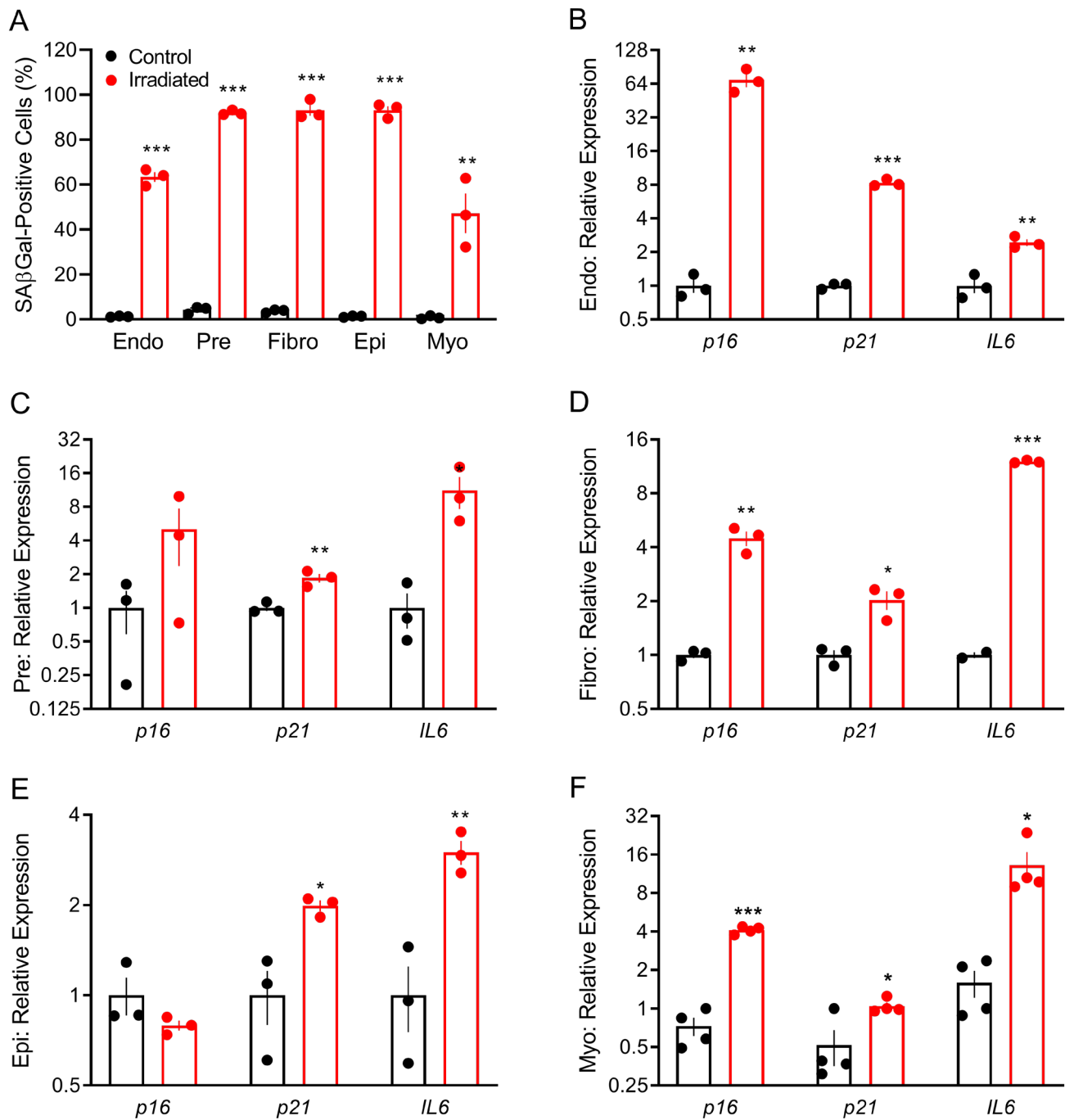


1 SUPPLEMENTARY FIGURE 1

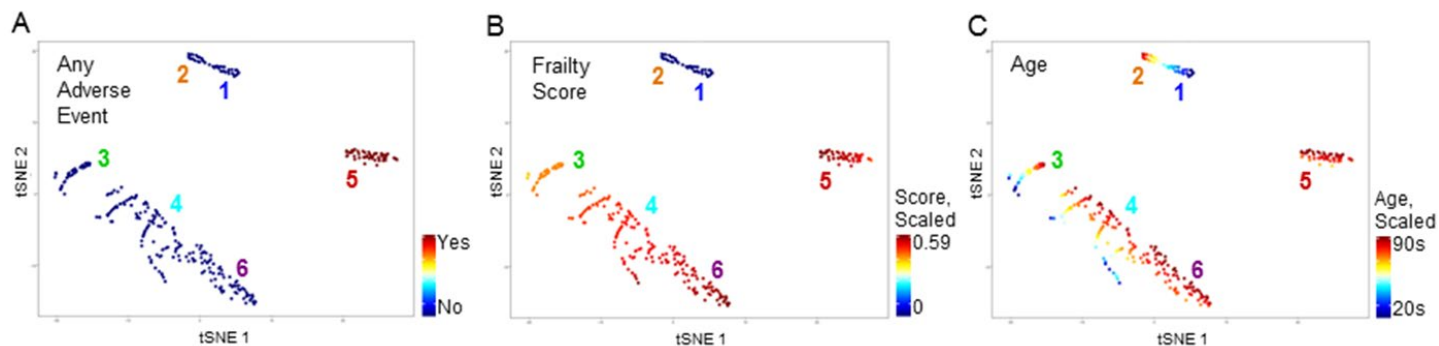


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3 Supplementary Figure 1. Markers of senescence in irradiated cells.

4 (A) Percentage of endothelial cells (endo), preadipocytes (pre), fibroblasts (fibro), epithelial cells (epi), and
5 myoblasts (myo) staining positively for SA-β-Gal. Expression of cyclin dependent kinase inhibitors and a SASP
6 factor in (B) endo, (C) pre, (D) fibro, (E) epi, and (F) myo cells in culture. (Mean ± SEM; individual two-tailed t-
7 tests with significance indicated as p < 0.05*, 0.01**, and 0.001***; n = 3 replicates per cell type).

8 **SUPPLEMENTARY FIGURE 2**



Supplementary Figure 2. Phenotypic cluster definition.

All study participants in which accumulation of deficit frailty status was determined and all proteins were measured were clustered based on the presence of (A) any post-surgical adverse event, (B) frailty score, and (C) chronological age (n = 343).

15 **Supplementary Table 3. Characteristics of participants used to study associations between circulating**
 16 **SASP and chronological age.**

	20-29	30-39	40-49	50-59	60-69	70-79	80-89	
Characteristic	Number (%) or Median (Q1,Q3)							p-value
n	38	37	38	39	37	38	40	
Female	20 (52.6%)	19 (51.4%)	20 (52.6%)	20 (51.3%)	20 (54.1%)	18 (47.4%)	20 (50%)	0.810 ¹
Male	18 (47.4%)	18 (48.6%)	18 (47.4%)	19 (48.7%)	17 (45.9%)	20 (52.6%)	20 (50%)	
Age in years	24.2 (23,26.1)	33.2 (32.4,36.4)	44.5 (43.2,47.6)	54.6 (52.8,56.9)	65.5 (63,67.5)	74.4 (71.9,76.9)	83.2 (81.7,86.3)	N/A
