Small molecule compounds that induce cellular senescence

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Summary

To date, dozens of stress-induced cellular senescence phenotypes have been reported. These cellular senescence states may differ substantially from each other, as well as from replicative senescence through the presence of specific senescence features. Here, we attempted to catalog virtually all of the cellular senescence-like states that can be induced by low molecular weight compounds. We summarized biological markers, molecular pathways involved in senescence establishment, and specific traits of cellular senescence states induced by more than fifty small molecule compounds.

Key words: cellular senescence; cell stress; DNA damage; DNA replication stress; epigenetic modifiers; aging.

Cellular senescence is a stable arrest of the cell cycle and is characterized by complex phenotypic changes. It was first described in studies of human fibroblasts that ceased proliferation following an extended cultivation (Hayflick & Moorhead, 1961; Hayflick, 1965). Discovered by Hayflick and Moorhead, senescence in normal human cells was shown to depend on telomere dysfunction originating mainly from replication-associated telomere shortening (Harley et al., 1990; Allsopp, 1996; Bodnar et al., 1998). This type of senescence is also known as replicative senescence and is the prototypical cellular senescence state. Other forms of senescence (i.e., not linked to proliferation-dependent telomere shortening) include a variety of prematurely developed cellular senescence phenotypes, similar but not identical to replicative senescence. Many proliferative cell types can undergo so-called stress-induced premature senescence (SIPS) upon exposure to subcytotoxic stresses (UV, \( \gamma \)-irradiation, \( H_2O_2 \), hyperoxia, etc.) (Toussaint et al., 2000, 2002). Oncogene-induced senescence (OIS) represents another complex senescence phenotype that depends on activation and/or overexpression of oncogenes (Serrano et al., 1997; Bianchi-Simiraglia & Nikiforov, 2012). The mechanism of OIS involves DNA damage that may be a result of DNA hyper-replication (Di Micco et al., 2006), replication fork reversal (Neelsen et al., 2013), depletion of nucleotide pools (Mannava et al., 2013), and/or increased levels of reactive oxygen species (ROS) (Lee et al., 1999). Conceptually and mechanistically, OIS is closely related to tumor-suppressor loss-induced senescence (Chen et al., 2005; Di Miti & Alimonti, 2016). Cell-to-cell fusion-induced senescence can also be considered a premature senescence subtype (Chuprin et al., 2013; Burton & Faragher, 2015). The distinctive phenotypic changes typical of various types of cellular senescence are cell enlargement and flattening, senescence-associated \( \beta \)-galactosidase activity (SA-\( \beta \)-gal), formation of senescence-associated heterochromatin foci (SAHF), persistent DNA damage response (DDR), and senescence-associated secretory phenotype (SASP). However, these and several other facultative features of cellular senescence that manifest in each particular case of cell cycle arrest greatly depend on the senescence-inducing stimulus and the cell type (Campisi, 2013; Salama et al., 2014).

The contribution of cellular senescence to organismal aging is a question of ongoing research (van Deursen, 2014). However, strong evidence for this connection has been reported recently. Specifically, it was shown that clearance of age-accumulated p16\(^{INK4a} \)-positive senescent cells in mice could extend their healthy lifespan (Baker et al., 2011, 2016). Several chemical compounds that specifically target senescent cells have been identified in the last 2 years (so-called senolytic drugs) (Xu et al., 2015b; Zhu et al., 2015a,b). It was shown that clearance of senescent cells by such drugs may alleviate age-related vasomotor dysfunction and frailty, enhance adipogenesis, rejuvenate haematopoietic stem cells after total-body irradiation, and, generally, extend lifespan (Xu et al., 2015a; Zhu et al., 2015b; Roos et al., 2016). Furthermore, these studies confirm the known pathological impact of cellular senescence, exemplified by cellular dysfunction, impairment of tissue regeneration, detrimental effects on tissue microenvironment, etc. (Burton & Krizhanovsky, 2014). It is evident that along with its detrimental effects, cellular senescence has clearly defined beneficial physiological functions. For instance, it has been shown recently that cellular senescence plays a role in the differentiation of megakaryocytes (Besancenot et al., 2010), the maturation of the placenta (Chuprin et al., 2013), the restriction of fibrosis (Krizhanovsky et al., 2008; Jun & Lau, 2010; Zhu et al., 2013), tissue repair (Demaria et al., 2014), and embryonic development (Nacher et al., 2006; Munoz-Espin et al., 2013; Storer et al., 2013). The role of cellular senescence in cancer prevention is well documented (Burton & Krizhanovsky, 2014; Munoz-Espin & Serrano, 2014).

It is generally agreed in the field that the most important features of cellular senescence are SASP and resistance to apoptosis (Munoz-Espin & Serrano, 2014; Burton & Faragher, 2015). SASP stimulates immune system-dependent elimination of unwanted precancerous cells or specific embryonic cells that undergo senescence. Notably, cellular senescence may serve as an alternative to apoptosis in embryonic development as well as in cancer prevention (Childs et al., 2014). It has been shown that failure to undergo senescence triggers apoptosis in a compensatory manner to eliminate transient structures during development (Munoz-Espin et al., 2013; Storer et al., 2013). Therefore, it may be reasonable to consider some of the cellular senescence states (e.g., SIPS), along with apoptosis, autophagy, necrosis, etc., in terms of the cell stress response rather than aging. However, it is unclear whether or not...
Table 1 Low molecular weight compounds that induce cellular senescence

| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|--------------------------------------|-----------|----------------------------------------------------|-------------------------------|----------------------------|---------------------------------------------|-------------------------------------------|-----------|
| DNA replication stress inducers      |           |                                                    |                               |                            |                                             |                                           |           |
| PHIDICOLIN Inhibitor of DNA          | HFF       | Senescence-like arrest                             | Growth arrest                 | p53-p21↑                   | S                                           | 150–200 µM for ~10 days                   | Marusyk et al. (2007) |
| polymerase α                         | MCF10     | Prolonged S-phase senescence-like arrest           | Large flattened cells with increased nuclear size (CS-like morphology) SA-β-Gal γH2A.X foci increased H3K9-trimethylation | p-Rb↓ p21↑ p-Rb↓ p53-p21↑ | S                                           | 1 µg ml⁻¹ for 4 days reversible state of S-phase arrest RPA foci | Maya-Mendoza et al. (2014) |
| HYDROXYUREA Ribonucleotide reductase inhibitor | HFF       | Senescence-like arrest                             | Growth inhibition             | p53-p21↑                   |                                            | 400–800 µM for ~3 weeks                    | Yeo et al. (2000) |
|                                      | McA-RH7777| Senescence-like arrest                             | Growth inhibition             | p21↑                       | G1                                          |                                           | Hong et al. (2004) |
|                                      | HFF       | Senescence-like arrest                             | Growth inhibition             | p53-p21↑ p-p53 Ser15↑ p-p53 Ser20↑ p16-independent |                                           |                                             | Marusyk et al. (2007) |
|                                      | REF52     | Senescence-like arrest                             | CS-like morphology SA-β-Gal γH2A.X foci | p53-p21↑ |                                           |                                             | Park et al. (2000) |
|                                      | MCF10A    | Senescence-like arrest                             | SA-β-Gal                      | p16↑ p21↑ p27↑             | G1                                          |                                           | Sumikawa et al. (2005), Kobayashi et al. (2012) |
|                                      | K562      | Senescence-like arrest                             | SA-β-Gal                      | p53-p21↑                   |                                            |                                             |                                      |
| Thymidine Excess of thymidine inhibits DNA replication by reducing the amount of dCTP synthesized | HeLa      | Premature senescence                             | Growth inhibition             | p21↑                       | S                                           | 1.5 µM for 7–10 days                      | Nair et al. (2015) |
|                                      | TIG-7     | Senescence-like arrest                             | CS-like morphology SA-β-Gal   | p53-p21↑                   | G2                                          | 200 µM for 7 days Chk1Ser345p↑ Chk2Thr68p↑ | Masterson & O'Dea (2007) |
|                                      | HeLa      | Senescence-like arrest                             | Growth inhibition             | p53-p21↑ p-Rb↓ p27↑         | S                                           |                                             | Eriko et al. (1999), Suzuki et al. (2001) |
|                                      | A549      | Senescence-like arrest                             | CS-like morphology SA-β-Gal γH2AX foci SASP | p21↑             |                                             |                                             |                                      |
|                                      | A549      | Senescence-like arrest                             | CS-like morphology γH2AX foci SASP | p21↑             | G1                                          | 100 µM for 48 h elevated ROS levels p53 activation in A549 cells |                                      |

