

1 **TGF- β signaling alters H4K20me3 status via miR-29 and contributes to cellular**
2 **senescence and cardiac aging**

3 Guoliang et al.

4

5

6

7

8

9

10

11

12

13

14

15

16

17

18

19

20

21

22

23

24

25

26

27

28

29

30

31

32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62

Supplementary Figures

63

64

65

66

67

68

69

70

71

72

73

74

75

76

77

78

79

80

81

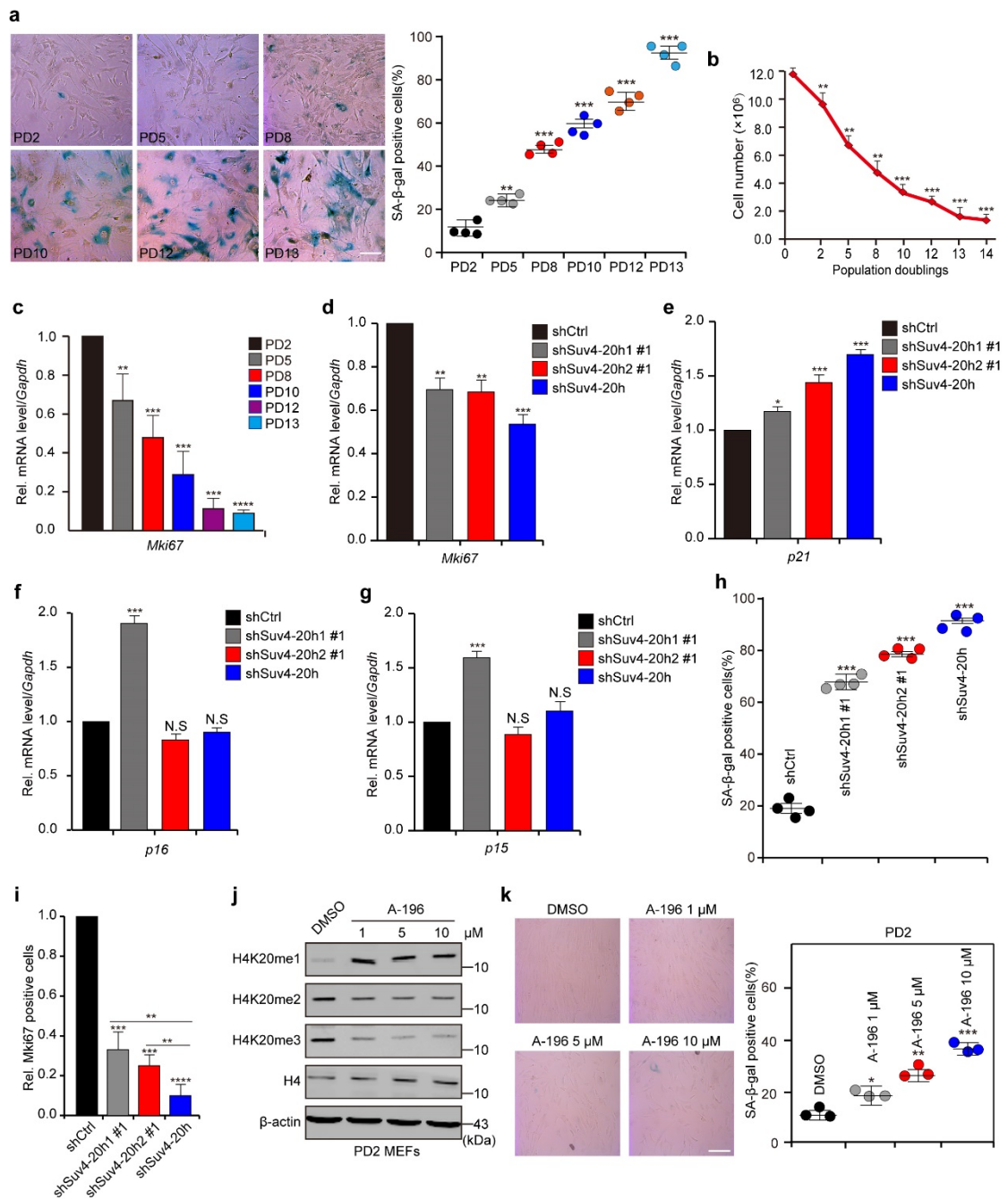
82

83

84

85

86



87 **Supplementary Figure 1. Loss of H4K20me3 promotes emergence of senescent**
 88 **phenotypes.**

89 **a-c**, MEFs progressively accumulated positive SA-β-gal stained cells and showed a
 90 decreased proliferation rate over time. MEFs from different growth stages were
 91 subjected to SA-β-gal staining (**a**), growth curve analysis (**b**) and *Mki67* measurement
 92 by RT-qPCR (**c**). Scale bar, 20 μm. ***p*<0.01, ****p*<0.001 (One-way ANOVA with
 93 Dunnett's multiple comparison test). **d-g**, RT-qPCR measurements of *Mki67* (**d**), *p21*

94 (e), p16 (f), and p15 (g) in Suv4-20h1 knockdown (shSuv4-20h1), Suv4-20h2
95 knockdown (shSuv4-20h2) or both (shSuv4-20h) cells. **h**, Statistical analysis of SA- β -
96 gal staining in **Figure. 1i**. **i**, Statistical analysis of the Mki67 signal in **Figure. 1j**. **j**,
97 Western blot for changes in H4K20 methylation in cells treated with the indicated doses
98 of A-196. β -actin served as a loading control. **k**, Cells from **j** were subjected to SA- β -
99 gal staining (left) and analyzed (right, One-way ANOVA with Dunnett's multiple
100 comparison test). Scale bar, 20 μ m. The error bars represent the s.d. obtained from
101 triplicate independent experiments. Two-tailed unpaired Student's t-tests were
102 performed. **p<0.01, ***p<0.001.

103

104

105

106

107

108

109

110

111

112

113

114

115

116

117

118

119

120

121

122

123

124

125

126

127

128

129

130

131

132

133

134

135

136

137

138

139

140

141

142

143

144

145

146

147

148

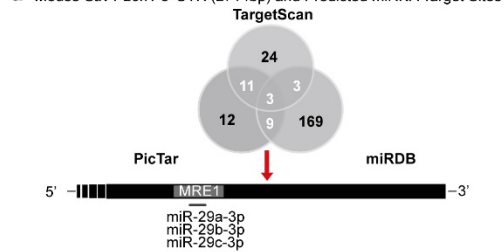
149

150

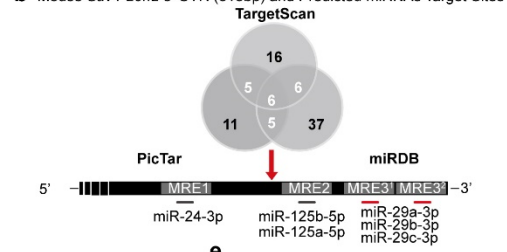
151

152

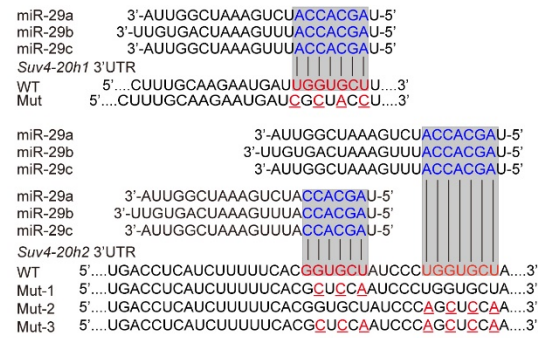
a Mouse *Suv4-20h1* 3' UTR (2714bp) and Predicted miRNA Target Sites



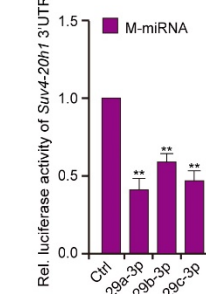
b Mouse *Suv4-20h2* 3' UTR (613bp) and Predicted miRNAs Target Sites



