mRNA metabolic process and RNA processing
Q9BVP2	GNL3	mRNA metabolic process and RNA processing
Q9BZE4	GTPBP4	mRNA metabolic process and RNA processing
Q9H583	HEATR1	mRNA metabolic process and RNA processing
P31943	HNRNPH1	mRNA metabolic process and RNA processing
P61978	HNRNPK	mRNA metabolic process and RNA processing
P52272	HNRNPM	mRNA metabolic process and RNA processing
Q12905	ILF2	mRNA metabolic process and RNA processing
Q12906	ILF3	mRNA metabolic process and RNA processing
Q9H0A0	NAT10	mRNA metabolic process and RNA processing
P78316	NOP14	mRNA metabolic process and RNA processing
P46087	NOP2	mRNA metabolic process and RNA processing
Q9Y2X3	NOP58	mRNA metabolic process and RNA processing
P62906	RPL10A	mRNA metabolic process and RNA processing
P62424	RPL7A	mRNA metabolic process and RNA processing
P32969	RPL9P7	mRNA metabolic process and RNA processing
P23396	RPS3	mRNA metabolic process and RNA processing
P62701	RPS4X	mRNA metabolic process and RNA processing
P62241	RPS8	mRNA metabolic process and RNA processing
O43818	RRP9	mRNA metabolic process and RNA processing
O76021	RSL1D1	mRNA metabolic process and RNA processing
Q15393	SF3B3	mRNA metabolic process and RNA processing
O15042	U2SURP	mRNA metabolic process and RNA processing
Q9BVJ6	UTP14A	mRNA metabolic process and RNA processing
P49368	CCT3	Protein folding
P50990	CCT8	Protein folding
Q99615	DNAJC7	Protein folding
P15924	DSP	Protein folding
P11021	HSPA5	Protein folding
P17066	HSPA6	Protein folding
P38646	HSPA9	Protein folding
P26038	MSN	Protein folding
P17987	TCP1	Protein folding
P78347	GTF2I	NA
O15160	POLR1C	NA
O14830	PPEF2	NA
O75368	SH3BGRL	NA

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Biological functions enriched among the top ranked 87 PPAR γ interactors defined in Supplementary Table 1. NA, not available.

135 **Table S3**
 136 **Tandem affinity purification identified proteins interacting with PPAR γ and NBS1 in unperturbed cells. –**
 137 **Related to Figure 1.**

Uniprot ID	Log ₂ FC	Adj. P value	Gene name	Protein name
O60934	6.24	0	NBS1	Nibrin
P37231	6.03	0	PPARG	Peroxisome proliferator-activated receptor gamma
P49959	5.22	0	MRE11A	Meiotic recombination 11 MRE11A
Q92878	5.01	0	RAD50	DNA repair protein RAD50
O95816	3.7	1.71E-08	BAG2	BAG family molecular chaperone regulator 2
Q9UL15	2.9	4.03E-07	BAG5	BAG family molecular chaperone regulator 5
O75594	2.56	2.25E-02	PGLYRP1	Peptidoglycan recognition protein 1
P04637	2.46	7.99E-09	TP53	Cellular tumor antigen p53
P62269	2.2	0	RPS18	40S ribosomal protein S18
Q07021	2.1	2.05E-08	C1QBP	Complement component 1 Q subcomponent-binding protein, mitochondrial
P23588	1.96	4.10E-10	EIF4B	Eukaryotic translation initiation factor 4B
Q02878	1.93	0	RPL6	60S ribosomal protein L6
P18124	1.9	1.60E-13	RPL7	60S ribosomal protein L7
P34932	1.84	0	HSPA4	Heat shock 70 kDa protein 4
P50914	1.8	0	RPL14	60S ribosomal protein L14
O95071	1.8	3.37E-02	UBR5*	E3 ubiquitin-protein ligase UBR5
P98175	1.78	4.19E-11	RBM10	RNA-binding protein 10
P62701	1.66	7.94E-07	RPS4X	40S ribosomal protein S4, X isoform
P62333	1.65	2.25E-03	PSMC6*	26S protease regulatory subunit 10B
Q9Y2W1	1.55	7.34E-11	THRAP3*	Thyroid hormone receptor-associated protein 3
P11142	1.54	0	HSPA8	Heat shock cognate 71 kDa protein

138
 139 Proteins were co-purified with PPAR γ -2xStrep and sequentially with Flag-NBS1 from 293T whole cell extracts
 140 [unperturbed cells or with HU treatment (Supplementary Table 4)]. Log₂FC indicates the fold enrichment of proteins
 141 immunoprecipitated from the PPAR γ -2xStrep and Flag-NBS1 expressing cells as compared to GFP-Strep-Flag
 142 expressing cells (negative control). The adjusted P value is indicated and the data were obtained from three
 143 independent experiments.