(Continued)
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|-------------------------------------|-----------|---------------------------------------------------|-------------------------------|---------------------------|---------------------------------------------|---------------------------------------------|------------|
| 2′,2′-DIFLUORODEOXYCYTIDINE (GEMCITABINE) Inhibits ribonucleotide reductase Inhibits CTP synthetase | AsPC1 PANC-1 | Premature senescence | Growth inhibition CS-like morphology SA-β-Gal | p21↑ | Sub-G1 | 100 nM for 4 days | Modrak et al. (2009) |
| CYCLOPENTENYL CYTOSINE Inhibits CTP synthetase | MCF-7 | Premature senescence | Growth inhibition CS-like morphology SA-β-Gal | p53▼ p53, p21↑ | G2 | 0.125–1 μM for 5 days | Huang et al. (2011) |
| (2) DNA-damaging agents (2a) DNA topoisomerases inhibitors | | | | | | | |
| DORORUBICIN DNA intercalator Induces DSBs by poisoning DNA topoisomerase II | 11 cell lines derived from different types of human solid tumors HCT116 | Senescence-like phenotype | Growth inhibition CS-like morphology SA-β-gal | Can be dependent or not on p53 activation | | | Chang et al. (1999a) |
| DOXORUBICIN DNA intercalator Induces DSBs by poisoning DNA topoisomerase II | MCF7 | Premature senescence | CS-like morphology SA-β-gal | p53▼ p53-p21▼ p21↑ | G2 phase | 50–100 nM for 1–4 days treatment led to the appearance of a substantial fraction of polyploid nuclei in p53−/− and p21−/− lines | Chang et al. (1999b) |
| | HCT116 | Senescence-like phenotype | | | | | |
| | Neonatal rat cardiomyocytes H9c2 | Premature senescence | CS-like morphology SA-β-gal | p53▼ p-p38↑ p-JNK↑ p-ERK↑ MAPK (p-38 and JNK)▼ mTOR▼ p53, p21↑ | G2 | 100 ng mL−1 for 1–4 days low p53 levels during prolonged cell cycle arrest lead to senescence, while high levels of p53—to either quiescence or cell death | Leontieva et al. (2010) |
| | WI38 | Premature senescence | CS-like morphology SA-β-gal | | | | |
| | A549 | Transient senescence-like state | CS-like morphology SA-β-gal | | G2 | 50–200 nM for 72 h | Litwiniec et al. (2010) |
| | | | | | | | |
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|-------------------------------------|-----------|---------------------------------------------------|-------------------------------|-----------------------------|-----------------------------------------------|---------------------------------------------|------------|
| MMTV-Wnt1 mice                     | MCF7      | Premature senescence                              | Growth inhibition SA-β-gal   | p53<sup>▼</sup>, +/- p21<sup>▼</sup> | G1 (p53-and p21-dependent), G2 (p21-independent) | 4 mg kg<sup>-1</sup> day<sup>-1</sup> for 5 days, MCF7 treated with 200 nM for 24 h in vivo | Jackson et al. (2012) |
| Cardiac progenitor cells            |           | Premature senescence                              | CS-like morphology SA-β-gal γH2AX | p16†                      | G1                                             | 0.1–1 μM for 24–48 h in vivo                | Piegari et al. (2013) |
| DU145                               | LNCaP PC3 | Premature senescence                              | Growth inhibition CS-like morphology SA-β-Gal | p21†, p27†, p-Rb†, p53-independent | G1                                             | 10 nM for 1–5 days                           | Park et al. (2006) |
| ETOPOSIDE                           | LS174T A2780 MCF7 WI38 | Premature senescence                              | Growth arrest CS-like morphology SA-β-gal | p53<sup>▼</sup>, p-p53<sup>▼</sup>, p21† | G1                                             | 2 μM for 24 h                               | te Poele et al. (2002) |
| Daunorubicin DNA intercalator       | Jurkat    | Senescence-like phenotype                         | Growth inhibition CS-like morphology SA-β-Gal | p53-p21†                  | G2                                             | 0.75–3 μM for 72 h Polyploid (higher DNA contents (>G2)) | Litwiniec et al. (2013) |
| Mitoxantrone Topoisomerase II       | A549      | Senescence-like phenotype                         | SAHF                         |                             |                                               |                                             |                        |
| Topoisomerase II                    | WI-38     | Premature senescence                              | SASP                          | p21†, p16†                 | G2                                             | 91 nM for 24 h                              | Mansilla et al. (2003) |
| MITOXANTRONE Topoisomerase II       | A549      | Senescence-like phenotype                         | Growth inhibition CS-like morphology SA-β-Gal | p21†, p-ATMSer1981†        | G1                                             | 2 nM for 2–5 days                            | Zhao et al. (2010) |

(Continued)
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|--------------------------------------|-----------|-----------------------------------------------|----------------------------|---------------------------|-----------------------------------------------|------------------------------------------|-----------|
| **CAMPTOTHECIN AND SN-38**           | LS174T    | Senescence-like arrest                        | Growth arrest              | p53-p21↑ p16↑            | G1                                            | 6–100 ng mL⁻¹ for 24–168 h                 | te Poele et al. (2002)                  |
| Topoisomerase I poison               | MCF-7     |                                               | CS-like morphology         |                           | S                                             |                                         |                                      |
| Induces SSBs                         | A2780     |                                               | SA-β-gal                   |                           | G2                                            |                                         |                                      |
|                                       | HCT116    |                                               |                            |                           |                                               |                                         |                                      |
|                                       | LS174T    | Senescence-like arrest                        | Growth arrest              |                           | S                                             |                                         |                                      |
|                                       | HCT116    |                                               |                            |                           | G2                                            |                                         |                                      |
|                                       | H1299     | Premature senescence                         | CS-like morphology         |                           |                                               |                                         |                                      |
|                                       | HCT116    | Premature senescence                         | Growth inhibition          |                           |                                               |                                         |                                      |
|                                       | HeLa      | Senescence-like growth arrest                 | Growth inhibition          |                           |                                               |                                         |                                      |
| (2b) DNA cross-linkers               | CNE1      | Senescence-like arrest                        | Growth inhibition          |                           |                                               |                                         |                                      |
| DNA-alkylating agent                 |           |                                               |                            |                           |                                               |                                         |                                      |
| Induces DNA intrastrand cross-links  |           |                                               |                            |                           |                                               |                                         |                                      |
| Normal human lung fibroblasts        |           |                                               |                            |                           |                                               |                                         |                                      |
| Human non-small cell lung cancer cells |           |                                               |                            |                           |                                               |                                         |                                      |
| HCT116                               |           | Premature senescence                         | Growth inhibition          |                           |                                               |                                         |                                      |
|                                       |           |                                               |                            |                           |                                               |                                         |                                      |
| HepG2                                |           | Premature senescence                         | Growth inhibition          |                           |                                               |                                         |                                      |
| CCL23                                |           | Premature senescence                         | Growth inhibition          |                           |                                               |                                         |                                      |
| CAL27                                |           |                                               |                            |                           |                                               |                                         |                                      |
| UM-SCC1                              |           |                                               |                            |                           |                                               |                                         |                                      |
| UM-SCC14A                            |           |                                               |                            |                           |                                               |                                         |                                      |
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|-------------------------------------|-----------|------------------------------------------------------|-------------------------------|-----------------------------|---------------------------------------------|---------------------------------------------|-----------|
| **DNA-alkylating agent**            | A549      | Premature senescence                                 | Growth inhibition CS-like morphology SA-β-Gal γH2AX | p21↑                        | G2                                          | 0.01–0.02 μg mL⁻¹ for 6 days                | McKenna et al. (2012) |
| **DNA-alkylating agent**            | Murine bone marrow cells | Premature senescence                                 | Growth inhibition SA-β-Gal | p16↑, p19↑                  | G2                                          | 30 μM for 6 h                               | Meng et al. (2003) |
| **DNA-alkylating agent**            | WI38      | Premature senescence                                 | Growth inhibition CS-like morphology SA-β-gal | MAPK(p38, ERK)↑, p-p38↑, p-JNK↑, p-ERK↑, p21↑, p16↑ | G2                                          | 7.5–120 μM for 24 h                         | Probin et al., 2006, 2007 |
| **DNA-intercalating agent**         | Lymphoma-bearing C57BL/6 mice | Premature senescence                                 | SA-β-gal                      | p53↑, p16↑                  | G2                                          | 300 mg kg⁻¹ day⁻¹ for 7 days in vivo        | Schmitt et al. (2002) |
| **DNA-intercalating agent**         | TIG-7     | Premature senescence                                 | Growth inhibition SA-β-gal    | MAPK (p-p38, p-JNK, p-ERK)↑, p21↑, p16↑ | G1                                          | 10 μM for 14 days                           | Palaniyappan (2009) |
| **DNA-intercalating agent**         | DU145     | Premature senescence                                 | CS-like morphology SA-β-gal   |                          | G1                                          | 0.25–10 μM for 3 days                       | Ewald et al. (2009) |