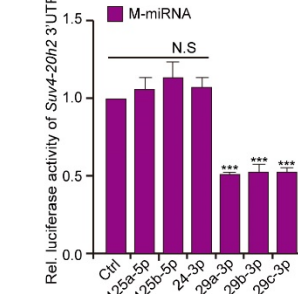
c



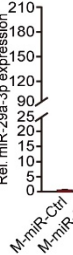
d



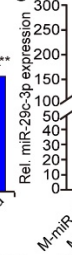
e



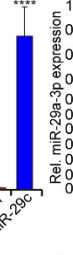
f



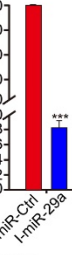
g



h



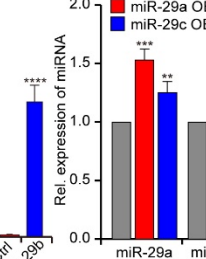
i



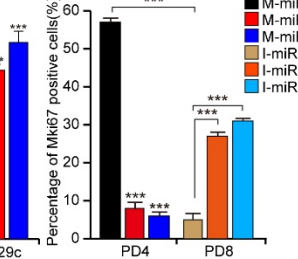
j



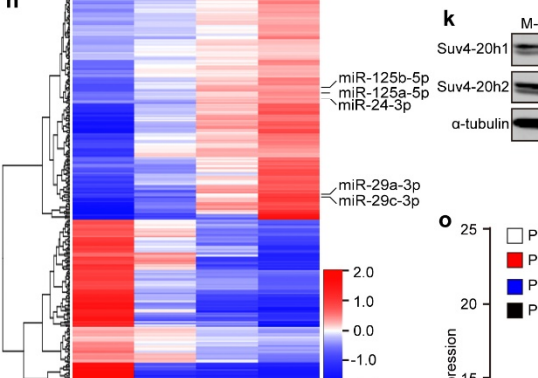
m



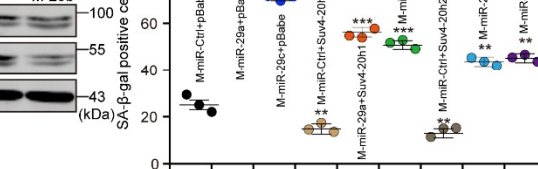
p



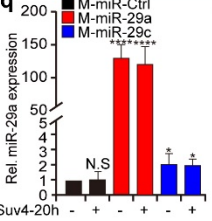
n



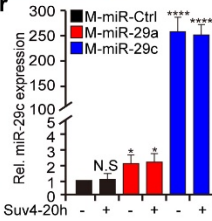
k



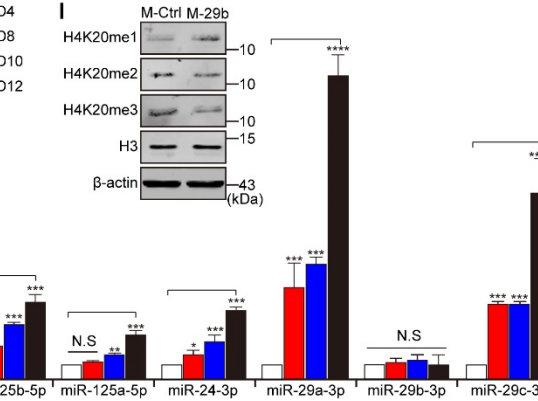
q



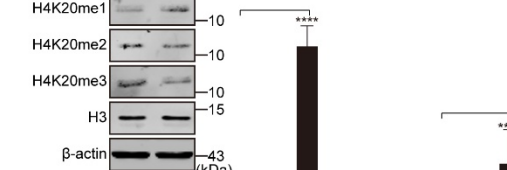
r



o



l



Supplementary Figure 2. miR-29 targets Suv4-20h to accelerate cellular senescence.

a, b, Schematic diagrams showing the of miRNAs predicted to target the 3' UTR of

156 *Suv4-20h1* (**a**) or *Suv4-20h2* (**b**) by analyzing three different miRNA databases
157 (TargetScan, PicTar and miRDB). **c**, Representative mode of miR-29 targeting the 3'
158 UTRs of *Suv4-20h1* and *Suv4-20h2*. **d, e**, Luciferase activity assays showed cells co-
159 transfected with miRNA mimics and constructs containing the 3' UTR of *Suv4-20h1* (**d**)
160 or 3' UTR of *Suv4-20h2* (**e**). **f-j**, RT-qPCR showed expression of the indicated miRNA
161 after transfection with the corresponding miRNA mimics or inhibitors. **k, l**, Western
162 blots showed changes in Suv4-20h protein (**k**) and H4K20 methylation (**l**). α -tubulin
163 and β -actin served as loading controls. **m**, RT-qPCR measurements of lentivirus-
164 induced expression of miR-29. **n, o**, Differential miRNA profiling (**n**) and RT-qPCR
165 measurement (**o**) of the indicated miRNAs during MEFs senescence. **p**, Statistical
166 analysis of Mki67-positive cells from **Figure 2f,j**. **q, r**, RT-qPCR analysis of miR-29
167 expression in cells with mimic-mediated expression of miR-29 followed by
168 reintroduction of Suv4-20h. **s**, Statistical analysis of SA- β -gal staining from **Figure 2i**
169 (One-way ANOVA with Dunnett's multiple comparison test). The error bars represent
170 the s.d. obtained from triplicate independent experiments. Two-tailed unpaired
171 Student's t-tests were performed. **p<0.01, ***p<0.001.

172

173

174

175

176

177

178

179

180

181

182

183

184

185

186

187

188

189

190

191

192

193

194

195

196

197

198

199

200

201

202

203

204

205

206

207

208

209

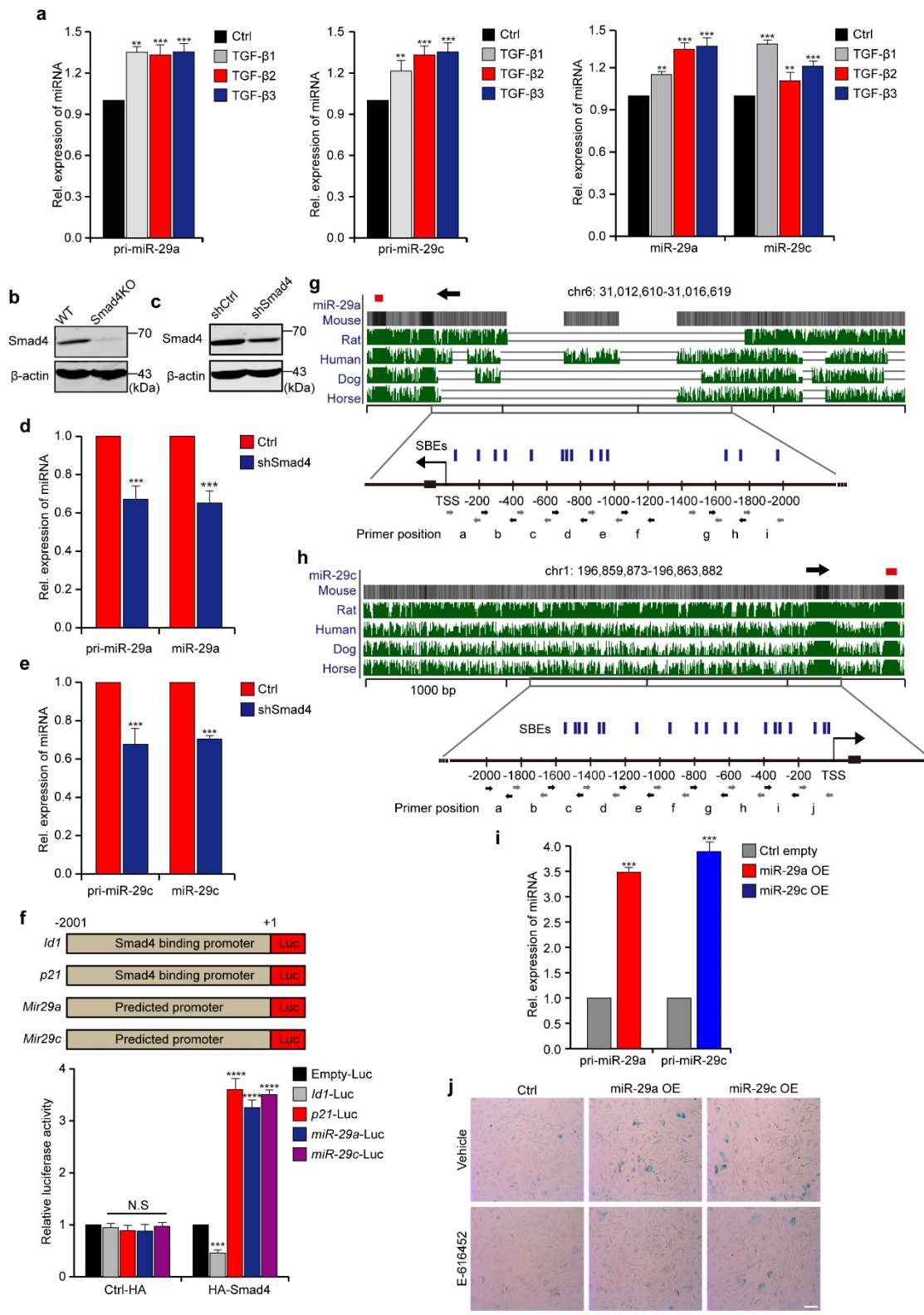
210

211

212

213

214



215 **Supplementary Figure 3. TGF-β signaling regulates transcription of *Mir29* in a**

