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Senescence-associated secretory phenotype and its impact on oral immune homeostasis

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The senescence-associated secretory phenotype (SASP), which accumulates over the course of normal aging and in age-related diseases, is a crucial driver of chronic inflammation and aging phenotypes. It is also responsible for the pathogenesis of multiple oral diseases. However, the pathogenic mechanism underlying SASP has not yet been fully elucidated. Here, relevant articles on SASP published over the last five years (2017–2022) were retrieved and used for bibliometric analysis, for the first time, to examine SASP composition. More than half of the relevant articles focus on various cytokines (27.5%), growth factors (20.9%), and proteases (20.9%). In addition, lipid metabolites (13.1%) and extracellular vesicles (6.5%) have received increasing attention over the past five years, and have been recognized as novel SASP categories. Based on this, we summarize the evidences demonstrating that SASP plays a pleiotropic role in oral immunity and propose a four-step hypothetical framework for the progression of SASP-related oral pathology—1) oral SASP development, 2) SASP-related oral pathological alterations, 3) pathological changes leading to oral immune homeostasis disruption, and 4) SASP-mediated immune dysregulation escalating oral disease. By targeting specific SASP factors, potential therapies can be developed to treat oral and age-related diseases.

KEYWORDS

oral homeostasis, senescence-associated secretory phenotype, cellular senescence, age-related disease, oral-systemic disease

1 Introduction

Oral immune homeostasis is a delicate balance established and shaped by the interaction between pathogen invasion and host immune response (1). Any disruption to this balance results in local or systemic diseases. Some pathophysiological changes are attributed to the environmental impact of senescent cells (2). The primary non-spontaneous effects of senescent cells appear to be closely linked to the senescence-associated secretory phenotype (SASP).

SASP, a product of senescent cells, is mainly classified into the following categories: 1) pro-inflammatory cytokines (such as interleukin (IL)-1α, IL-1β, IL-6, and IL-8); 2) chemokines (such as CXCL-1/3 and CXCL-10); 3) proteases: including matrix remodeling enzymes and plasminogen activators; 4) growth factors (such as VEGF, TGF-β and GM-CSF); 5) bioactive lipids (like oxidized lipid mediators); 6) extracellular vesicles (EVs); and 7) others (2, 3). SASP profiles exhibit a significant cell type-dependent heterogeneity, and SASP strength and composition are spatially and temporally dependent (4). In this paper, we summarize the different SASP categories, and reveal the most relevant cell types *via* bibliometric analysis, and propose a framework for the role of SASP in oral immune homeostasis to provide insights into the potential of SASP as a novel therapeutic target.

2 Methods

2.1 Data source and retrieval strategy

We searched the Web of Science Core Collection and PubMed databases for articles related to SASP factors published from 2017 to 2022. The retrieval strategy for the Web of Science Core Collection database was as follows: (((ALL=(cytokine) OR ALL=(chemokine) OR ALL=(protease) OR ALL=(growth factor) OR ALL=(lipid) OR ALL=(proinflammatory factor)) AND (ALL=(senescence associated secretory phenotype) OR ALL=(sasp))) AND (DOP==(2017-01-01:2022-04-01))) AND ((LA==("ENGLISH")) NOT (DT==("REVIEW"))). The retrieval strategy for the PubMed database was as follows: (((((((cytokine) OR (chemokine)) OR (protease)) OR (growth factor)) OR (proinflammatory factor)) OR (lipid) AND ((y_5[Filter]) AND (English[Filter]))) AND ((senescence associated secretory phenotype) OR (sasp) AND ((y_5[Filter]) AND (English[Filter])))) NOT review[PT].

Inclusion criteria were as follows: Research articles 1. related to SASP; 2. published between 2017-01-01 and 2022-04-01; and 3. written in English. Exclusion criteria were as follows: 1. Literature whose content is not closely related to SASP factors; 2. Studies including guidance, consensus, industry standards, interviews, comments, announcements, advertisements, or letters to the editor; and 3. informally published studies, such as graduate theses.