(2c) DNA-damaging drugs with complex effects

| Small molecules that induce cellular senescence, N. V. Petrova et al. |
|---------------------------------------------------------------|
| DNA-intercalator: Actinomycin D                              | Normal human fibroblasts                                    | Premature senescence          | Growth inhibition CS-like morphology SA-β-Gal γ-H2AX foci | p53-p21↑ | G1 | 0.04 mg mL⁻¹ for 12 h | Robles and Adami (1998) |
| Can poison topoisomerase I and II, and, thus induce SSBs and DSBs | Human mesenchymal stem cells                  | Premature senescence          | Growth inhibition CS-like morphology SA-β-Gal γ-H2AX foci | p53-p21↑, p16↑ | G2 | 400 mM for 3–21 days | Minieri et al. (2015) |
| BLEOMYCIN:                                                   | Normal human fibroblasts                                    | Premature senescence          | Growth inhibition SA-β-gal   | p53-p21↑ | G1 | 0.06 units mL⁻¹ for 12–24 h | Robles and Adami (1998) |
| Induces DNA breaks                                           | A549 Rat primary type II cells C57BL/6J mice               | Premature senescence          | Growth inhibition CS-like morphology SA-β-gal   | p16↑, p21↑ | G2 | 50 μg mL⁻¹ for 120 h or 5 mg kg⁻¹ day⁻¹ for 7–21 days in vivo | Aoshiba et al. (2003) |
|                                                            | A549                                                        | Premature senescence          | Growth inhibition CS-like morphology SA-β-gal   | p53-p21↑ | G2 | 50 μM mL⁻¹ for 1–7 days siRNA for caveolin-1 reduces SA-β-gal | Linge et al. (2007) |
Table 1 (Continued)

| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|--------------------------------------|-----------|-----------------------------------------------------|-------------------------------|-----------------------------|---------------------------------------------|---------------------------------------------|------------|
| TEMOZOLOMIDE DNA-alkylating agent    | BJ        | Premature senescence                               | CS-like morphology SA-β-gal | p21↑                        | 100 μg mL⁻¹ for 24 h                         |                                             | Pazelli et al. (2012) |
| Alkylates/methylates DNA Induces DNA damage | C57BL/6J mice | Premature senescence | γH2AX                       | p21↑                        | 2.5 mg kg⁻¹ day⁻¹ for 7–21 days             |                                             | Aoshiba et al. (2013) |
|                                       | U-87 MG   | Premature senescence                               | Growth inhibition SA-β-Gal  | p33↑, p21↑                  | G2                                          | 100 μM for 3 h the gradual appearance of hyperploid cells | Hirose et al. (2001) |
| (3) Epigenetic modifiers             | Me4405 IR3 | Premature senescence                               | Growth inhibition SA-β-Gal  | p33↑, p21↑                  | G2                                          | 25–100 μM for 72 h the gradual appearance of hyperploid cells | Mhaidat et al. (2007) |
| 5-Aza-2'-deoxycytidine Inhibitor of DNA methyltransferases induces DSBs | MDAH041    | Premature senescence                               | CS-like morphology SA-β-gal | p16↑                        | S                                           | 1 μM for 6 days                                | Vogt et al. (1998) |
|                                      | HepG2     | Premature senescence                               | Growth inhibition SA-β-gal  | p16↑                        |                                             |                                             | Venturelli et al. (2013) |
|                                      | NMRI mice | Premature senescence                               | SA-β-gal                    | p16↑                        |                                             |                                             | Xing et al. (2013) |
|                                      | U2OS      | Premature senescence                               | Growth inhibition SA-β-gal  | p16↑                        |                                             |                                             | Widodo et al. (2007) |
|                                      | H28       | Premature senescence                               | Growth inhibition SA-β-gal  | p16↑                        |                                             |                                             | Amatari et al. (2011) |
| SODIUM BUTYRATE Class I and II histone deacetylase (HDAC) inhibitor | WI38       | Senescence-like state                               | Growth inhibition SA-β-gal  | p- Rb ↓                      | G1                                          | 0.5 mM for ~20 days                           | Ogryzko et al. (1996) |
|                                      | NIH3T3    | Senescence-like state                               | CS-like morphology SA-β-gal | p21↑                        | G1                                          | 5–10 mM for 48 h activation of p21 expression may be both p53-dependent and p53-independent | Xiao et al. (1997) |
|                                      | HHUA      | Senescence-like state                               | SA-β-gal                    | p21↑                        | G1                                          | 1–4 mM for 2–5 days activation of p21 expression may be both | Terao et al. (2001) |
|                                      | Hec1-A    | Senescence-like state                               | SA-β-gal                    | p-Rb↓                       | G1                                          |                                             | Xiong et al. (2013) |
|                                      | SKOV-3    | Senescence-like state                               | SA-β-gal                    | p-Rb↓                       | G2                                          |                                             | Chen et al. (2013) |
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/Other notes (if any) | References |
|-------------------------------------|-----------|---------------------------------------------|-------------------------------|---------------------------|---------------------------------------------|-------------------------------------------|------------|
| HeLa                               |           | Senescence-like state                       | CS-like morphology, SA-β-gal | p21↑                      | G1                                          | 4 mM for 24 h or 0.5 mM for 14 days       | Place et al. (2005) |
| SiHa                               |           | Senescence-like state                       |                               | p21↑, p16↑                 | G1                                          | 4 mM for 24–72 h DDR without detectable DNA damage | Abramova et al. (2006), Pospelova et al. (2009) |
| Wi-38                              | BJ 293T   | Premature senescence                        | CS-like morphology, SASP     | p53- and RB-independent   | G1                                          | 4 mM for 3–6 days SASP dependent upon ATM and NF-κB | Pozolli et al. (2012) |
|                                    |           |                                             |                               |                           |                                             | 10 ng mL⁻¹ for ~30 days                    | Oryzsko et al. (1996) |
| E1A + Ras-transfected rat and mouse embryonic fibroblasts | Wi38      | Senescence-like state                       | Growth inhibition            | p21↑                      | G1 phase                                    | 2 μM for 24–72 h or 0.5 μM for 9 days     | Place et al. (2005) |
|                                   | BJ 293T   | Premature senescence                        | CS-like morphology, SA-β-gal | p53- and RB-independent   | G1                                          | 1 mM for 3 days lack of DNA damage         | Pozolli et al. (2012) |
|                                   |           |                                             | SASP                          |                           |                                             |                                          |            |
|                                   | AS49      | Premature senescence                        | Growth inhibition            | p21↑                      | G1                                          | 0.5–1.0 μM for 48 h                       | Zhao et al. (2010) |
|                                   |           |                                              | SASP                          |                           |                                             |                                          |            |
|                                   | B143      | Premature senescence                        | CS-like morphology, SA-β-gal | p21↑, p27↑                 | G1                                          | 0.4–1 μM for 5 days induced polypliod cells | Xu et al. (2005) |
|                                   | MG-63     |                                              |                               |                           | G2                                          |                                          |            |
|                                   | Saos-2    |                                              |                               |                           |                                             |                                          |            |
|                                   | SJSA      |                                              |                               |                           |                                             |                                          |            |
|                                   | U2OS human osteosarcoma cell lines        |                                              |                               |                           |                                             |                                          |            |
|                                   |          |                                              |                               |                           |                                             |                                          |            |
|                                   | LB589 (Panobinostat)                      | Premature senescence                        | CS-like morphology, SA-β-Gal | p53-independent           | G1                                          | 15 μM for 21 days aysor 2–10 mg kg⁻¹ day⁻¹ for 17 days in vivo (mice) | Cain et al. (2013) |
|                                   | B143      |                                              |                               |                           | G2                                          |                                          |            |
|                                   | MCF7      |                                              |                               |                           |                                             |                                          |            |
|                                   | HT1080    |                                              |                               |                           |                                             |                                          |            |
|                                   |          |                                              |                               |                           |                                             |                                          |            |
|                                   | D283-Med  |                                              |                               |                           |                                             |                                          |            |
|                                   | DAOY      |                                              |                               |                           |                                             |                                          |            |
|                                   | PFSK      |                                              |                               |                           |                                             |                                          |            |
|                                   | Bel-7402  |                                              |                               |                           |                                             |                                          |            |
|                                   | Bel-7404  |                                              |                               |                           |                                             |                                          |            |
|                                   |          |                                              |                               |                           |                                             |                                          |            |
| Small compounds/  | Cell line      | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References            |
|-------------------|----------------|-----------------------------------------------------|--------------------------------|----------------------------|-----------------------------------------------|---------------------------------------------|------------------------|
| CURCUMIN AND C646 | TIG3           | Senescence-like state                               | SA-β-gal                      | p53-, p21- and p16-independent | G2                                            | 6–9 µM for 2–15 days Global H3, H4 hypothesis Lack of DNA damage | Prieur et al. (2011)   |
| p300 histone      |                |                                                     | SAHF                           |                            |                                               |                                             |                        |
| acetyltransferase inhibitors |        |                                                     |                                |                            |                                               |                                             |                        |
| HCT116            | MCF 7          | Premature senescence                               | CS-like morphology             | p21†                       | G2                                            | 10 µM (HCT116) or 15 µM (MCF 7) or 7.5 µM (U2OS) for 24 h | Mosieniak et al. (2012) |
| U2OS              | CAF myofibroblasts | Premature senescence                               | CS-like morphology             | p16↑                        |                                               |                                             |                        |
| VSMC endothelial cells derived from aorta |                | Premature senescence                               | Growth inhibition              | p21↑                        | G2                                            | 5–7.5 µM (VSMCs) and 2.5–5 µM (endothelial cells) for 3–7 days ROS- and ATM-independent | Grabowska et al. (2015) |
| BRD4770           | K562           | Premature senescence                               | Growth inhibition              | p16↑                        |                                               |                                             |                        |
| G9a histone       | SW620          |                                                     | SA-β-Gal                       | p21↑                        |                                               |                                             |                        |
| methyltransferase inhibitor |        |                                                     |                                | p27↑                        |                                               |                                             |                        |
| SYUIQ-5           |                |                                                     |                                | p-ATM↑                       |                                               |                                             |                        |
| (4) Inhibitors of telomerase activity |        |                                                     |                                |                             |                                               |                                             |                        |
| BMVC4             |                |                                                     |                                |                             |                                               |                                             |                        |
| Stabilizes G-quadruplexes |        |                                                     |                                |                             |                                               |                                             |                        |
| HeLa              |                |                                                     |                                |                             |                                               |                                             |                        |
| VAI13             |                |                                                     |                                |                             |                                               |                                             |                        |
| Saco52            |                |                                                     |                                |                             |                                               |                                             |                        |
| U2OS              |                |                                                     |                                |                             |                                               |                                             |                        |
| PYRIDOSTATIN      |                |                                                     |                                |                             |                                               |                                             |                        |
| Stabilizes G-quadruplexes |        |                                                     |                                |                             |                                               |                                             |                        |
| HT1080            |                |                                                     |                                |                             |                                               |                                             |                        |
| Stabilizes G-quadruplexes |        |                                                     |                                |                             |                                               |                                             |                        |
| COMPOUND 115405   |                |                                                     |                                |                             |                                               |                                             |                        |
| G-quadruplex ligand |                |                                                     |                                |                             |                                               |                                             |                        |
| TAKA et al.       |                |                                                     |                                |                             |                                               |                                             |                        |
| PERYLENE DERIVATIVES PM2 AND PIPER |        |                                                     |                                |                             |                                               |                                             |                        |
| Induce G-quadruplex formation from both telomeric DNA and hTERT promoter region |        |                                                     |                                |                             |                                               |                                             |                        |
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|-------------------------------------|-----------|---------------------------------------------------|-------------------------------|-----------------------------|---------------------------------------------|---------------------------------------------|------------|
| HARMINE, β-carboline alkaloid       | MCF7      | Premature senescence                              | Growth inhibition             | p53-p21↑                    | 20–30 μM for 48–96 h                        | Zhao and Wink (2013)                         |
| INDOLE DERIVATIVES (INDOLE-3-CARBINOL (I3C) AND INDOLY SULFATE) Phytochemicals that downregulate hTERT expression | MCF7 | Premature senescence                              | Growth inhibition             | p53↑, p-p53Ser15↑                 | G1                                           | 200 μM for 48 h                               | Marconett et al. (2011) |
| HK-2                                | CRF rats  | Premature senescence                              | Growth inhibition             | p53↑                        | 250 μM for 48–120 h or 4 g kg⁻¹ for 16 weeks | Shimizu et al. (2010)                       |
| INDOLE DERIVATIVES (INDOLE-3-CARBINOL (I3C) AND INDOLY SULFATE) Phytochemicals that downregulate hTERT expression | HK-2      | Premature senescence                              | Growth inhibition             | p53↑                        | 250 μM for 48–72 h | 10 mg for 130 days | Shimizu et al. (2011) |
| BIBR1532 Non-nucleosidic TERT inhibitor | NCI-H460  | Senescence-like phenotype                         | Growth inhibition             | NF-κB↓                      | 250 μM for 48–72 h | ROS↑                                            | Damm et al. (2001) |
| AZIDOTHYMIDINE (AZT) Reverse transcriptase inhibitor | MCF-7 | Senescence-like arrest                             | Growth inhibition             | p21↑, p27↑                   | 50 μM for 18 weeks | in vivo (ATL patients) the Jurkat T-cell line, treated under the same conditions, did not enter growth arrest | Datta et al. (2006) |
| Inhibits telomerase activity         | HTLV-I    | Premature senescence                              | Growth inhibition             | mTOR, MEK↓                   | 0.5 μM for 3–7 days                | Leontieva and Blagosklonny (2013) |
| MASC C57BL/6 mice                   |            | Premature senescence                              | Growth inhibition             | mTOR↑, pRb↓                  | 1 μM for 5 days                   | Leontieva et al. (2013) |
| 12 sarcoma cell lines generated directly from patient samples | MCF7 | Premature senescence                              | Growth inhibition             | pRb↓                        | G1                                           | 9–27 μM for 2–4 days or 100 mg kg⁻¹ day⁻¹ for 3 weeks | Perez et al. (2015) |
|                                    |           |                                                   |                               |                             |                                              | in vivo (mice)                              |
|                                    |           |                                                   |                               |                             |                                              | 1 μM for 5–7 days, 0.6 or 3.0 mg kg⁻¹ day for 10 cycles each consisting of 3 weeks | Hu et al. (2015) |
|                                    |           |                                                   |                               |                             |                                              | in vivo (dogs)                              |