216 **Smad-dependent manner during MEFs senescence.**

217 **a, RT-qPCR measurement of miR-29 expression in cells treated with or without TGF-**

218 $\beta 1/\beta 2/\beta 3$. **b, c**, Knockout (**b**) or knockdown (**c**) of Smad4 was confirmed by western
219 blotting. **d, e**, RT-qPCR measurement of miR-29a (**d**) and miR-29c (**e**) upon Smad4
220 knockdown. **f**, Luciferase assays showed cells co-transfected with Smad4 and fused
221 constructs containing the predicted promoters. **g, h**, Bioinformatics analysis of *Mir29a*
222 (**g**) and *Mir29c* (**h**) about SBEs residing in the predicted promoters. The arrows show
223 the transcription directions. The letters represent the primer positions for ChIP. **i**, RT-
224 qPCR to measure miR-29 expression following lentivirus-mediated expression of miR-
225 29. **j**, Cells with ectopic expression of miR-29 were subjected to SA- β -staining. The
226 error bars represent the s.d. obtained from triplicate independent experiments. Two-
227 tailed unpaired Student's t-tests were performed. **p<0.01, ***p<0.001.

228

229

230

231

232

233

234

235

236

237

238

239

240

241

242

243

244

245

246

247

248

249

250

251

252

253

254

255

256

257

258

259

260

261

262

263

264

265

266

267

268

269

270

271

272

273

274

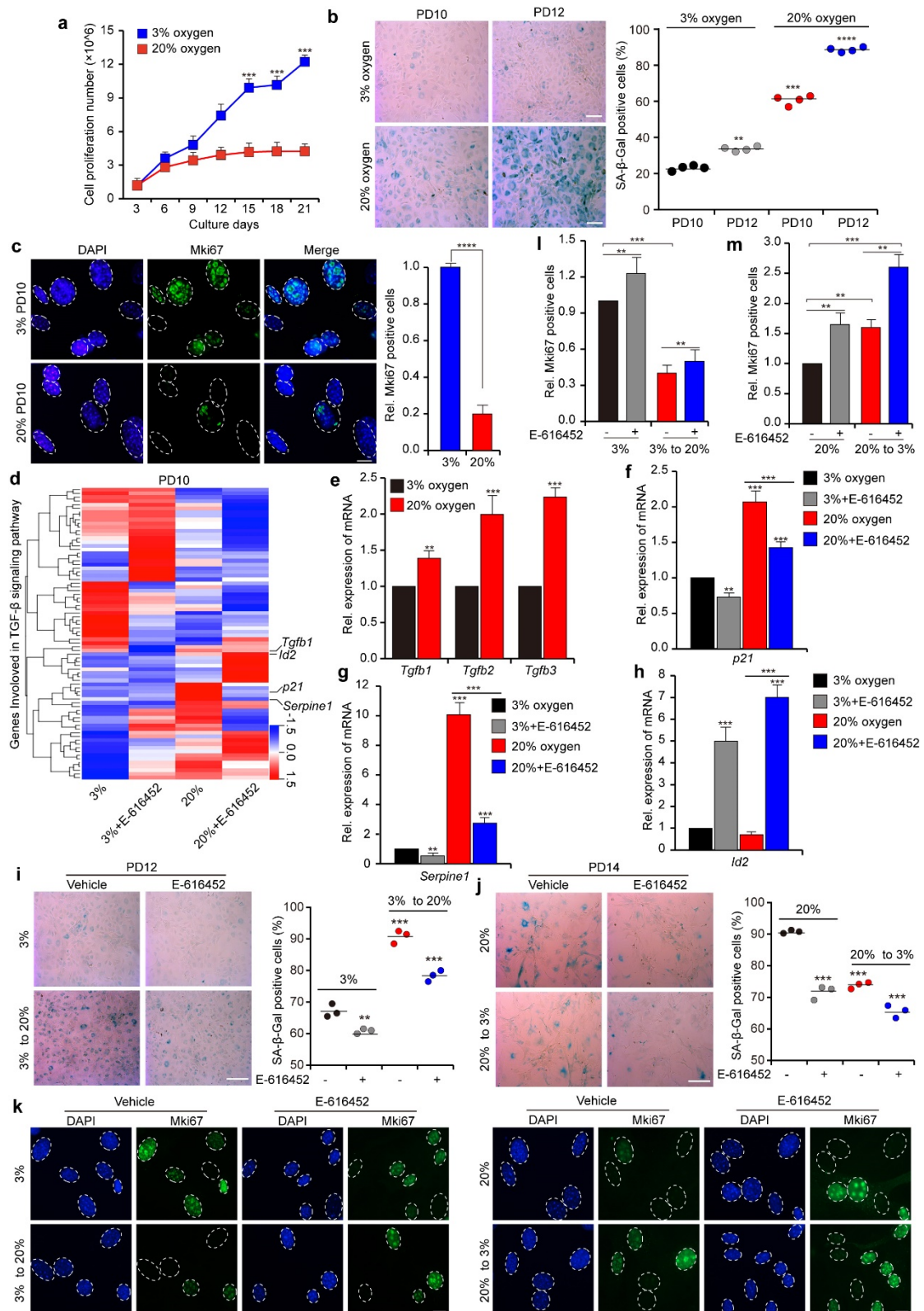
275

276

277

278 **Supplementary Figure 4. Hyperoxia-induced oxidative stress activates TGF- β**

279 **signaling to accelerate senescence.**



280 **a**, The growth curves showed the differences in the proliferation rates of MEFs cultured
281 under physioxia (3% oxygen) and relative hyperoxia (20% oxygen). **b**, Cells from PD10
282 and PD12 incubated in 3% or 20% oxygen were subjected to SA- β -gal staining. Scale
283 bar, 20 μ m. One-way ANOVA with Dunnett's multiple comparison test was performed.
284 **c**, PD10 MEFs grown in 3% oxygen and 20% oxygen were used to immunofluorescent
285 staining analysis of Mki67. Scale bar, 5 μ m. Two-tailed unpaired Student's t-tests were
286 performed. **d**, PD10 MEF cells grown in 3% or 20% oxygen, with or without inhibitors
287 were collected for analysis of differential mRNA transcript expression. **e-h**, RT-qPCR
288 measurement validated the mRNA expression results from **d**. **i, j**, MEFs grown in 3%
289 oxygen and transferred to 20% oxygen (**i**), as well as MEFs cultured in 20% oxygen
290 and transferred to 3% oxygen (**j**), were treated with or without inhibitors and subjected
291 to SA- β -gal staining. Scale bar, 20 μ m. One-way ANOVA with Dunnett's multiple
292 comparison test was performed. **k-m**, Immunofluorescent staining analysis of the
293 Mki67 signal of MEFs with the same treatment as in **j**. Graphs with p-values are
294 representatives of triplicate independent experiments. **p<0.01, ***p<0.001,
295 ****p<0.0001.