2.2 Data processing and analysis

After retrieval, data screening and quality control were performed. This was conducted by reading titles and abstracts to remove literature that met the exclusion criteria. The data from papers that met the inclusion criteria were downloaded and merged. After removing duplicates, CiteSpace 6.1.R1 was used for data analysis. Next, different words or phrases expressing the same meaning were merged. For example, nuclear factor-kappa b and nf kappa b were merged as NF-kappa B. To clearly demonstrate the relationships among different type of SASP factors clearly, we further merged the same type SASP factors. For example, NF-kappa B, CCN1, and cyclin d1 were merged as proinflammatory factors, and stem cells, mesenchymal stem cells, and cancer stem cells were merged as pluripotent stem cells. Finally, a keyword co-occurrence network was built to visualize the relationships among knowledge domains and identify important SASP factors that have attracted attention in recent years.

3 Results

3.1 Analysis of the proportion of reported SASP factors

In total, 564 articles were included in the bibliometric analysis. In the last 5 years, the most cited SASP factors have been cytokines, including IL-6, IL-1, IL-8, CXCL-8; tumor necrosis factors (TNFs); and interferons. These proinflammatory cytokines account for 27.5% of the reported SASP factors. Among them, interleukins were determined to be the most important SASP factors, accounting for nearly half of these proinflammatory cytokines. The second-most cited SASP factors are growth factors and proteases. These two types of SASP factors account for 20.9% of variations in the SASP factors. IGF-1, TGF-β, and VEGF are the most frequently cited growth factors. Matrix metalloproteinases (MMPs) are the most cited proteases. In the past five years, more than half of the articles related to SASP have focused on various cytokines, growth factors, and proteases (Figure 1A).

3.2 Analysis of relevant cell type

As shown in Figure 1B, a tight and complex network of interactions is formed among different types of SASP factors and between SASP factors and different types of cells. The main cell