(Continued)
| Small compounds/ Mechanism of action | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|--------------------------------------|---------------------------------------------------|-------------------------------|-----------------------------|---------------------------------------------|-------------------------------------------|------------|
| **1205Lu** | Premature senescence | Growth inhibition | pRb↓, mTOR↓ | G1 | 1 μM for 8 days or 90 mg kg⁻¹ day for 14 days | Yoshida et al. (2016) |
| **WM983** | Premature senescence | Growth inhibition | p53-p21↑ | G1 | 1–10 μg mL⁻¹ for 24 h | Park et al. (2009) |
| **WM983BR** | Premature senescence | Growth inhibition | p-Rb↓ | G1 | 500 μM for 6 days or 200 mg kg⁻¹ for 21 days | Rader et al. (2013) |
| **WM451Lu** | Premature senescence | Growth inhibition | p53↑ | G1 | 5 or 10 μM for 1–7 days; no apoptosis was observed | Efeyan et al. (2007) |
| **WM239A** | Premature senescence | Growth inhibition | p21↑ | G1 | 10 μM for 5 days | Huang et al. (2011) |
| **WM3918** | Premature senescence | Growth inhibition | p53↑ | G1 | 2.5–10 μM for 24–72 h; induces apoptosis | Manfe et al. (2012) |
| **RTE** | Premature senescence | Growth inhibition | p53↑ | G1 | 10 μM for 3 days | Ling et al. (2014) |
| **MDCK** | Premature senescence | Growth inhibition | p21↑ | G1 | 0.1–1 μg mL⁻¹ for 24 h; telomerase was selectively repressed; normal human fibroblasts were resistant to treatment | Cozzi et al. (2006) |
| Small molecules that induce cellular senescence, N. V. Petrova et al. | 437 | 883 | 437 | 883 |
|---|---|---|---|---|
| **Table 1** (Continued) | | | | |
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
| | | | | | | | |
| PEPO05 (INGENOL-3-ANGELATE) | H358, H441, H322 | Premature senescence | Growth inhibition | p21↑, pRb↓ | G2 | 100 nM for 30 min reduced telomerase activity | Oliva et al. (2008) |
| | SK-MEL-5, MCF7, Colo-205 | Senescence-like arrest | Growth inhibition | p21↑, ERK1/2↑, p-Rb↓ | G2 | 10-1000 ng mL^-1 for 24 h | Mason et al. (2010) |
| | D04, D08, MM127, MM455 | Premature senescence | Growth inhibition | p21↑, ERK↑, p-Rb↓ | G1 | 0.2-1 μg mL^-1 for 24 h telomerase was selectively repressed normal human fibroblasts were resistant to treatment | Cozzi et al. (2006) |
| | | | | | | | |
| PEPO08 (20-O-ACETYL-INGENOL-3-ANGELATE) | SK-MEL-5, MCF7, Colo-205 | Senescence-like arrest | Growth inhibition | p21↑, ERK↑ | G2 | 10-1000 ng mL^-1 for 24 h or 5-6 days | Mason et al. (2010) |
| | | | | | | | |
| ROS inducers | F65 | Senescence-like arrest | CS-like morphology | p21↑, SA-β-Gal | G1 | 200 μM for 2 h | Chen and Ames (1994) |
| | IMR-90 | Senescence-like arrest | CS-like morphology | p53-p21↑, SA-β-Gal | G1 | 300 μM for 2 h | Chen et al. (1998) |
| | IMR-90 | Senescence-like arrest | CS-like morphology | p53-p21↑, SA-β-Gal | G1 | 150 μM for 2 h | Frippiat et al. (2002) |
| | 2BS | Premature senescence | CS-like morphology | p53-p21↑, SA-β-Gal | G1 | 10 μM for 3 weeks accumulation of DNA damage accelerated telomere shortening | Duan et al. (2005) |
| | A549 | Premature senescence | CS-like morphology | p53-p21↑, SA-β-Gal | G1 | 100 μM for 2 h | Yoshizaki et al. (2009) |
| | Primary human keratinocytes | Premature senescence | CS-like morphology | p53-p21↑, SA-β-Gal | G1 | 50 μM for 2 h | Ido et al. (2012) |
| | HUVEC | Premature senescence | CS-like morphology | p53-p21↑, SA-β-Gal, SASP | G1 | 100 μM for 1 h | Suzuki et al. (2013) |
| | hMESCs | Premature senescence | CS-like morphology | p53-p21↑, SA-β-Gal | G1 | 200 μM for 1 h | Burova et al. (2013), Borodkina et al. (2014) |
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|--------------------------------------|-----------|---------------------------------------------------|--------------------------------|----------------------------|-----------------------------------------------|------------------------------------------|------------|
| **Wt-38**                            | Premature senescence | SA-β-Gal                                           | γH2A.X and p-53BP1 foci         | p-p38↑, p53-p21↑, p-Rb↓, p21↑                 |                                              | 500 μM for 2 h                          | Gorbunova et al. (2002) |
| **IMR-90**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 150 μM for 2 h caveolin ↑               | Chretien et al. (2008) |
| **IMR-90**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 200 μM for 2 h                          | Zdanov et al. (2006) |
| **HUVEC**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 100 μM for 1 h                          | Ota et al. (2008) |
| **MRC-5**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 5 x 30 μM for 1 h day^{-1}              | von Zglinicki et al. (2000) |
| **Wt-38**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 100 μM for 2 days                         | Dumont et al. (2000) |
| **HUVEC**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 5 x 30 μM for 1 h day^{-1}              | von Zglinicki et al. (2000) |
| **Wt-38**                            | Premature senescence | SA-β-Gal                                           | CS-like morphology             | p21↑, p-ERK↑, p-Akt↑, p-p38↑, p21↑, p53↑      |                                              | 4 x 100 μM for 1 h per every two doubling | Chretien et al. (2008) |
| **Human mesangial cells**            | Senescence-like arrest | SA-β-Gal                                           | CS-like morphology             | JAK2-STAT↑                                 |                                              | 30 μM for 1 h                           | Zhou et al. (2013) |
| **Phenyl-2-pyridyl ketoxime (PPK0)** | **Primary human fibroblasts were isolated from newborn foreskins** | SA-β-Gal                                           | CS-like morphology             | p53-p21↑, p16↑, ERK1/ERK2↑, ROS- and NO↑- dependent |                                              | 1 mM for 3 days                         | Yang et al. (2016) |