296

297

298

299

300

301

302

303

304

305

306

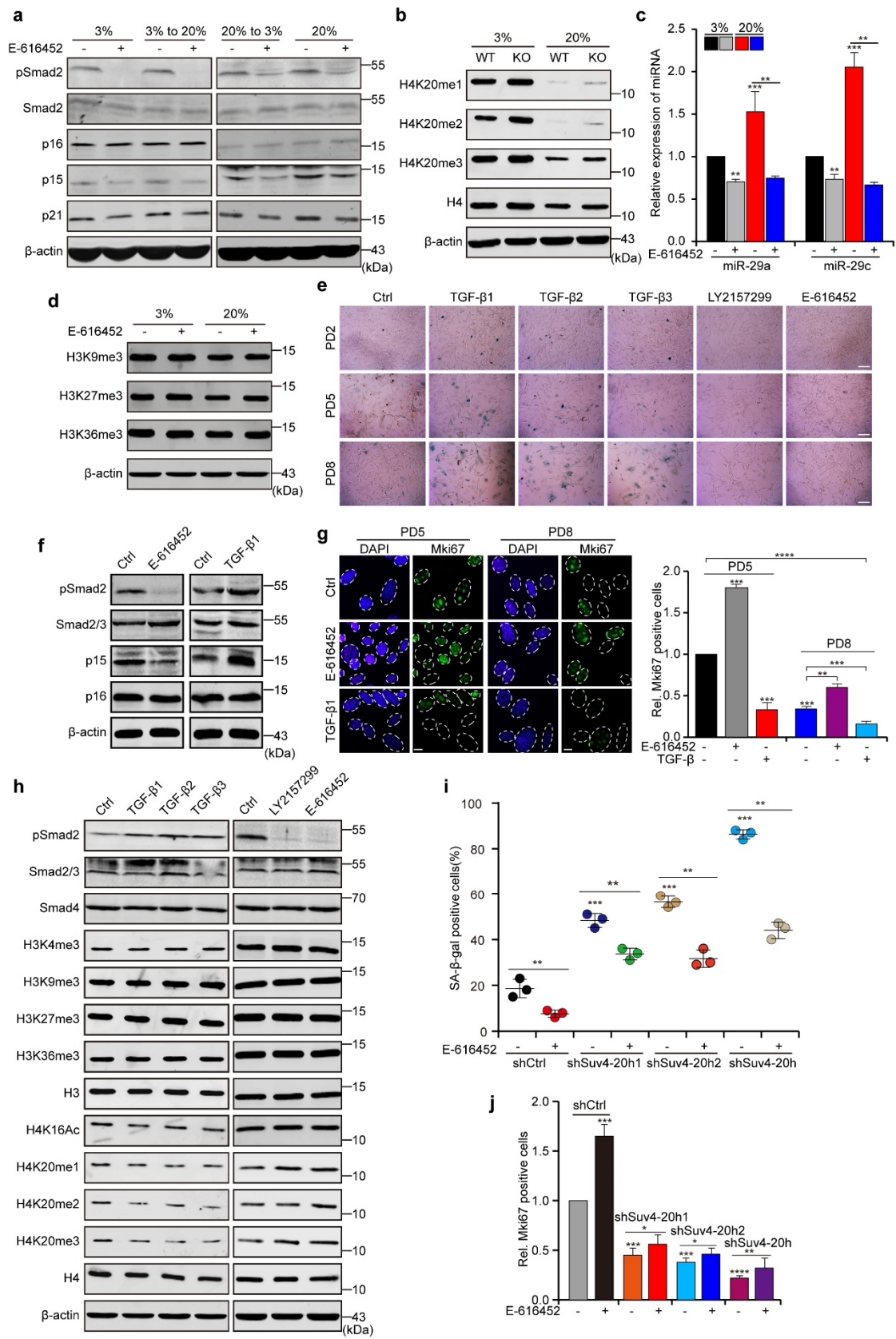
307

308

309

310

311
 312
 313
 314
 315
 316
 317
 318
 319
 320
 321
 322
 323
 324
 325
 326
 327
 328
 329
 330
 331
 332
 333
 334
 335
 336
 337
 338
 339
 340
 341



Supplementary Figure 5. TGF-β signaling regulates senescence by reducing

342 **H4K20me3 abundance.**

343 **a**, Western blots showed protein changes in MEFs grown in 3% or 20% oxygen. β -actin
344 served as a loading control. **b**, Western blots showed changes in H4K20 methylation in
345 MEFs grown in 3% or 20% oxygen upon Smad4 knockout (KO). WT is designated for
346 wild-type. β -actin served as a loading control. **c**, RT-qPCR measurement of miR-29
347 expression in MEFs grown in 3% or 20% oxygen under treatment with or without E-
348 616452. **d**, Western blot for alterations of histone modifications in cells cultured in 3%
349 or 20% oxygen in the presence or absence of E-616452. β -actin served as a loading
350 control. **e**, MEFs from PD2, PD5 and PD8 treated with TGF- β or inhibitors (LY2157299
351 and E-616452) were subjected to SA- β -gal staining. Scale bar, 20 μ m. **f**, Western blot
352 for confirmation of the efficacy of the treatments with E-616452 or TGF- β . **g**,
353 Immunofluorescent microscopy for Mki67 staining in PD5 and PD8 MEFs (left) with
354 the same treatment as in **f**. Signals were analyzed by ImageJ software (right). Student's
355 t-test was applied. Scale bar, 5 μ m. **h**, Western blot for the indicated proteins in cells
356 with the same treatment as in **e**. β -actin served as a loading control. **i**, Statistical analysis
357 of the SA- β -gal staining in **Figure 4g**. One-way ANOVA with Dunnett's multiple
358 comparison test was performed. **j**, Statistical analysis of the Mki67-positive cells in
359 **Figure 4h**. The error bars represent the s.d. obtained from triplicate independent
360 experiments. Two-tailed unpaired Student's t-tests were performed. ** $p < 0.01$,
361 *** $p < 0.001$.

362

363

364

365

366

367

368

369

370

371

372

373

374

375

376

377

378

379

380

381

382

383

384

385

386

387

388

389

390

391

392

393

394

395

396

397

398

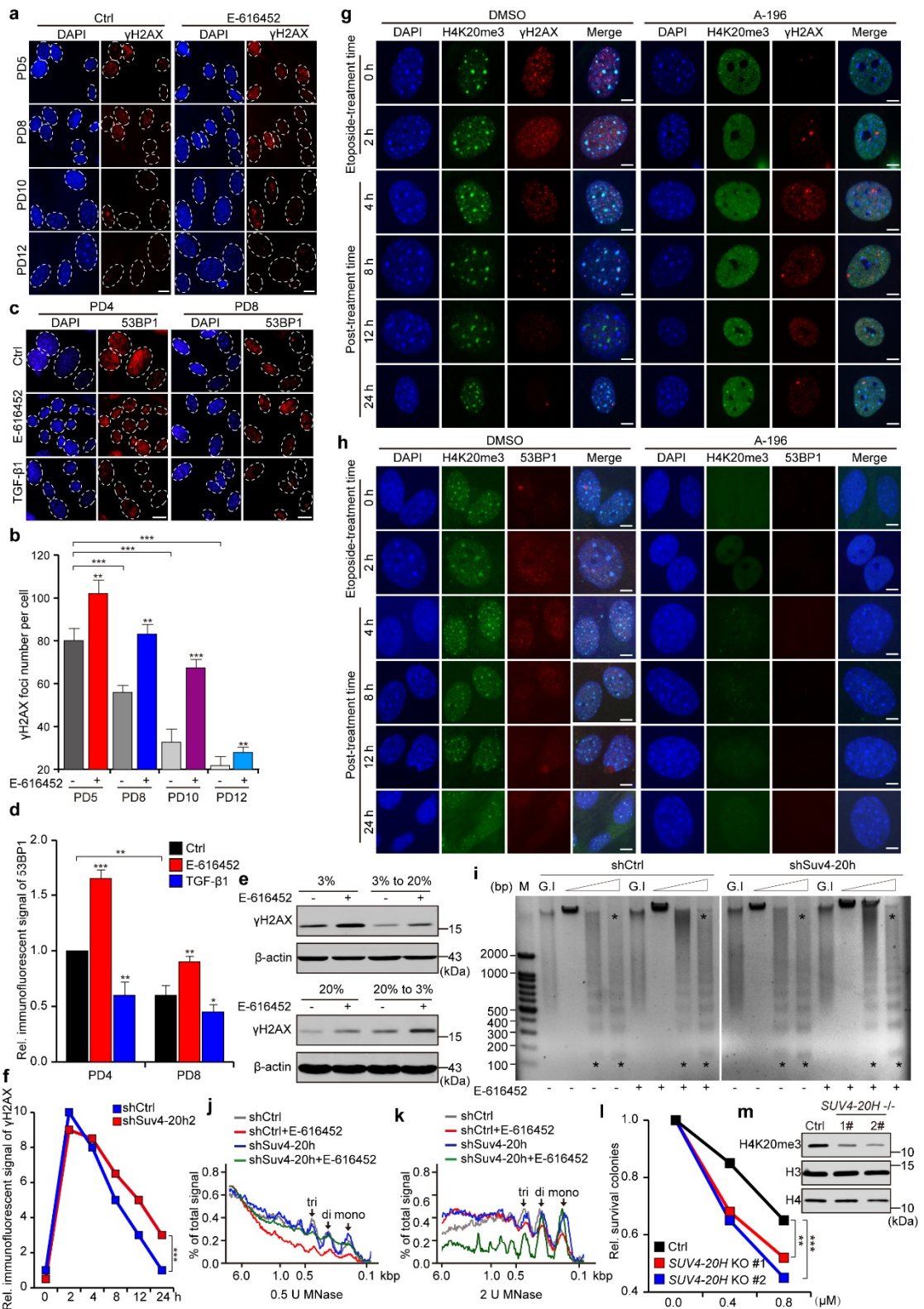
399

400

401

402

403



Supplementary Figure 6. Loss of H4K20me3 leads to DNA damage repair defects and lowers cell survival rates upon DNA damage stress.