144 * indicates proteins specifically enriched in unperturbed cells but not present in the HU treated cells. THRAP3 and
 145 UBR5 (red) are validated in Fig. 1e, f. Proteins are listed by the Log₂FC.

146 **Table S4**
 147 **Tandem affinity purification identified proteins interacting with PPAR γ and NBS1 upon HU treatment. –**
 148 **Related to Figure 1.**

Uniprot ID	Log ₂ FC	Adj. P value	Gene name	Protein name
O60934	6.25	0	NBS1	Nibrin
P37231	6.09	0	PPARG	Peroxisome proliferator-activated receptor gamma
P49959	5.02	0	MRE11A	Meiotic recombination 11 MRE11A
Q92878	4.78	0	RAD50	DNA repair protein RAD50
O95816	3.88	6.05E-09	BAG2	BAG family molecular chaperone regulator 2
Q9UL15	3.65	7.27E-08	BAG5	BAG family molecular chaperone regulator 5
Q92552	3.16	3.37E-02	MRPS27*	28S ribosomal protein S27, mitochondrial
O75594	2.7	2.17E-02	PGLYRP1	Peptidoglycan recognition protein 1
Q02878	2.61	0	RPL6	60S ribosomal protein L6
P18124	2.58	0	RPL7	60S ribosomal protein L7
Q07021	2.36	1.18E-09	C1QBP	Complement component 1 Q subcomponent-binding protein, mitochondrial
P04637	2.08	6.15E-07	TP53	Cellular tumor antigen p53
P50914	2.07	0	RPL14	60S ribosomal protein L14
P43686	1.89	1.08E-07	PSMC4*	26S protease regulatory subunit 6B
Q9BRT6	1.87	4.32E-02	LLPH*	Protein LLP homolog
O60318	1.83	2.30E-02	MCM3AP*	Germinal-center associated nuclear protein
P11388	1.82	3.49E-09	TOP2A*	DNA topoisomerase 2-alpha
P23588	1.79	1.21E-09	EIF4B	Eukaryotic translation initiation factor 4B
P62269	1.78	0	RPS18	40S ribosomal protein S18
P98175	1.76	1.56E-11	RBM10	RNA-binding protein 10
Q75N03	1.73	1.62E-02	CBLL1*	E3 ubiquitin-protein ligase Hakai
P40429	1.71	2.38E-02	RPL13A*	60S ribosomal protein L13a
Q02880	1.7	3.50E-04	TOP2B*	DNA topoisomerase 2-beta
P36578	1.69	9.59E-14	RPL4*	60S ribosomal protein L4
Q9HAN9	1.67	3.64E-04	NMNAT1*	Nicotinamide/nicotinic acid mononucleotide adenylyltransferase 1
P62917	1.65	0	RPL8*	60S ribosomal protein L8
Q07020	1.64	6.87E-10	RPL18*	60S ribosomal protein L18
P62266	1.62	6.37E-07	RPS23*	40S ribosomal protein S23
P62847	1.61	3.98E-09	RPS24*	40S ribosomal protein S24
P34932	1.61	8.63E-14	HSPA4	Heat shock 70 kDa protein 4
P62701	1.59	9.46E-07	RPS4X	40S ribosomal protein S4, X isoform
P11142	1.58	0	HSPA8	Heat shock cognate 71 kDa protein
P62158	1.56	2.83E-06	CALM2*	Calmodulin
P62249	1.55	5.53E-09	RPS16*	40S ribosomal protein S16
P62241	1.54	1.43E-08	RPS8*	40S ribosomal protein S8
P49368	1.51	2.20E-02	CCT3*	T-complex protein 1 subunit gamma

149
 150 Proteins were co-purified with PPAR γ -2xStrep and sequentially with Flag-NBS1 from 293T whole cell extracts with
 151 HU treatment. Log₂FC (Fold-Change) indicates the fold enrichment of proteins immunoprecipitated from the
 152 PPAR γ -2xStrep and Flag-NBS1 expressing cells as compared to GFP-Strep-Flag expressing cells (negative control).
 153 The adjusted P value is indicated and the data were obtained from three independent experiments. * indicates
 154 proteins specifically enriched in the HU treated cells but not present in unperturbed cells. Proteins are listed by the
 155 Log₂FC.