(Continued)
| Small compounds/ Mechanism of action | Cell line | Cellular senescence state (as described by authors) | Senescence markers documented | Signaling pathways involved | Cell cycle phase of the stable growth arrest | Triggering conditions/ Other notes (if any) | References |
|-------------------------------------|-----------|---------------------------------------------------|------------------------------|---------------------------|---------------------------------|---------------------------------|-----------|
| PHENYLAMINONAPHTHOQUINONES (Q7 AND Q9) | T24       | Senescence-like arrest                            | CS-like morphology SA-β-Gal  | MAPK ▼                       | G2                               | 4 μM for 1–3 days alone or with 1 mM ascorbate | Felipe et al. (2013) |
| PARAQUAT                            | TIG-7     | Premature senescence                             | CS-like morphology SA-β-Gal  |                          |                                  | 100 μM for 4 days               | Joguchi et al. (2004)      |
| BALB/c mice                         | BJ        | Premature senescence                             | CS-like morphology SA-β-Gal  | p53↑ p21↑                   | p27↑                            | 25 mg kg⁻¹ for 3 days (intraperitoneal injection) | Ota et al. (2008)         |
|                                    | BJ        |                                                   |                              |                           |                                 | 350 μM for 16 h thioredoxin 1↑   | Young et al. (2010)        |

*Cell lines: 1205Lu, human lung melanoma cells; 28S, human embryonic lung fibroblasts; A2780, human ovarian carcinoma cells; AS49, human lung adenocarcinoma epithelial cells; AsPC-1, human pancreas adenocarcinoma cells; B143, human osteosarcoma cells; Bel-7402, human hepatocellular carcinoma cells; Bel-7404, human hepatocellular carcinoma cells; BJ, normal human foreskin fibroblasts; CAF, cancer-associated fibroblasts; CAL27, human oral adenosquamous carcinoma cells; CNE1, human nasopharyngeal carcinoma cells; D283-Med, human medulloblastoma cells; DAOY, human cerebellar medulloblastoma cells; DU145, human prostate carcinoma cells; F65, human foreskin fibroblasts; H1299, human lung carcinoma cells; H28, human mesothelioma cells; H92, rat cardiomyoblasts; HCA2, normal human foreskin fibroblasts; HCT116, human colorectal carcinoma cells; HCT8, human ileocoeal colorectal adenocarcinoma cells; Hec1-A, human uterus/ endometrium adenocarcinoma cells; HeLa, human cervix adenocarcinoma cells; HepG2, human hepatocellular carcinoma cells; HFF, human foreskin normal fibroblasts; HUVEC, human umbilical vein endothelial cells; IMR-90, human fetal lung fibroblasts; Jurkat, human T-cell leukemia cells; K562, human bone marrow myelogenous leukemia lymphoblasts; L1, human embryonic lung fibroblasts; LNCaP, human prostate carcinoma cells; LS174T, human colorectal adenocarcinoma cells; Mac1, human cutaneous T-cell lymphoma (CTCL); Mac2a, human cutaneous T-cell lymphoma (CTCL); MASC, mouse multipotent astrocytic stem cell; MCA-RH7777, rat hepatoma cells; MCF10, human breast fibrocytic cells; MCF7, human breast adenocarcinoma cells; MDAH041, derive from the fibroblasts of a patient with Li-Fraumeni syndrome; MDCK, canine epithelial kidney cells; MEF, mouse embryonic fibroblasts; MEL10 (SK-MEL-147), human melanoma cells; MG-63, human osteosarcoma cells; MRC-5, human lung fibroblast; MyLa2000, human cutaneous T-cell lymphoma (CTCL); NCl-H460, human lung carcinoma cells; NIH3T3, mouse embryo fibroblasts; PANC-1, human pancreatic carcinoma cells; PC-3, human prostate cancer cells; PFSK-1, human neuroectodermal cells derived from cerebral brain tumor; RFS52, rat embryonic fibroblasts; RPE, human retinal pigment epithelial cells; RTE, rat trabecular epithelial cells; Saos-2, human osteosarcoma cells; SiHa, human cervix squamous cell carcinoma cells; SJSA-1, human osteosarcoma cells; SKN-SH, human neuroblastoma cells; SKOV-3, human ovarian adenocarcinoma cells; SW620, human colon cancer cells; T24, human bladder carcinoma cells; TIG-3, human embryonic lung fibroblasts; TIG-7, human embryonic lung fibroblasts; U2OS, human osteosarcoma cells; U-87 MG, human glioblastoma, astrocytoma cells; U-M SCC1, human squamous carcinoma of the oral cavity cells; UM-SCC14A, human squamous carcinoma of the oral cavity cells; VA-13, human lung fibroblasts; VSMC, vascular smooth muscle cells; W38, human lung fibroblasts; WM239A, human melanoma cells; WM391B, human melanoma cells; WM451Lu, human melanoma cells; WM9838, human melanoma cells; WT 9-7, human cells from a patient with autosomal-dominant polycystic kidney disease (ADPKD). |

†Abbreviations: CS, cellular senescence; DSBs, double-stranded DNA breaks; SA-β-gal, senescence-associated β-galactosidase; SAHF, senescence-associated heterochromatin foci; SASP, senescence-associated secretory phenotype; SSBs, single-stranded DNA breaks. |

‡Symbols: ▲, increased activity/expression reported; ▼, decreased activity/expression reported; ▼, involvement of the protein/pathway was verified by gene(s) knockout or knockdown, inhibitory analysis, and/or using cell lines carrying inactivating mutations.
the cellular senescence that is widely implicated in normal aging, chronic diseases, tumor suppression, tumorigenesis, cell differentiation, and embryogenesis represents a single physiological cellular state.

