

Supplementary Table 1. Primer Sequences

Gene	Forward	Reverse
<i>βActin</i>	ACCGTGAAAAGATGACCCAG	GTACGACCAGAGGCATACAG
<i>Cdkn1a</i>	GCAGATCCACAGCGATATCC	CAACTGCTCACTGTCCACGG
<i>Cdkn2a</i>	CCCAACGCCCCGA ACT	GCAGAAGAGCTGCTACGTGAA
<i>Gstp1</i>	TGCCATCTTGAGACACCTTG	GTTGGTGTAGATGAGGGTGAC
<i>Gdf15</i>	TCCCCACCTGTATCGCTC	TCTGTCCTGTGCATAAGAACC
<i>Dusp3</i>	GCTAGTTATCGCCTACCTCATG	CTCCTTGGCTAGTCTGTCATTG
<i>Hsp90aa1</i>	ATCCCCGTGCTGTATTGTC	TCTCCAGGTGTTTCTTTGCTG
<i>Ing1</i>	AAGCACACCAGGACATCAG	GATTCTCATTGTTTCGCTGCC
<i>Kpnb1</i>	ATCACATTGCTGGAGATGAGG	TCCGCCCTTCAGTTAATAGTTC

Supplementary Table 2. Antibodies used for immunostaining

Antibody	Source	Catalog Number	Concentration
Anti-Insulin	Abcam	Ab195956	1:200
Anti-HMGB1	BioVision	A1005-50	1:400
Anti-Ki67	Abcam	Ab15580	1:500
488 Donkey anti-Guinea Pig IgG	Jackson ImmunoResearch Laboratories	706-545-148	1:200
594 Donkey anti-Rabbit IgG IgG	Jackson ImmunoResearch Laboratories	711-585-152	1:200

Supplementary Table 3. Viability of β-Cells by Experiment

Experiment #	Proportion PI-negative (%)	
	βgal-	βgal+
1	86	86
2	80	74

3	80	86
4	82	77
5	87	78
6	89	81

Supplementary Table 4. Viability of MIN6 Cells by Experiment

Passage #	Proportion PI-negative (%)	
	Control	Bleomycin
33	63	64
34	45	40
34	42	40
39	42	51
40	33	40
40	41	38

Supplementary Table 5. Proteomic Results for Core SASP Factors Described in the Literature

Protein	Bgal p-value	MIN6 p-value	Bgal Log2FC	MIN6 Log2FC
IL1A	0.079	0.362	0.054	0.042
IL6	0.267	0.072	-0.261	0.278
CCL2	0.169	0.004	0.051	0.111
CCL3	0.882	0.091	0.004	-0.042
CCL5	0.353	0.000	-0.051	-0.204
CXCL10	0.849	0.008	-0.021	0.191
CXCL1	0.040	0.090	0.196	0.096
CTSB	0.395	0.581	-0.093	-0.078
CXCL3 CXCL2	0.011	0.045	0.171	0.209
FGF2	0.120	0.001	-0.125	-0.107

IGFBP3	0.336	0.143	-0.092	-0.061
IGFBP5	0.573	0.000	0.097	-0.877
LAMA1 LAMB1 LAMC1	0.037	0.005	0.188	0.306
NGF	0.402	0.020	-0.047	-0.128
PLAU	0.613	0.009	-0.114	0.111
SERPINE1	0.278	0.067	-0.159	0.186
VEGFA	0.189	0.000	0.562	-0.957

Supplementary Table 6. Proteomic results for 3 SASP factors characteristic of other tissues as described in the literature

Cell type	Species	SASP Factor	Condition		Change direction
			β Gal ⁻	β Gal ⁺	
β -cells	Mouse	IL1A	100%	104%	Increase
		IL1B	100%	123%	Increase
		IL6	100%	83%	Decrease
	Human	IL1A	100%	105%	Increase
		IL1B	100%	102%	Increase
		IL6	100%	94%	Decrease
			Control	Bleomycin	
MIN6	Mouse cell line	IL1A	100%	103%	Increase
		IL1B	100%	115%	Increase
		IL6	100%	121%	Increase

Supplementary Table 7. Relative expression levels of different islet hormones used to identify different islet subpopulations in scRNASeq analysis.

	alpha C	alpha S	alpha SR	beta C	beta S	beta SR	delta C	delta S	delta SR	pp C	pp S	pp SR
Ins2	7.2	3.73	6.47	11.5	11.7	11.5	7.27	3.16	6.48	7.87	4.01	7.15
Ins1	5.57	2.16	4.9	9.98	10.2	9.95	5.73	1.9	4.97	6.08	2.48	5.55
Gcg	11.2	11.1	11.2	1.27	1.08	1.27	1.39	1.09	1.02	2.78	1.99	2.47
Sst	0.554	0.257	0.402	0.416	0.284	0.362	9.47	9.56	9.45	2.19	1.73	2.28
Ppy	2.32	1.96	2.52	1.19	0.853	0.834	3.21	2.45	2.71	10.2	10.1	9.96

C= control, S =S961, SR= S961 then 2 weeks recovery

Supplementary Figure 1. Bleomycin-induced DNA damage as an *in vitro* model of β -cell senescence.

(A) Workflow for inducing senescence in MIN6 cells. MIN6 cells were cultured and treated with 50 μ M bleomycin for 48 hours, followed by 3-4 days of culture in regular MIN6 media, to induce senescence.

(B) Bleomycin treatment induced a two-fold increase in *Cdkn1a* expression in MIN6 cells compared to the control cells. Results show mean \pm SEM; * $p = 0.03$; $n = 6$ (Paired t-test).

(C) Bleomycin treatment significantly increased the percentage of MIN6 cells that exhibited SA- β gal activity. MIN6 samples with higher passage number exhibited higher proportion of β gal⁺ cells. * $p = 0.015$ (Paired t-test).

(D) Nuclear HMGB1 staining decreased in cells treated with bleomycin. Cumulative frequency graph showed a decrease in the percentage of cells that had nuclear staining and an overall decrease in staining intensity.

(E) Micrograph showing a local decrease of Ki67 staining as proliferation marker in MIN6 cells treated with bleomycin.

(F) Micrograph of DAPI stained nuclei showing a significant increase of nucleus areas after treatment with bleomycin. This is a well-known morphological change of senescence.

(G) qPCR results for *Cdkn1a* and *Cdkn2a* mRNA expression in untreated MIN6 cells after days in culture supporting the capacity of immortalized cell lines to enter senescence; $n=5$, * $p<0.05$

(H) Normalized values of SA- β Gal activity of different passages of MIN6 cells shows a significant increase of paired β Gal⁺ cells with higher passages. This supports the capacity of subpopulations MIN6 cells produce SASP.

Supplementary Figure 2. Bleomycin treated MIN6 cells timecourse expression of individual genes qPCR data shown in **Fig. 2B**. Red bars are statistically significantly different ($p<0.05$) than time 0; $n=5$.

Supplementary Figure 3. Transcriptional expression of the complete SASP signature in β -cells from animals with insulin resistance. Heatmap showing average expression levels by scRNA-seq. Expression levels are shown as the log of the fold change from overall average, and the midpoint of 0 represents average expression across all samples.

Supplementary Figure 4. Heatmap showing expression of all β -cell SASP targets in each cluster. Expression levels are shown as z-scores, and the midpoint of 0 represents average expression across all samples. Cluster 4 did not appear in the control group, so all control cluster 4 squares are marked in black.