156 **Table S5 – Related to STAR Methods.**
 157 **Characteristics of (a) control subjects and (b) PAH patients used in this study.**
 158 (a) Control

Control ID	Cells/ Tissues	Assays	Age (yr)/ Gender	Race/ Ethnicity	Cause of Death
Control-1	Tissue	IF	41/F	White/ Non- Hispanic	Grade 4 subarachnoid hemorrhage, ruptured anterior cerebral artery aneurysm
Control-2	Tissue	IF	43/M	White/ Non- Hispanic	Fatal gunshot to head
Control-3	Tissue	IF	57/F	White/ Non- Hispanic	Intracranial hemorrhage/stroke
Control-4	Tissue	IF	28/F	White/ Non- Hispanic	MVC-anoxia
Control-5	SPAEC	Comet	47/M	White/ Non- Hispanic	Head trauma-bicycle vs. car accident
Control-6	Tissue	IF	56/F	White/ Non- Hispanic	Cerebrovascular accident
Control-7	SPAEC	Comet	55/F	Unknown/ Hispanic or Latino	Cerebrovascular stroke
Control-8	Tissue/ LPAEC	IF, comet, protein expression	57-F	White/ Non- Hispanic	Acute myocardial infarction
Control-9	LPAEC	Comet	12/M	Unknown/ Non- Hispanic	Head trauma rollover MVC ejection
Control-10	LPAEC	Comet	49/M	White/ Non- Hispanic	Head trauma
Control-11	SPAEC	Protein expression	33/F	White/ Non- Hispanic	Head trauma. Blunt injury.

Control-12	LPAEC	IP, protein expression	54/M	Unknown/Hispanic or Latino	Cerebrovascular/stroke ICH
Control-13	LPAEC	IP, protein expression	34/F	Asian/Non-Hispanic	Cerebrovascular/stroke ICH
Control-14	LPAEC	IP	1/M	White/Non-Hispanic	Anoxia/drowning
Control-15*	LPAEC	IP, protein expression	35/M	White/Non-Hispanic	Gunshot wound
Control-16	LPAEC	IP, protein expression	46/M	Asian/Unknown	Cerebrovascular/stroke ICH

159

160 (b) PAH patients

Patient ID	Cells/Tissues	Assays	Age (yr)/Gender	Race/Ethnicity	Diagnosis	BMPR2 mutation	(s/d/m)PAPa	PVRb (mmHg)	6 Min Walkc (Wood Units)	PAH Medications (m), up to transplant date
PAH-1	Tissue	IF	15-F	White/Non-Hispanic	IPAH	No	(175/66/102)	25.24	387	sildenafil, epoprostenol
PAH-2	Tissue	IF	40-F	White/Non-Hispanic	IPAH	No	(84/26/47)	NA	294	ambrisentan, sildenafil, iloprost, epoprostenol
PAH-3	LPAEC	Protein expresso pm	33-F	Black or African American/Non-Hispanic	FPAH	Yes	(75/33/48)	15.57	326.1	epoprostenol, bosentan, sildenafil, treprostinil
PAH-4	Tissue	IF	56-F	White/Non-Hispanic	IPAH	No	(83/39/57)	11.41	137.2	sildenafil, ambrisentan, treprostinil
PAH-5	Tissue,S/ LPAEC	IF, comet,	27-F	White/Non-Hispanic	IPAH	Yes	(110/49/69)	12.11	359.7	sildenafil, treprostinil, bosentan, iloprost

PAH-6	SPAEC	Comet	40-M	White/ Hispanic or Latino	IPAH	No	(118/49/64)	73	420	sildenafil, ambrisentan, treprostinil
PAH-7	SPAEC	Comet	37-M	White/ Non- Hispanic	FPAH	Yes	(119/51/77)	14.22	309	sildenafil, sitaxsentan, ambrisentan, epoprostenol, imatinib (investigational), treprostinil
PAH-8	LPAEC	Comet, IP, protein expression	32-F	White/ Non- Hispanic	IPAH	No	(68/38/49)	15.34	238	bosentan, epoprostenol
PAH-9	Tissue/ LPAEC	IF, comet, IP, protein expression	30-M	White/ Non- Hispanic	APAH- Congenita I ASD	No	(128/60/85)	NA	160	sildenafil, bosentan
PAH-10	LPAEC	IP, protein expression	16-F	White/ Non- Hispanic	IPAH	N/A	(NA/NA/95)	N/A	102.4	sildenafil, subcutaneous treprostinil
PAH-11	LPAEC	Protein expression	50-F	White/ Hispanic or Latino	APAH- D&T	N/A	(113/43/65)	16.18	384	sildenafil, ambrisentan, bosentan, treprostinil, epoprostenol
PAH-12	LPAEC	Protein expression	22-F	White/ Non- Hispanic	FPAH	Yes	(98/46/66)	10.19	506	sildenafil, ambrisentan, tadalafil, treprostinil

161

162 a (s/d/m) PAP= Systolic, diastolic, and mean pulmonary arterial pressure. b PVR= Pulmonary vascular resistance. c 6 min walk = distance walked in six minutes.