To date, dozens of stress-induced cellular senescence phenotypes have been reported. These senescence states may differ substantially from each other, as well as from replicative senescence, through the presence of specific senescence features. Additionally, it has been reported that some stress-induced senescence states can be overcome, thus challenging the dogma that cellular senescence is an irreversible form of growth arrest (Romanov et al., 2001; Beausejour et al., 2003). Such caveats can lead to confusion regarding the terminology of stress-induced cellular senescence states; it is not clear whether senescence-like growth arrest (and variations thereof) resembles ‘true’ cellular senescence. The indispensable characteristics of this ‘true’ cellular senescence are also elusive. It can be argued that SASP (arising along with morphological changes and SA-β-gal) may be the most important physiologically relevant feature of cellular senescence; however, SASP has not been studied in most cases of stress-induced senescence. Here, we attempt to catalog virtually all of the cellular senescence-like states that can be induced by low molecular weight compounds (Table 1). We summarize the biological markers, molecular pathways involved in senescence establishment, and specific traits of cellular senescence states induced by small compounds, as well as the treatment conditions used. In total, we analyzed more than 50 chemical inducers of cellular senescence and senescence-like states. These chemical compounds can be functionally classified into eight groups: (1) DNA replication stress inducers (aphidicolin, hydroxyurea, thymidine, bromodeoxyuridine, difluorodeoxycytidine, cyclopentenyl cytosine); (2) DNA-damaging agents, including (2a) DNA topoisomerase inhibitors (doxorubicin, etoposide, daunorubicin, mitoxantrone, camptothecin), (2b) DNA cross-linkers (cisplatin, mitomycin C, busulfan, cyclophosphamide, diaziquone), and (2c) drugs with complex effects (actinomycin D, bleomycin, temozolomide); (3) epigenetic modifiers that inhibit DNA methyltransferases (5-aza-2-deoxycytidine), histone deacetylases (sodium butyrate, trichostatin A, MS-275, SAHA, LBH589, phenylbutyric acid, valproic acid), histone acetyltransferases (curcumin, C646), and histone methyltransferases (BRD47770); (4) inhibitors of telomerase activity (SU1,590, BMVC4, pyridostatin, compound 115405, perylene and indole derivatives, harmine, BIBR1532, azidothymidine); (5) inhibitors of cyclin-dependent kinases (palbociclib, roscovitine, ribociclib); (6) activators of p53 (nutilin 3a, FL118); (7) activators of protein kinase C (TPA/PMA, PEP005, PEP008); and (8) reactive oxygen species (ROS) inducers (hydrogen peroxide, tert-butyl hydroperoxide, phenyl-2-pyridyl ketoxime, phenylaminonaphthoquinones, paraquat).

The table highlights the fact that cancer cells can undergo cellular senescence in vitro just as well as their normal nontransformed counterparts. It is apparent that there is no senescence marker or pathway unique to normal or cancer cells. In most cases, increased SA-β-gal, morphological changes, and persistent DDR foci were recorded. SAHF were found in only a few cases (aphidicolin, etoposide, palbociclib, and epigenetic modifiers). SASP was also noted only in some cases; however, this is likely because SASP is not commonly analyzed as a senescence biomarker. Apparently, an implicit consensus was established that the demonstration of SA-β-gal, morphological changes, and persistent DDR is sufficient to document a cellular senescence-like state. It is notable that authors designated these phenotypes as a state of premature senescence or senescence-like cell cycle arrest, regardless of the set of biomarkers observed in each case.

Extremely prolonged drug exposure (from hours to days) was typically required to induce cellular senescence, as is evidenced by the table. In marginal situations, as in the case of aphidicolin-induced cell cycle arrest, the full set of senescence biomarkers (SA-β-gal, cell enlargement, SAHF, and DDR foci) was maintained, while the drug was present in the culture medium and lost upon drug removal (Meya-Mendoza et al., 2014). The requirement for prolonged incubation time was found for all groups of chemical compounds analyzed; however, the mechanism of senescence development appeared to differ among these groups. Whereas replication stress inducers, different DNA-damaging agents, and telomerase inhibitors likely generate a persistent DDR following prolonged introduction of a small number of DNA lesions or telomere uncapping, long-term incubation with epigenetic modifiers likely causes transcriptional activation of repressed loci (particularly INK4A, which encodes p16 CDK inhibitor). This hypothesis is supported by the fact that, in contrast to DNA damage-induced cellular senescence, which depends on p21 CDK inhibitor, epigenetically induced senescence is mostly dependent on p16. This characterizes epigenetically induced senescence as ‘causeless’—epigenetic modifiers directly activate molecular pathways maintaining the cellular senescence state without generating any cell stress. In this regard, senescence induced by epigenetic modifiers can resemble developmentally programmed or organismal aging-associated cellular senescence, while replication stress- and DNA damage-induced senescence are examples of stress-induced premature senescence states.

It follows from the table that replication stress- and DNA damage-induced cellular senescence mostly depend on the p53-p21 pathway. The same is basically true for cellular senescence induced by physical stressors such as ionizing radiation (IR) and ultraviolet (UV) (Latonen et al., 2001; Suzuki et al., 2006). It is well known that IR as well as UV can stimulate senescence in a variety of normal and cancer cell lines (Chainiaux et al., 2002; Meng et al., 2003; Jones et al., 2005; Jee et al., 2009). Mechanistically, this type of cellular senescence mostly depends on DNA damage induced by these stressors; this links IR and UV to chemical DNA-damaging agents. Moreover, IR and UV, along with most of the DNA-damaging agents presented in the table, induce apoptosis rather than senescence when used at higher doses. These observations further emphasize the relationship between apoptosis and senescence. Accordingly, these cell stress response pathways may operate either as alternatives or as supplement to each other. While prominent (but short term) DNA damage induces apoptosis, prolonged mild DNA damage activates cellular senescence. The p53 transcription factor emerges as a master regulator controlling these cell fate decisions (Purvis et al., 2012).

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Conflict of interest

None declared.

References

Abramova MV, Pospelova TV, Nikulenkov FP, Hollander CM, Fornace AJ Jr, Pospelov VA (2006) G1/S arrest induced by histone deacetylase inhibitor sodium butyrate in E1A + Ras-transformed cells is mediated through down-regulation of E2F activity and stabilization of beta-catenin. J. Biol. Chem. 281, 21040–21051.

Alsopp RC (1996) Models of initiation of replicative senescence by loss of telomeric DNA. Exp. Gerontol. 31, 235–243.
Amatori S, Bagaloni I, Viti D, Fanelli M (2011) Premature senescence induced by DNA demethylating agent (Decitabine) as therapeutic option for malignant pleural mesothelioma. Cancer Cell 17, 113–115.

An HM, Xue YF, Shen YL, Du Q, Hu B (2013) Sodium valproate induces cell senescence in human hepatocarcinoma cells. Molecules 18, 14935–14947.

Asohia K, Tsuji T, Nagai A (2003) Bleomycin induces cellular senescence in alveolar epithelial cells. Eur. Respir. J. 22, 436–443.

Asohia K, Tsuji T, Kameyama S, Itoh M, Semba S, Yamaguchi K, Nakamura H (2013) Senescence-associated secretory phenotype in a mouse model of bleomycin-induced lung injury. Exp. Toxicol. Pathol. 65, 1053–1062.

Baker DJ, Wijshake T, Tchkonia T, LeBrasseur NK, Childs BG, van de Sluis B, Kirkland JL, van Deursen JM (2011) Clearance of p16INK4a-positive senescent cells delays age-associated disorders. Nature 479, 232–236.

Baker DJ, Childs BG, Dunk M, Wijers ME, Sieben CJ, Zhong J, Saltness RA, An HM, Xue YF, Shen YL, Du Q, Hu B (2013) Sodium valproate induces senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.

Baker DJ, Childs BG, Rinaldi F, Liang W, Wang L, Vogt K, Goshki A, Petronilli V, Hening G, Hu B (2014) Small molecules that induce cellular senescence.
Frippiat C, Dewelle J, Remacle J, Toussaint O (2002) Signal transduction in H2O2-induced senescence-like phenotype in human diploid fibroblasts. Free Radic. Biol. Med. 33, 133–144.

Gorbunova V, Seluanov A, Pereira-Smith OM (2002) Expression of human telomerase (hTERT) does not prevent stress-induced senescence in normal human fibroblasts but protects the cells from stress-induced apoptosis and necrosis. J. Biol. Chem. 277, 38540–38549.

Grabowska W, Kucharewicz K, Wnuk M, Lewinska A, Suszek M, Przybylska D, Mosieniak G, Sikora E, Biełak-Zmijewska A (2015) Curcumin induces senescence of primary human cells building the vasculature in a DNA damage and ATM-independent manner. Age 37, 9744.

Han Z, Wei W, Dunaway S, Darnowski JW, Calabresi P, Sedivy J, Hendrickson EA, Balan KV, Pantazis P, Wyche JH (2002) Role of p21 in apoptosis and senescence of human colon cancer cells treated with camptothecin. J. Biol. Chem. 277, 17154–17160.

Harley CB, Fritsch AB, Greider CW (1990) Telomeres shorten during ageing of human cells. Nature 345, 458–460.

Hayflick L (1965) The limited in vitro lifetime of human diploid cell strains. Exp. Cell Res. 37, 614–636.

Hayflick L, Moorhead PS (1961) The serial cultivation of human diploid cell strains. Exp. Cell Res. 25, 585–621.