404 **a, b**, Immunofluorescent staining of γ H2AX in serially passaged cells treated with or
405 without E-616452 (**a**). Signals were analyzed by using ImageJ software (**b**). Scale bar,
406 5 μ m. **c, d**, Immunofluorescent staining of 53BP1 from PD4 and PD8 cells treated with
407 inhibitor or TGF- β 1 (**c**). Signals were analyzed using ImageJ software (**d**). Scale bar, 5
408 μ m. **e**, Protein levels of γ H2AX in cells treated with or without E-616452, were tested
409 by western blotting. **f**, Statistical analysis of the γ H2AX signals in **Figure 5e**. **g, h**
410 Immunofluorescent staining for γ H2AX (**g**) and 53BP1 (**h**) in cells incubated with A-
411 196 for 48 h followed by etoposide treatment according to the indicated time course.
412 Scale bar, 5 μ m. **i-k**, Nuclei from Suv4-20h knockdown cells treated with or without E-
413 616452 and incubated with 0 U, 0.5 U or 2 U of MNase for 5 min followed by DNA
414 extraction, agarose gel electrophoresis and ethidium bromide staining. The asterisks
415 indicate prominent signal changes (**i**). Signals were analyzed using ImageJ software (**j**,
416 **k**). **l**, Colonies analysis of **Figure 5k** using ImageJ software. **m**, Western blot for
417 H4K20me3 in HEK293T (Ctrl) and *SUV4-20H*-knockout cells (*SUV4-20H*^{-/-} #1 and
418 #2). H3 served as a loading control. The error bars represent the s.d. obtained from
419 triplicate independent experiments. Two-tailed unpaired Student's t-tests were
420 performed. **p<0.01, ***p<0.001.

421

422

423

424

425

426

427

428

429

430

431

432

433

434

435

436

437

438

439

440

441

442

443

444

445

446

447

448

449

450

451

452

453

454

455

456

457

458

459

460

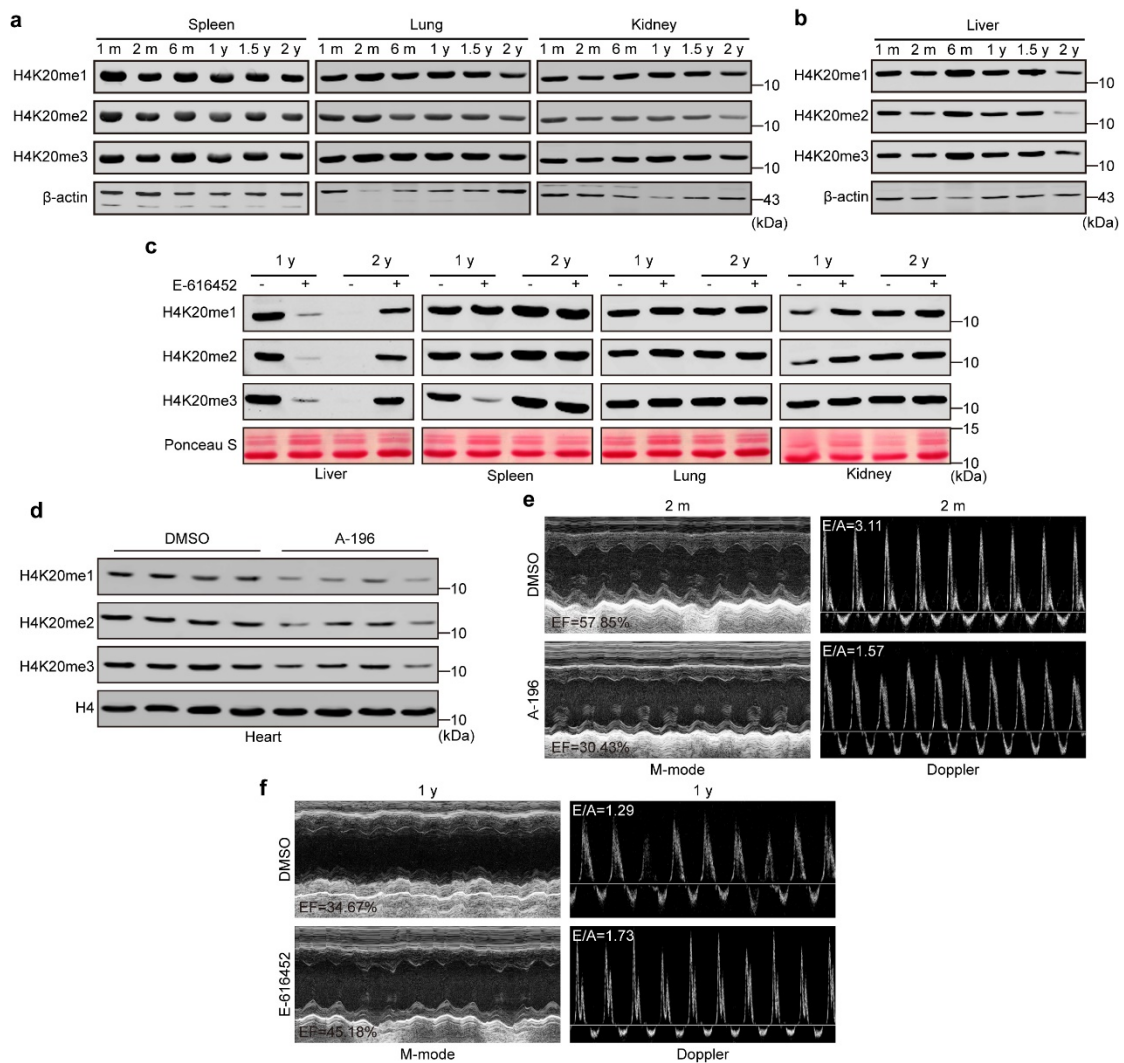
461

462

463

464

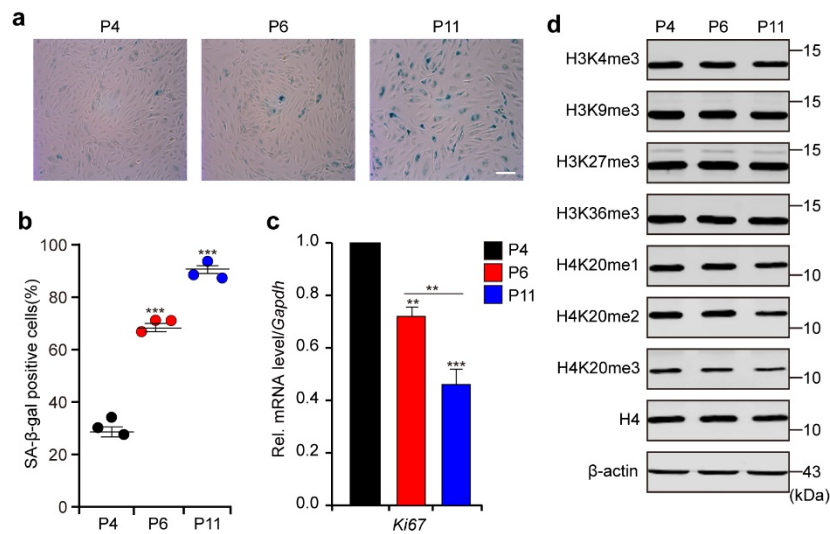
465



Supplementary Figure 7. TGF- β signaling regulates cardiac aging by reducing H4K20me3 abundance.

a, Tissues (spleen, lung and kidney) collected from mice of the indicated age were used to test H4K20 methylation by western blotting. **b**, Western blot for H4K20 methylation in livers collected from **a**. **c**, Immunoblotting for H4K20 methylation in livers, spleens, lungs and kidneys collected from the indicated mice fed with E-616452. H4, β -actin and Ponceau S served as loading controls. **d**, Western blot for H4K20 methylation in hearts from two-month-old mice. Four mice were treated with vehicle (DMSO), whereas four mice were treated with A-196. H4 served as a loading control. **e**, **f**, Representative echocardiographs vehicle (DMSO)- or A-196-treated 2-month-old mice (2m, **e**) and DMSO- or E-616452-treated 1-year-old mice (1 y, **f**). Data were acquired under B-mode, M-mode and pulse-waved Doppler.