BMI	24 (21.7,27.4)	26.7 (24.3,28.8)	25.8 (23.6,27.8)	27.2 (23.4,30.3)	28.1 (24.8,31.2)	28.6 (24.8,31)	27.1 (24.7,29.5)	0.004 ²
Frailty score	0 (0,0.03) n-miss = 9	0 (0,0.03) n-miss = 10	0.03 (0.03,0.08) n-miss = 8	0.06 (0,0.1) n-miss = 4	0.07 (0.03,0.13) n-miss = 4	0.1 (0.06,0.17) n-miss = 6	0.19 (0.1,0.27) n-miss = 10	<0.001 ²

17 ¹Kruskal-Wallis, ²Spearman Correlation

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Supplementary Table 4: Circulating SASP factors are associated with chronological age.

Protein	Alias	Model 1		Model 2	
		r-value	q-value	r-value	q-value
ACTIVIN A	INHBA	0.671	<0.001	0.105	0.022
ADAMTS13	VWF	-0.163	0.011	-0.184	<0.001
CCL3	MIP1A, SCYA3	0.415	<0.001	0.393	<0.001
CCL4	MIP1B, SCYA4	0.526	<0.001	0.446	<0.001
CCL5	RANTES, SCYA5	-0.138	0.031	-0.153	0.001
CCL17	TARC, SCYA17	0.237	<0.001	0.235	<0.001
CCL22	MDC, SCYA22	0.187	0.003	0.155	0.001
FAS	APT1, TNFRSF6	0.482	<0.001	0.376	<0.001
GDF15	MIC1, NAG1, NRG1	0.746	<0.001	0.320	<0.001
GDNF		-0.054	0.425	-0.040	0.416
ICAM1	CD54	0.192	0.003	0.082	0.077
IL6		0.330	<0.001	0.015	0.759
IL7		-0.130	0.041	-0.160	<0.001
IL8	CXCL8	0.198	0.002	0.106	0.021
IL15		0.267	<0.001	0.127	0.005
MMP2	CLG4A	0.119	0.061	0.098	0.031
MMP9	CLG4B	0.037	0.574	0.013	0.759
OPN	SPP1, PSEC0156	0.260	<0.001	0.228	<0.001
PAI1	SERPINE1, PLANH1	-0.033	0.587	-0.027	0.575
PAI2	SERPINEB2, PLANH2	-0.042	0.534	-0.032	0.511
SOST		0.303	<0.001	0.280	<0.001
TNFα		0.349	<0.001	0.317	<0.001
TNFR1		0.632	<0.001	0.440	<0.001
VEGFA	VP	0.172	0.007	0.185	<0.001
Model 1: FDR-corrected spearman correlation of chronological age versus SASP protein.					
Model 2: FDR-corrected spearman correlation of chronological age versus SASP protein adjusted for sex and BMI.					