Hendrayer SF, Al-Khalaf HH, Aboussekhra A (2013) Curcumin triggers p16-dependent senescence in active breast cancer-associated fibroblasts and suppresses their paracrine procarcinogenic effects. Neoplasia 15, 631–640.

Hirose Y, Berger MS, Pieper RO (2001) p53 effects include both the duration of G2/M arrest and the fate of temozolomide-treated human glioblastoma cells. Cancer Res. 61, 1957–1963.

Hong SH, Hong B, Kim DC, Rho MS, Park J, Rha SH, Jun HS, Jeong JS (2004) Involvement of mitogen-activated protein kinases and p21Waf1 in hydroxyurea-induced G1 arrest and senescence of MCA-RH7777 rat hepatoma cell line. Exp. Mol. Med. 36, 493–498.

Hu W, Sung T, Jessen BA, Thibault S, Finkelstein MB, Khan NK, Sacaan AI (2015) Mechanistic investigation of bone marrow suppression associated with palbociclib and its differentiation from cytotoxic chemotherapies. Clin. Cancer Res. 22, 2000–2008.

Huang M, Whang P, Lewicki P, Mitchell BS (2011) Cyclopentenyl cytosine induces senescence in breast cancer cells through the nucleolar stress response and telomerase inhibitory activity. Br. J. Pharmacol. 167, 393–406.

Ido Y, Duranton A, Lan F, Caccione JM, Chen TC, Breton L, Ruderman NB (2012) Acute activation of AMP-activated protein kinase prevents H2O2-induced premature senescence in primary human keratinocytes. PLoS ONE 7, e35092.

Jackson JG, Pant V, Li Q, Chang LL, Quintas-Cardama A, Garza D, Tavana O, Yang P, Lakey R, Blaney SM, Lau CC (2005) Valproic acid induces growth arrest, apoptosis, and senescence in medulloblastomas by increasing histone hyperacetylation and regulating expression of p21Cip1, CDK4, and CMYC. Mol. Cancer Ther. 4, 1912–1922.

Lafay F, Belin Le, Marmottant P, Breton L, Ruderman NB (2012) Senescent breast cancer cells exhibit conserved features of senescence. Aging Cell 11, 2417–2426.

Latenon L, Taya Y, Laiho M (2001) UV-radiation induces dose-dependent regulation of p53 response and modulates p53-HDM2 interaction in human fibroblasts. Oncogene 20, 6784–6793.

Lee AC, Fenster BE, Ito H, Takeda K, Bae NS, Hirai T, Yu ZX, Ferrans VJ, Howard BH, Finkel T (1999) Ras proteins induce senescence by altering the intracellular levels of reactive oxygen species. J. Biol. Chem. 274, 7936–7940.

Leontieva OV, Blagosklonny MV (2013) CDK4/6-inhibiting drug substitutes for p21 and p16 in senescence: duration of cell cycle arrest and MTOR activity determine geroncrosis. Cell Cycle 12, 3063–3069.

Leontieva OV, Gustikov AV, Blagosklonny MV (2010) Weak p53 permits senescence during cell cycle arrest. Cell Cycle 9, 4323–4327.

Leontieva OV, Demidenko ZN, Blagosklonny MV (2013) MEK drives cyclin D1 hyperrepression during geroncrosis. Cell Death Diff. 20, 1241–1249.

Li XN, Shu Q, Su JM, Perlaky L, Blaney SM, Lau CC (2005) Valproic acid induces growth arrest, apoptosis, and senescence in medulloblastomas by increasing histone hyperacetylation and regulating expression of p21Cip1, CDK4, and CMYC. Mol. Cancer Ther. 4, 1912–1922.

Ling X, Xu C, Fan C, Zhong K, Li F, Wang X (2014) FL118 induces p53-dependent senescence in colorectal cancer cells by promoting degradation of MdmX. Cancer Res. 74, 7487–7497.

Linge A, Weinhold K, Blasche R, Kasper M, Barth K (2007) Downregulation of cavelin-1 affects bleomycin-induced growth arrest and cellular senescence in A549 cells. Int. J. Biochem. Cell Biol. 39, 1964–1974.

Litwiniec A, Grzanka A, Helmin-Basa A, Gackowska L, Grzanka D (2010) Features of senescence and cell death induced by doxorubicin in A549 cells: organization and level of selected cytoskeletal proteins. J. Cancer Res. Clin. Oncol. 136, 717–736.

Litwiniec A, Gackowska L, Helmin-Basa A, Zuryn A, Grzanka A (2013) Low-dose etoposide-treatment induces endoreplication and cell death accompanied by cytoskeletal alterations in A549 cells: does the response involve senescence? The possible role of vimentin. Cancer Cell Int. 13, 9.

Manfe V, Biskup E, Johnsen P, Kamstrup MR, Krejsgaard TF, Morling N, Wulf HC, Gniadecik R (2012) MDM2 inhibitor nutlin-3a induces apoptosis and senescence in cutaneous T-cell lymphoma: role of p53. J. Invest. Dermatol. 132, 1487–1496.

Mannava S, Moparthy KC, Wheeler LJ, Natarajan V, Zuckerman EL, Fink EE, Im M, Flanagan S, Burhans WC, Zeiouni NC, Shewach DS, Mathews CK, Nikiforov MA (2013) Deletion of deoxyribonucleotide pools is an endogenous source of DNA damage in cells undergoing oncogene-dependent senescence. Am. J. Pathol. 182, 145–152.

Mansilla S, Pina B, Portugal J (2003) Daunorubicin-induced variations in gene transcription, commitment to proliferation arrest, senescence and apoptosis. Biochem. J. 372, 763–771.

Marconnet CN, Sundar SN, Tseng M, Tin AS, Tran KQ, Mahuron KM, Bjeldanes LF, Firestone GL (2011) Indole-3-carbinol downregulation of telomerase gene expression requires the inhibition of estrogen receptor-alpha and Sp1 transcription factor interactions within the hTERT promoter and mediates the G1 cell cycle arrest of human breast cancer cells. Carcinogenesis 32, 1315–1323.

Marusyk A, Wheeler LJ, Mathews CK, DeGregori J (2007) p53 mediates senescence-like arrest induced by chronic replicational stress. Mol. Cell. Biol. 27, 5336–5351.

Mason SA, Cozi SJ, Pierce CJ, Pavey SJ, Parsons PG, Boyle GM (2010) The induction of senescence-like growth arrest by protein kinase C-activating diterpene esters in solid tumor cells. Invest. New Drugs 28, 575–586.

Masterson JC, O’Dea S (2007) 5-Bromo-2-deoxyuridine activates DNA damage signalling responses and induces a senescence-like phenotype in p16-null lung cancer cells. Anticancer Drugs 18, 1053–1068.

Maya-Mendoza A, Mercurt-May MJM, Bartkova J, Bartek J, Streuli CH, Jackson DA (2014) Immortalised breast epithelia survive prolonged DNA replication stress and return to cycle from a senescent-like state. Cell Death Dis. 5, e1351.

Mckenna E, Traganos F, Zhao H, Darynkiewicz Z (2012) Persistent DNA damage caused by low levels of mitomycin C induces irreversible cell senescence. Cell Cycle 11, 3132–3140.

Meng A, Wang Y, Van Zant G, Zhou D (2003) Induction of irradiation and busulfan induced premature senescence in murine bone marrow hematopoietic cells. Cancer Res. 63, 5414–5419.

Mihaiadi NM, Zhang XD, Allen J, Avery-Kiejda KA, Scott RJ, Hersey P (2007) Temozolomide induces senescence but not apoptosis in human melanoma cells. Br. J. Cancer 97, 1225–1233.

Minieri V, Savoizzi S, Gambartoga G, Lo Iacono M, Accomasso L, Cibario Rocchietti E, Gailina C, Turinettov C, Giachino C (2015) Persistent DNA damage-induced premature senescence alters the functional features of human bone marrow mesenchymal stem cells. J. Cell Mol. Med. 19, 734–743.
Small molecules that induce cellular senescence, N. V. Petrova et al.
Suzuki M, Suzuki K, Kodama S, Watanabe M (2006) Interstitial chromatin alteration causes persistent p53 activation involved in the radiation-induced senescence-like growth arrest. Biochem. Biophys. Res. Commun. 340, 145–150.

Suzuki E, Takahashi M, Oba S, Nishimura H (2013) Oncogene- and oxidative stress-induced cellular senescence shows distinct expression patterns of proinflammatory cytokines in vascular endothelial cells. ScientificWorldJournal 2013, 754735.

Taka T, Huang L, Wongnoppavich A, Tam-Chang SW, Lee TR, TunTWechapikul W (2013) Telomere shortening and cell senescence induced by perylene derivatives in AS49 human lung cancer cells. Biogor. Med. Chem. 21, 883–890.