466
467
468
469
470
471
472
473
474
475
476
477
478
479
480
481
482
483
484
485
486
487
488
489
490
491
492
493
494
495
496



Supplementary Figure 8. H4K20 methylation is attenuated during HUVECs senescence.

a-c, HUVECs at different passage stages were subjected to SA-β-gal staining (**a, b**) and measurement of *Ki67* expression (**c**). Scale bar, 20 μm. One-way ANOVA with Dunnett's multiple comparison test was performed to analyze the SA-β-gal staining signals. **d**, The total protein levels of the indicated histone modifications were examined by immunoblotting. H4 and β-actin served as loading controls. The error bars represent the s.d. obtained from triplicate independent experiments. Two-tailed unpaired Student's t-tests were performed. **p<0.01, ***p<0.001.

497

498

499

500

501

502

503

504

505

506

507

508

509

510

511

512

513

514

515

516

517

518

519

520

521

522

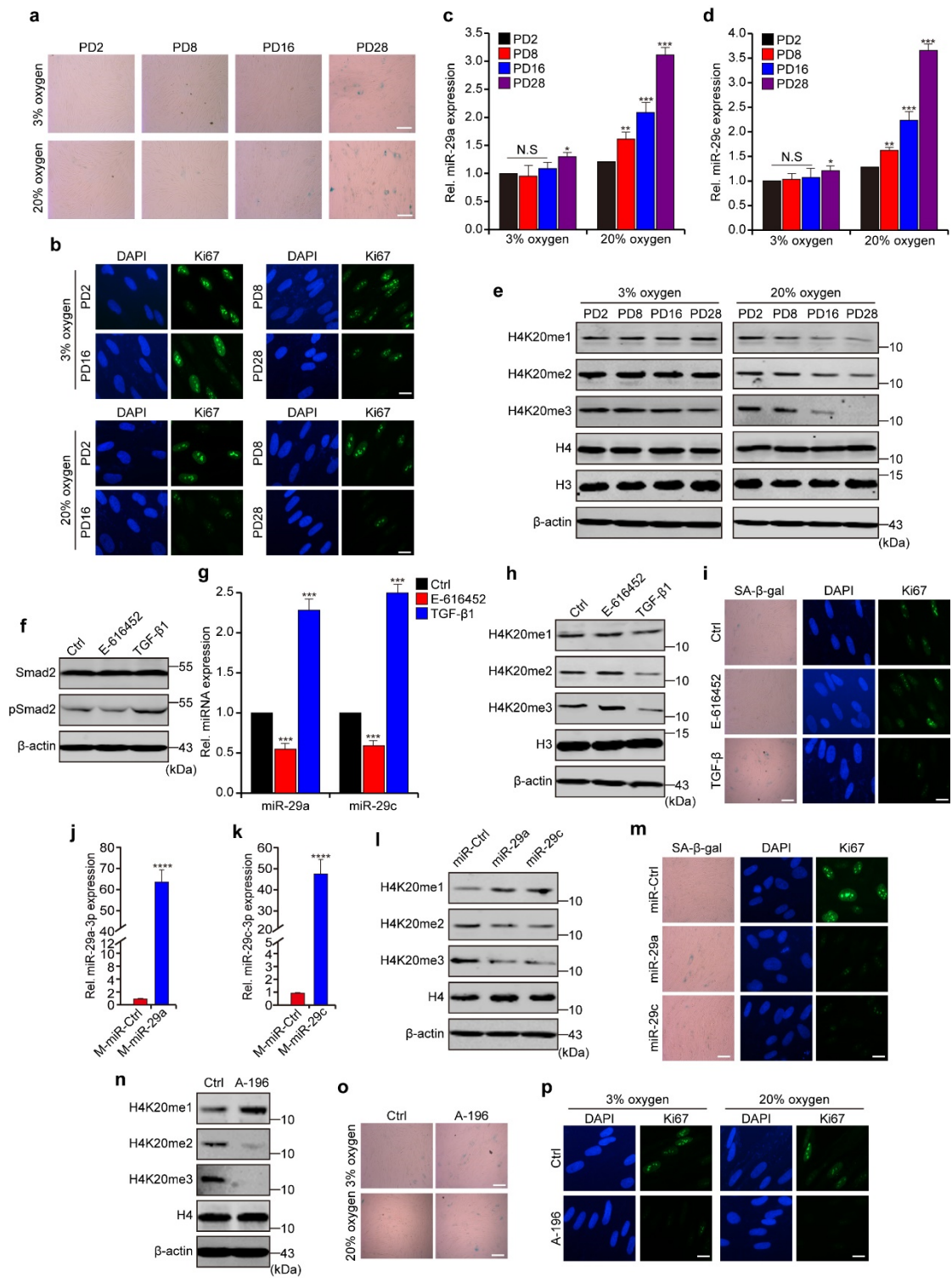
523

524

525

526

527



Supplementary Figure 9. Activated TGF-β signaling promotes HEFs senescence by reducing H4K20me3 via induction of miR-29 expression.

a, SA-β-gal staining of serially passaged HEFs (PD2, PD8, PD16 and PD28) individually grown in 3% and 20% oxygen. Scale bar, 20 μm. **b**, Immunofluorescent

528 staining of Ki67 of serially passaged HEFs cultured in 3% and 20% oxygen. Scale bar,
529 5 μ m. **c, d**, RT-qPCR measurement of miR-29 expression in different passages of HEFs.
530 miRNAs expression was normalized to U6. The error bar represents the s.d. from three
531 independent replicates. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, Student's t-test was
532 performed. **e**, Global H4K20 methylation was tested by western blotting in the HEFs
533 from **a**. H3, H4 and β -actin served as loading controls. **f**, Immunoblotting for the
534 indicated proteins in the presence of E-616452 or TGF- β 1. β -actin served as a loading
535 control. **g**, RT-qPCR measurement of miR-29 expression in different passages of HEFs.
536 **h**, Western blots of the indicated proteins in the HEFs derived from **f**. β -actin served as
537 a loading control. **i**, SA- β -gal staining and immunofluorescent staining of Ki67 in HEFs
538 with the same treatment as those in **f**. **j, k**, RT-qPCR measurement of miR-29 expression
539 in HEFs with ectopic expression of miR-29 mimics. **l**, Western blot for total H4K20
540 methylation in HEFs with ectopic miR-29 expression. β -actin served as a loading
541 control. **m**, SA- β -gal staining (left panel, scale bar, 20 μ m) and immunofluorescent
542 staining of Ki67 (right panel, scale bar, 20 μ m) in HEFs with the same treatment as
543 those in **l**. **n**, Immunoblotting of H4K20 methylation in cells treated with A-196. β -actin
544 served as a loading control. **o, p**, SA- β -gal staining (**o**, scale bar, 20 μ m) and
545 immunofluorescent staining of Ki67 (**p**, scale bar, 5 μ m) in cells with the same
546 treatment as those in **a**.