163 a-c Values are closest to transplant date. * Control line that did not meet PHBI inclusion criteria due to known history of amphetamine/methamphetamine use.

164 Abbreviations: SPAEC, small pulmonary arterial endothelial cells (<1 mm by dissection); LPAEC, large PAEC (>1 mm); IF, immunofluorescence; IP,

165 immunoprecipitation; F, female; M, male; MVC, motor vehicle accident; ICH, intracranial hemorrhage; IPAH, idiopathic pulmonary arterial hypertension;

166 FPAH, familiar PAH; APAH, associated PAH; ASD, atrial septal defect ; D&T, drug and toxin; NA, not available.

167 **Table S6 – Related to STAR Methods.**
 168 **Table of Oligonucleotides used.**
 169

Oligo	Sequence (5'-3')	Description
PPAR γ -delLBD_F	GTGGCCATCCGCATCTGACAGGGCTGCCAGTTTCG	Primer used for generating Flag-PPAR γ Δ LBD
PPAR γ -delLBD_R	CGAAACTGGCAGCCCTGTCAGATGCCGATGGCCAC	Primer used for generating Flag-PPAR γ Δ LBD
2Strep-PPAR γ -F	TAGTCCAGTGTGGTGGAAATTCGCCGCATGACCATGGTTGACACAG	Primer used for generating 2xStrep-PPAR γ
2Strep-PPAR γ -R	CACCGCCTCCCTCGAGCGGCCGCACGTACAAGTCCTTGATG	Primer used for generating 2xStrep-PPAR γ
siRES-PPAR γ _F	ATGACAGCGATCTCGCAATATTTATTGCTGTCAATTATC	Primer used for generating siRNA-resistant-Flag-PPAR γ
siRES-PPAR γ _R	TAAATATTGCGAGATCGCTGTCATCTAATTCCAGTG	Primer used for generating siRNA-resistant-Flag-PPAR γ
PPAR γ _F	GCCGTGGCCGCAGATTTGAAAGA	Primer used for quantitative real-time PCR
PPAR γ _R	TACGGAGAGATCCACGGAGCTGA	Primer used for quantitative real-time PCR
UBR5_F	CCTGCTCACTGCTACTAATCTG	Primer used for quantitative real-time PCR
UBR5_R	ATTCGAGGTGGCCTGTATTG	Primer used for quantitative real-time PCR
ATMIN_F	AACAGCACTGCAGTCTCACAA	Primer used for quantitative real-time PCR
ATMIN_R	CTGGTCTAGGGATTGGTTGGT	Primer used for quantitative real-time PCR
NBS1_F	CACTCACCTTGTCATGGTATCAG	Primer used for quantitative real-time PCR
NBS1_R	CTGCTTCTTGGACTCAA CTGC	Primer used for quantitative real-time PCR
PLIN2_F	ATGGCATCCGTTGCAGTTGAT	Primer used for quantitative real-time PCR
PLIN2_R	GGACATGAGGTCATACGTGGAG	Primer used for quantitative real-time PCR
ACOX1_F	GGAACCTCACCTTCGAGGCTTG	Primer used for quantitative real-time PCR
ACOX1_R	TTCCCCTTAGTGATGAGCTGG	Primer used for quantitative real-time PCR
CPT1B_F	CCTGCTACATGGCAACTGCTA	Primer used for quantitative real-time PCR
CPT1B_R	AGAGGTGCCCAATGATGGGA	Primer used for quantitative real-time PCR
ACSF2_F	ATGGCTGTCTACGTCCGGG	Primer used for quantitative real-time PCR
ACSF2_R	GACCATGCGATCCACCTCTC	Primer used for quantitative real-time PCR
ME1_F	CTGCTGACACGGAACCCTC	Primer used for quantitative real-time PCR
ME1_R	GATCTCCTGACTGTTGAAGGAAG	Primer used for quantitative real-time PCR
CFD_F	GACACCATCGACCACGACC	Primer used for quantitative real-time PCR
CFD_R	GCCACGTCGCAGAGAGTTC	Primer used for quantitative real-time PCR
IDH3A_F	GGACCTGGAGGAAAGTGGAT	Primer used for quantitative real-time PCR
IDH3A_R	GCTGCTATTGGGGTCTTCAA	Primer used for quantitative real-time PCR
β -actin_F	CATGCCATCCTGCGTCTGGA	Primer used for quantitative real-time PCR
β -actin_R	CCGTGGCCATCTCTTGCTCG	Primer used for quantitative real-time PCR