Terao Y, Nishida J, Horuchi S, Rong F, Ueoka Y, Matsuda T, Kato H, Furugen Y, Yoshida K, Kato K, Wake N (2001) Sodium butyrate induces growth arrest and senescence-like phenotypes in gynecologic cancer cells. Int. J. Cancer 94, 257–267.

Toussaint O, Medrano EE, von Zglinicki T (2000) Cellular and molecular mechanisms of stress-induced premature senescence (SIPS) of human diploid fibroblasts and melanocytes. Exp. Gerontol. 35, 927–945.

Toussaint O, Royer V, Salmon M, Remacle J (2002) Stress-induced premature senescence and tissue ageing. Biochem. Pharmacol. 64, 1007–1009.

Veena MS, Wilken R, Zheng JY, Gholkar A, Venkatesan N, Vira D, Ahmed S, Basak SK, Dalgarg CL, Ravichandran S, Bara RK, Kasahara N, Elashoff D, Fishbein MC, Whitelegge JP, Torres JZ, Wang MB, Sivratan ES (2014) p16 Protein and gigaxonin are associated with the ubiquitination of NFRkap in cisplatin-induced senescence of cancer cells. J. Biol. Chem. 289, 34921–34937.

Velichko AK, Petrova NV, Razin SV, Kantidze OL (2015) Mechanism of heat stress-induced cellular senescence elucidates the exclusive vulnerability of early S-phase cells to mild genotoxic stress. Nucleic Acids Res. 43, 6309–6320.

Venturelli S, Berger A, Weiland T, Essman F, Waiden M, Nuebling T, Hacker S, Schenk M, Schulze-Osthoff K, Salif HR, Fulda S, Brows P, Johnstone RW, Lauer UM, Bitzer M (2013) Differential induction of apoptosis and senescence by the DNA methyltransferase inhibitors 5-aza-2’-deoxycytidine in human tumor cells. Mol. Pharmacol. 85, 645–6463.

von Zglinicki T, Pilger R, Sitte N (2000) Accumulation of single-strand breaks in cisplatin-induced senescence of cancer cells. J. Cell Biol. 12, 2226–2236.

Vogt M, Haggblom C, Yearyin J, Christiansen-Weber T, Haas M (1998) Independent induction of senescence by p16INK4a and p21CIP1 in spontaneously immortalized human fibroblasts. Cell Growth Differ. 9, 139–146.

Wang X, Wong SC, Salmon M, Remacle J (2002) Stress-induced premature senescence elucidates the exclusive vulnerability of early S-phase cells to mild genotoxic stress. Nucleic Acids Res. 43, 6309–6320.

Yeow EJ, Hwang YC, Kang CM, Kim IH, Kim DI, Parka JS, Choy HE, Park WY, Park SC (2000) Senescence-like changes induced by hydroxyurea in human diploid fibroblasts. Exp. Gerontol. 35, 553–571.

Yoshida A, Lee KE, Diehl JA (2016) Induction of therapeutic senescence in vemurafenib-resistant melanoma by extended Inhibition of CDK4/6. Cancer Res. 76, 2990–3002.

Yoshizaki K, Fujiki T, Tsunematsu T, Yamashita M, Udono M, Shirahata S, Takakura Y (2009) Pro-senescent effect of hydrogen peroxide on cancer cells and its possible application to tumor suppression. Biosci. Biotechnol. Biochem. 73, 311–315.

Young JI, Patel A, Rai P (2010) Suppression of thioredoxin-1 induces premature senescence in normal human fibroblasts. Biochem. Biophys. Res. Commun. 392, 363–368.

Yuan Y, Wang Q, Pauk J, Kubiecik S, Kemp MM, Adams DJ, Shami AF, Wagner BK, Schreiber SL (2012) A small-molecule probe of the histone methyltransferase G9a induces cellular senescence in pancreatic adenocarcinoma. ACS Chem. Biol. 7, 1152–1157.

Zdanov S, Debaq-Chainaux F, Remacle J, Toussaint O (2006) Identification of p38MAPK-dependent genes with changed transcript abundance in H2O2-induced premature senescence of IMR-90 hTERT human fibroblasts. FEBS Lett. 580, 645–6463.

Zhou H, Wink M (2013) The beta-carboline alkaloid harmine inhibits telomerase activity of MCF-7 cells by down-regulating hTERT mRNA expression accompanied by an accelerated senescent phenotype. PeerJ 1, e174.

Zhao W, Lin ZX, Zhang ZQ (2004) Cisplatin-induced premature senescence with concomitant reduction of gap junctions in human fibroblasts. Cell Res. 14, 60–66.

Zhao H, Halicca HD, Traganos F, Jorgensen E, Darzynkiewicz Z (2010) New biomarkers probing depth of cell senescence assessed by laser scanning cytometry. Cytometry A 77, 999–1007.

Zhou JM, Zhu XF, Lu YJ, Deng R, Huang ZS, Mei YP, Wang Y, Huang WL, Li ZC, Gu LQ, Zeng YX (2006) Senescence and telomere shortening induced by novel potent G-quadruplex interactive agents, quindoline derivatives, in human cancer cell lines. Oncogene 25, 833–841.

Zhou H, Huang B, Han Y, Jin R, Chen S (2013) Procolubal induces JAK2-STAT pathway activation and protects human glomerular mesangial cells from tert-butyl hydroperoxide induced premature senescence. Can. J. Physiol. Pharmacol. 91, 671–679.

Zhu F, Li Y, Zhang J, Piao C, Liu T, Li HH, Du J (2013) Senescent cardiac fibroblast is critical for cardiac fibrosis after myocardial infarction. PLoS ONE 8, e74535.

Zhu Y, Tchkonia T, Fuhrmann-Stroissnigg H, Dai HM, Ling YY, Stout MB, Pirtskhalava T, Giorgadze N, KB, Schreiber SL (2012) A small-molecule probe of the histone methyltransferase G9a induces cellular senescence in pancreatic adenocarcinoma. ACS Chem. Biol. 7, 1152–1157.

Zdanov S, Debaq-Chainaux F, Remacle J, Toussaint O (2006) Identification of p38MAPK-dependent genes with changed transcript abundance in H2O2-induced premature senescence of IMR-90 hTERT human fibroblasts. FEBS Lett. 580, 645–6463.

von Zglinicki T, Pilger R, Sitte N (2000) Accumulation of single-strand breaks is the major cause of telomere shortening in human fibroblasts. Free Radic. Biol. Med. 28, 64–74.

Zhang JW, Zhang SS, Song JR, Sun K, Zong C, Zhao QD, Liu WT, Li R, Wu MC, Wei LX (2014) Autophagy inhibition switches low-dose camptothecin-induced premature senescence to apoptosis in human colorectal cancer cells. Biochem. Pharmacol. 90, 265–275.

Zhao L, Wink M (2013) The beta-carboline alkaloid harmine inhibits telomerase activity of MCF-7 cells by down-regulating hTERT mRNA expression accompanied by an accelerated senescent phenotype. PeerJ 1, e174.

Zhou W, Lin ZX, Zhang ZQ (2004) Cisplatin-induced premature senescence with concomitant reduction of gap junctions in human fibroblasts. Cell Res. 14, 60–66.

Zhao H, Halicca HD, Traganos F, Jorgensen E, Darzynkiewicz Z (2010) New biomarkers probing depth of cell senescence assessed by laser scanning cytometry. Cytometry A 77, 999–1007.

Zhou JM, Zhu XF, Lu YJ, Deng R, Huang ZS, Mei YP, Wang Y, Huang WL, Li ZC, Gu LQ, Zeng YX (2006) Senescence and telomere shortening induced by novel potent G-quadruplex interactive agents, quindoline derivatives, in human cancer cell lines. Oncogene 25, 833–841.

Zhou H, Huang B, Han Y, Jin R, Chen S (2013) Procolubal induces JAK2-STAT pathway activation and protects human glomerular mesangial cells from tert-butyl hydroperoxide induced premature senescence. Can. J. Physiol. Pharmacol. 91, 671–679.

Zhu F, Li Y, Zhang J, Piao C, Liu T, Li HH, Du J (2013) Senescent cardiac fibroblast is critical for cardiac fibrosis after myocardial infarction. PLoS ONE 8, e74535.

Zhu Y, Tchkonia T, Fuhrmann-Stroissnigg H, Dai HM, Ling YY, Stout MB, Pirtskhalava T, Giorgadze N, Johnson KO, Stout MB, Mezera V, Giorgadze N, Jensen MD, LeBarreur NK, Kirkland JL (2015a) Targeting senescent cells enhances adipogenesis and metabolic function in old age. Cell. 160, 435.

Zhu Y, Tchkonia T, Fuhrmann-Stroissnigg H, Dai HM, Ling YY, Stout MB, Pirtskhalava T, Giorgadze N, Johnson KO, Stout MB, Mezera V, Giorgadze N, Jensen MD, LeBarreur NK, Kirkland JL (2015b) The Achilles’ heel of senescent cells: from transcription to senolytic drugs. Aging Cell 14, 644–658.