547

548

549

550

551

552

553

554

555

556

557

558

559

560

561

562

563

564

565

566

567

568

569

570

571

572

573

574

575

576

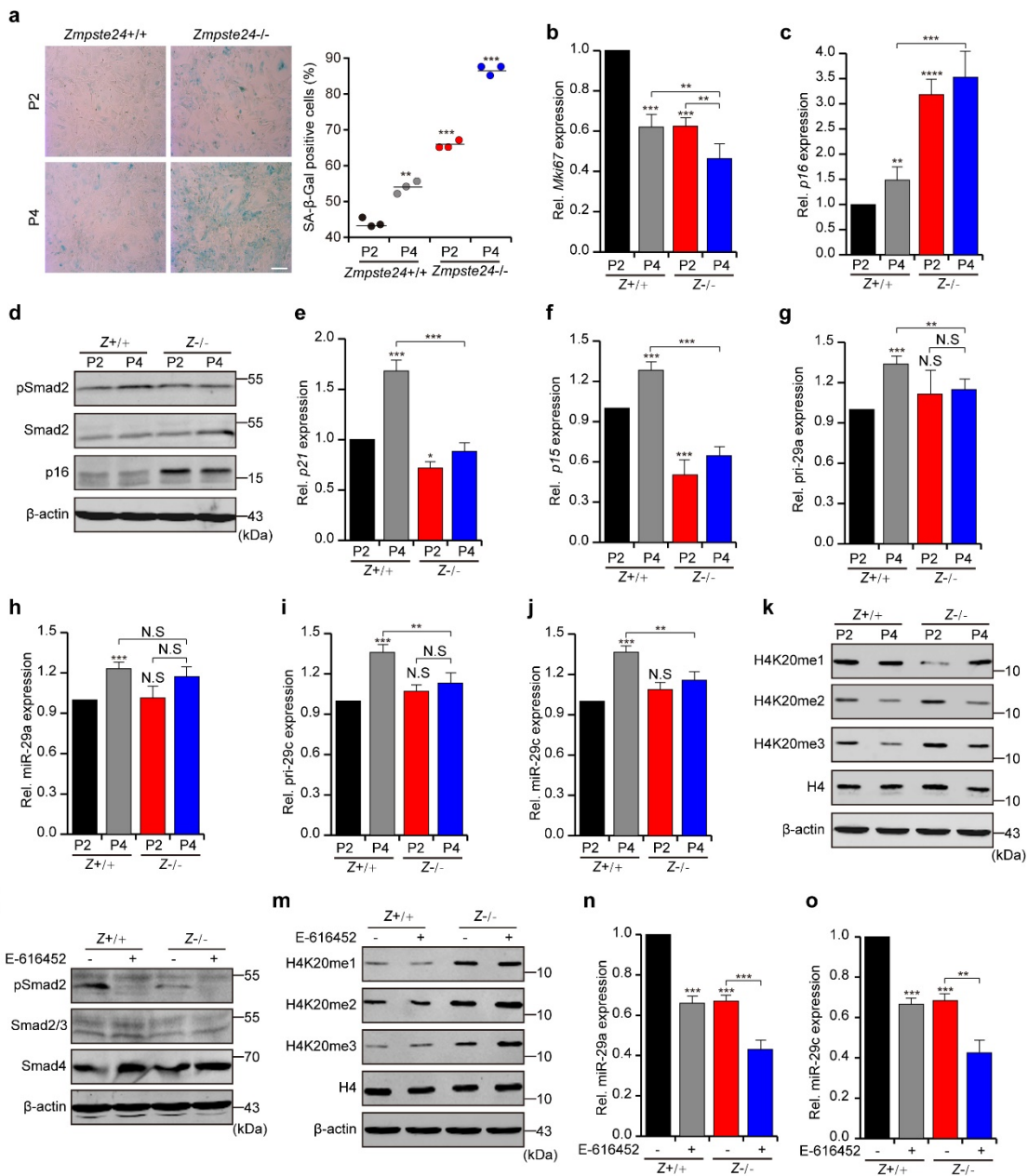
577

578

579

580

581



582 **Supplementary Figure 10. Depletion of *Zmpste24* blocks TGF-β-miR-29 signaling**
 583 **in the HGPS mouse model.**

584 **a**, Serial passages (P2 and P4) of wild-type (*Zmpste24*^{+/+}) and *Zmpste24*-knockout
 585 (*Zmpste24*^{-/-}) MEFs were subjected to SA-β-gal staining (left panel). Signals were
 586 analyzed by ImageJ software. One-way ANOVA with Dunnett's multiple comparison
 587 test was performed (right panel). **b**, **c**, mRNA expression levels of *Mki67* (**b**) and *p16*
 588 (**c**) were tested by RT-qPCR and normalized to that of *Gapdh*. **d**, Total protein levels of
 589 pSmad2, Smad2 and p16 were examined by immunoblotting in serially passaged wild-

590 type and *Zmpste24*-knockout cells. β -actin served as a loading control. **e-j**, RT-qPCR
591 measurement of the indicated genes (*p21*, **e** and *p15*, **f**) and miRNAs (pri-miR-29 and
592 miR-29, **g-j**). Expression of mRNA and pri-miRNA was normalized to that of *Gapdh*
593 and expression of miRNA was normalized to that of U6. Two-tailed unpaired Student's
594 t-tests were performed. The error bars represent the s.d. obtained from triplicate
595 independent experiments. **k**, Western blot for H4K20 methylation in the cells from **d**.
596 β -actin served as a loading control. **l, m**, Immunoblotting of the indicated proteins in
597 late-passage (P6) wild-type and *Zmpste24*-knockout MEFs treated with or without E-
598 616452. β -actin and H4 served as loading controls. **n, o**, RT-qPCR measurement of
599 miR-29a (**n**) and miR-29c (**o**) expression in cells from **l**. $Z^{+/+}$ and $Z^{-/-}$ represent wild-
600 type and *Zmpste24* knockout cells in **b-o**, respectively. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

601

602

603

604

605

606

607

608

609

610

611

612

613

614

615

616

617

618

619

620

621

622

623

624

625

626

627



641 **Supplementary Figure 11.** Uncropped scans of Western blots from Fig. 1a, b, e, h; Fig.

642 2c, d, h; Fig. 4a, d, e, f, i; Fig. 6b, d, j, l

643

644

645

646

647

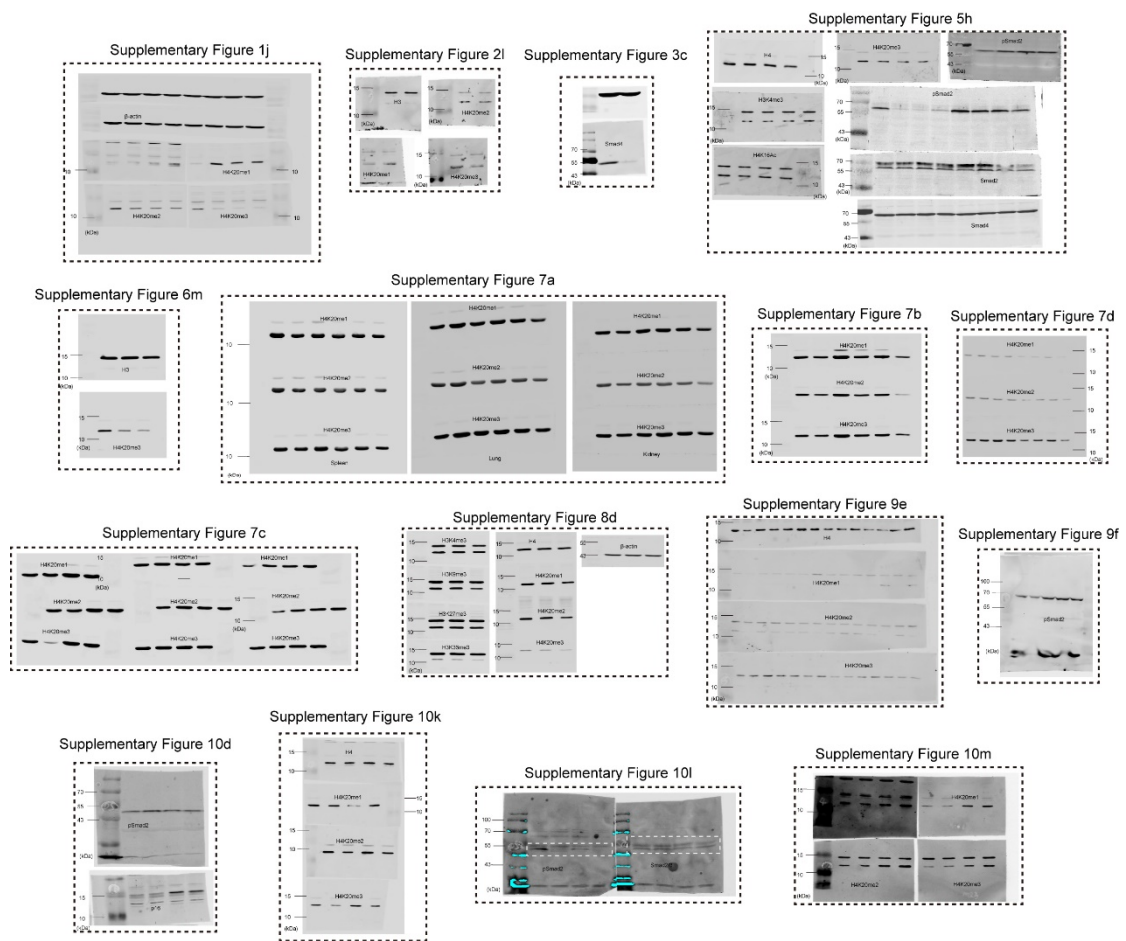
648

649

650

651

652
653
654
655
656
657
658
659
660
661
662
663
664
665
666
667
668
669
670
671
672
673
674
675
676
677
678
679
680
681
682



Supplementary Figure 12. Uncropped scans of Western blots from Supplementary Fig. 1j, Supplementary Fig. 2l; Supplementary Fig. 3b,c; Supplementary Fig. 5h, Supplementary Fig. 6m; Supplementary Fig. 7a, b, c, d; Supplementary Fig. 8d; Supplementary Fig. 9e, f; Supplementary Fig. 10d, k, l, m.