23 **Supplementary Table 5. Characteristics of participants used to study associations between circulating**
 24 **SASP and biological age.**

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	Aortic Stenosis	Ovarian Cancer	Biobank, 60+	
Characteristic	Number (%) or Median (Q1,Q3)			p-value
n	97	36	115	
Female	42 (43%)	36 (100%)	58 (50%)	<0.001 ¹
Male	55 (57%)	0 (0%)	57 (50%)	
Age in years	82.0 (76.0, 87.0)	71.7 (64.8, 77.1)	75.0 (68.2, 81.8)	<0.001 ²
BMI	29.1 (26.5, 33.2)	26.8 (22.6, 32.6)	27.8 (24.7, 31.0)	0.008 ²
Frailty score	0.23 (0.17, 0.29) n-miss = 0	0.14 (0.05, 0.27) n-miss = 0	0.10 (0.06, 0.18) n-miss = 20	<0.001 ²

26 ¹Pearson's Chi-Square, ²Kruskal-Wallis

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29 **Supplementary Table 6: Characteristics of biologically older versus younger ovarian cancer**
 30 **participants.**

	Non-frail	Frail	
	Mean (±SD) or Number (%) Median (Q1,Q3)		p-value
n	18	18	
Age in years	71.1 (7.5) 71.6 (65.4, 77.0)	71.2 (7.4) 71.7 (65.5, 77.0)	0.975 ¹
Stage: IIIc IV	15 (83.3%) 3 (16.7%)	15 (83.3%) 3 (16.7%)	1.000 ²
BMI	24.8 (4.4) 23.0 (21.9, 26.9)	30.6 (5.4) 31.6 (25.6, 34.2)	0.001 ¹
Frailty score	0.04 (0.03) 0.05 (0.01, 0.07)	0.27 (0.06) 0.27 (0.21, 0.31)	< 0.001 ¹

¹Kruskal-Wallis, ²Chi-Square

33 **Supplementary Table 7. Primers and probes for real-time PCR.**

Gene	Primers and probes catalog number or sequence	Source and/or reference
<i>TBP</i>	Hs.PT.58.20792004	IDT
<i>P16</i>	Forward primer: 5' CCAACGCACCGAATAGTTACG 3' Reverse primer: 5' GCGCTGCCCATCATCATG 3' Probe: 5' FAM - CCTGGATCGGCCTCCGAC - ZEN / IBFQ 3'	IDT(44)
<i>P21</i>	Hs.PT.58.40874346.g	IDT
<i>IL6</i>	Hs.PT.58.40226675	IDT

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Protein	Alias	Number Imputed Values	Percentage Imputed of All Samples per Target
ACTIVIN A	INHBA	0	0%
ADAMTS13	VWFCP	0	0%
CCL3	MIP1A, SCYA3	40	10%
CCL4	MIP1B, SCYA4	8	2%
CCL5	RANTES, SCYA5	0	0%
CCL17	TARC, SCYA17	1	0%
CCL22	MDC, SCYA22	0	0%
FAS	APT1, TNFRSF6	0	0%
GDF15	MIC1, NAG1, NRG1	5	1%
GDNF		52	13%
ICAM1	CD54	0	0%
IL6		0	0%
IL7		0	0%
IL8	CXCL8	0	0%
IL15		0	0%
MMP2	CLG4A	0	0%
MMP9	CLG4B	0	0%
OPN	SPP1, PSEC0156	16	4%
PAI1	SERPINE1, PLANH1	0	0%
PAI2	SERPINEB2, PLANH2	0	0%
SOST		0	0%
TNFα		1	0%
TNFR1		0	0%
VEGFA	VPH	1	0%