683

684

Supplementary Table 1 Diastolic and systolic data of left ventricle

685

	2 m (DMSO)	2 m (A -196)	1 y (DMSO)	1 y (E-616452)
LVAW; d	0.799 mm±0.054	0.632 mm±0.021	1.172 mm±0.027	1.385 mm±0.038
LVID; d	3.622 mm±0.115	4.128 mm±0.120	4.181 mm±0.160	3.956 mm±0.113
LVPW; d	0.826 mm±0.031	0.852 mm±0.043	0.959 mm±0.061	0.873 mm±0.057
LVAW; s	1.065 mm±0.041	1.092 mm±0.026	1.278 mm±0.016	1.225 mm±0.034
LVID; s	2.876 mm±0.053	4.234 mm±0.106	3.222 mm±0.082	2.476 mm±0.133
LVPW; s	0.692 mm±0.026	0.639 mm±0.017	0.985 mm±0.024	0.905 mm±0.027

686

Diastolic and systolic parameters of left ventricle under B- and M-mode. ± represents
 687 s.d. from mice of each group, respectively. Numbers of mice per group were indicated
 688 in the legend of **Figure 6e,f**. LVAWd, left ventricular anterior wall thickness at end-
 689 diastole; d, diastole; LVIDd, left ventricular internal dimension at end-diastole; LVPWd,
 690 left ventricular posterior wall thickness at end-diastole; LVAWs, left ventricular anterior
 691 wall thickness at end-systole; s, systole; LVIDs, left ventricular internal dimension at
 692 end-systole; LVPWs, left ventricular posterior wall thickness at end-systole.

693

694

695

696

697

698

699

700

701

702

703

Supplementary Table 2 Primers used in our study

Target	Forward (5'-3')	Reverse (5'-3')
qPCR <i>Mki67</i>	ATCATTGACCGCTCCTTTAGGT	GCTCGCCTTGATGGTTCCT
qPCR <i>Suv4-20h1</i>	ACTAGCGCCTTTCCTTCGAG	GCCGAAATCTCACAGGATTGTTG
qPCR <i>Suv4-20h2</i>	GCAGAGCTGCGTGAAGAGG	ACAGGCAGTATTTCCCATCTGA
qPCR <i>p16</i>	CGCAGGTTCTTGGTCACTGT	TGTTACAGAAAGCCAGAGCG
qPCR <i>p21</i>	CCTGGTGATGTCCGACCTG	CCATGAGCGCATCGCAATC
qPCR <i>p15</i>	CCCTGCCACCCCTTACCAGA	CAGATACCTCGCAATGTCACG
qPCR <i>Tgfb1</i>	GGCGGTGCTCGCTTTGT	GCGGGTGACCTTTTAGCATAG
qPCR <i>Tgfb2</i>	CGGAGCGACGAGGAGTA	CGGACGATTCTGAAGTAGGGT
qPCR <i>Tgfb3</i>	GGACTGGCGGAGCACAA	CGCTGCTTGGCTATGTGC
qPCR <i>Serpine1</i>	CTTCAGCCCTTGCTTGCC	GGACCACCTGTGAAACACT
qPCR <i>Ryr</i>	GCAGGTGGATGTGGAA	GTAGGAATGGCGTAGCA
qPCR <i>Myh7</i>	GAATGGCAAGACGGTGAC	TCCAGGAAGCGTAGCG
qPCR <i>Ki67</i>	GCCTGCTCGACCCTACAGA	GCTTGTCAACTGCGGTTGC
qPCR <i>Gapdh</i>	GTGTTCTACCCCAATGTGT	ATTGTCATACCAGGAAATGAGCTT
qPCR <i>GAPDH</i>	AGCCACATCGCTCAGACAC	GCCCAATACGACCAATCC
qPCR Pri-miR-29a	AGCCCTGAAGTAAGTGTC	AGGTCTTCATCCGAGCAT
qPCR Pri-miR-29c	ATGGGTGGAAGAGGGTT	TCTCCAGGTTACAGACGAG
qPCR mmu-miR-29a-3p	GGCTAGCACCATCTGAAATCGGTTA	
qPCR mmu-miR-29c-3p	GGCTAGCACCATTGAAATCGGTTA	
qPCR mmu-miR-29b-3p	GCGTAGCACCATTGAAATCAGTGTT	
qPCR U6	CGCAAGGATGACACGCAAATTC	
ChIP miR-29a -414- -208	AACTATTGCACGGACTTCACCTTC	CTGGACACTTACTTCAGGGCTGTAC
ChIP miR-29a -621- -415	AGTAAAAAGTGTCACGCTTATCAAAA	CCTAATTTACAGCGATCCTATG
ChIP miR-29a -828- -622	GCAGCAGTACTGATGATAGTGATAA	TCCAGATCGATCTGTTAGAGTCG
ChIP miR-29a -1035- -829)	TTATTGGAGTCCCTGACACATTC	TACTGCTACTACTATTACAGTTTTAAA
ChIP miR-29a -1236- -1021	GATTTGAGAATGAGCAGGAA	CAGGGACTCCAATAACTACAC
ChIP miR-29a -1656- -1450	GTCCCATGCACACTGGCTACTTC	GTTCTTACTGAATIGTTTGAAGCA
ChIP miR-29a -1780- -1559	AATAGATTCCCAGTCTGTAGC	CTACCCGAGGAAACAAAC
ChIP miR-29a -2001- -1752	CCGAGCTCATTCTCCAGCCC	GGTGCCAGGCTACAGACTGGG
ChIP miR-29c -207- -1	GGGTTCCCTTGGGCTGCAC	GGACCGACTGGTGGTGTTCCTTC

ChIP miR-29c -414- -208	CACAGCAGAGGGTAGACTACAGAGG	TCTTCCACCCATGGAATGCTG
ChIP miR-29c -621- -415	GCTCAAAGTGTGGCTGTATG	CTGCAATTCTTACTCCTAAAACAC
ChIP miR-29c -828- -622	GTTGTTAACATCTCATAGGTCATTG	AGACAAAAACAATGCAAAGCTT
ChIP miR-29c -1035- -829	AGGTACATTTTAAAACATACCTTGTT	TGAACCCAAGTAAGTCACCTGTG
ChIP miR-29c -1242- -1036	TCACATTATATTGCTCTATTACCC	AAAAATAAGATTACAAGCTATGTT
ChIP miR-29c -1449- -1243	CCCCTCCCCAGTCTAGCTCT	GTATCCCCAGAGTCACAATATTCAA
ChIP miR-29c -1656- -1450	GAGAAGTCAGTGCAGCATTGCTAG	GATTTAGCTCAGACAATCTGGATGG
ChIP miR-29c -1863- -1657	ACTGCCATCTAATCTATTTATGACC	AGAAACTGGGCCAGCCA
ChIP miR-29c -2001- -1864	CATAGAGGGAGGGGATTGG	TATTAACAAGGGTCAAAATCCTG
Promoter miR-29a-3p	GGGGTACCCCGAGCTCATTCTCCAGCC	CGCTCGAGCACAAGAGGTCATGTGCA
Promoter miR-29c-3p	GGGGTACCCATAGAGGGAGGGGATTG	CGCTCGAGGGACCGACTGGTGGTGT
3' UTR- <i>Suv420h1</i>	GAGCTCTTGAATCTTGTGCGTGAC	GTCGACCTCTTACATTGAATCCC
3' UTR- <i>Suv420h1</i> -Mut	CAAGAATGATCGCTACCTATTTTTTC	TAGGTAGCGATCATTCTTGCAAAG
3' UTR- <i>Suv4-20h2</i>	CGAGCTCCTTACAGGGAGCGGAATG	GCGTCGACCCAGTAGCACCAGGGATA
3' UTR- <i>Suv4-20h2</i> -Mut	GAGTCCTTACAGGGAGCGGAATGGA	
3' UTR- <i>Suv4-20h2</i> -Mut-1		GTCGACCCAGTTGGAGCTGGGATAGC
3' UTR- <i>Suv4-20h2</i> -Mut-2		GTCGACCCAGTAGCACCAGGGATTGG