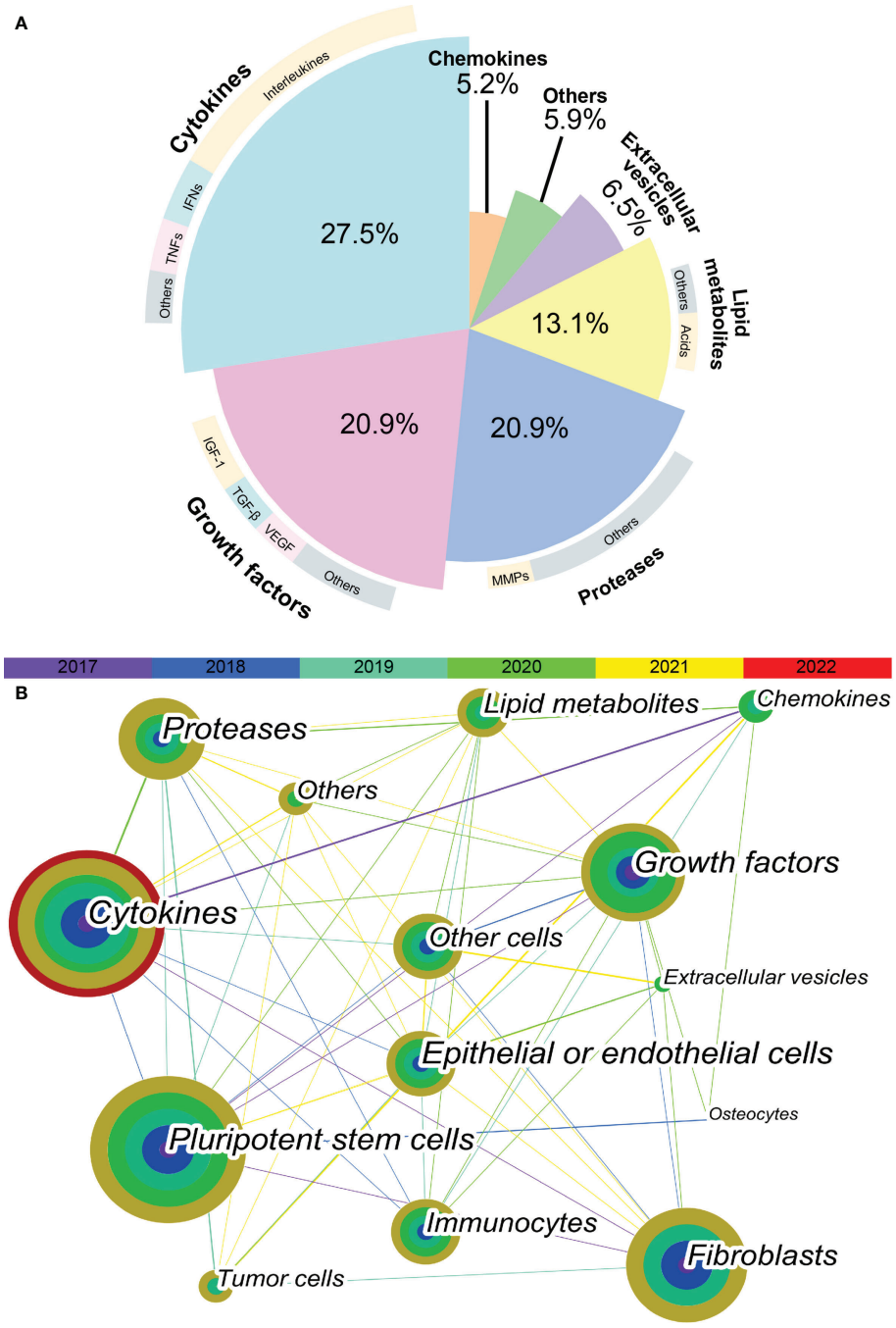

FIGURE 1
Keyword co-occurrence network visualization. **(A)** The proportion of different SASP factors in articles published in recent 5 years. Demonstrated as a pie plot. The inner are different types of SASP factors. The outer are most cited SASP factors of each type. **(B)** The keyword co-occurrence network shows the relationship between different types of SASP factors and different types of cells. Each node is a concentric circle, and the thickness of each layer of the concentric circle represents the frequency of use of this keyword in a certain year. The color of each line represents the year of the first co-occurrence between the two keywords. The thickness of each line represents the frequency of co-occurrence between the two keywords.

types of interest in recent studies of SASP were pluripotent stem cells, fibroblasts, epithelial or endothelial cells, and immunocytes. The cell types in which the different SASP factors mainly act are listed in Table 1. The results of bibliometric analysis demonstrated that different SASP factors tend to affect different cell types. For example, cytokines mainly act on pluripotent stem cells, whereas proteases mainly act on tumor cells. Pluripotent stem cells, fibroblasts, and epithelial or endothelial cells are often affected by SASP factors.

4 Discussion

4.1 Main components of SASP

4.1.1 Cytokines

The most prominent cytokines are members of the IL-1, IL-6, and TNF families. The membrane-binding IL-1α is an upstream regulator of age-related cytokine networks (5). The secreted IL-1β is excreted from cells in the early stages of the inflammatory process and then binds to the IL-1 receptor to trigger an inflammatory response (6). IL-6 initiates intracellular signaling by binding to its membrane-binding receptor, IL-6Rα, or its soluble receptor, sIL-6R (7). An enhanced TNF signaling is considered pertinent to immune system defects (8).

4.1.2 Chemokines

Chemokines act as local sensors of infection and inflammation (9). The most-studied chemokines in the field of aging in the past 5 years are the CXCL family members IL-8, CXCL-1, -2, and -3 and CCL family members like MCP-1, -2, and -4 and MIP-3α and-1α.

4.1.3 Growth factors

The diffusion of growth factors into the surrounding environment induces cell activation and proliferation, stimulates granulation tissue formation, regulates inflammatory responses, induces angiogenesis, and participates in matrix remodeling and re-epithelialization (10).

4.1.4 Extracellular proteases

Matrix metalloproteinases (MMPs) and tissue inhibitors of metalloproteinases (TIMPs) The MMP family is capable of degrading various components of extracellular matrix (ECM) proteins. TIMPs abrogate the proteolytic activity of MMPs by competing with them (11).

Serine proteases and their inhibitors Urokinase-type plasminogen activator (uPA) and tissue-type plasminogen activator (tPA) can modulate immune responses by activating MMPs to alter ECM composition, thus promoting the migration of macrophages and dendritic cells and modulating cytokine activity (12).

TABLE 1 Varies cell types where different SASP factors mainly act on.

SASP factors	Cell types	Rank
Cytokines	Pluripotent stem cells	1st
	Immunocytes	2nd
	Fibroblasts	3rd
Growth factors	Pluripotent stem cells	1st
	Fibroblast	2nd
	Other cells	3rd
Proteases	Tumor cells	1st
	Pluripotent stem cells	2nd
	Immunocytes	3rd
Lipid metabolites	Epithelial or endothelial cells	1st
	Other cells	2nd
	Immunocytes	3rd
Extracellular vesicles	Other cells	1st
	Epithelial or endothelial cells	2nd
	Fibroblasts	3rd
Chemokines	Tumor cells	1st
	Osteocytes	2nd
	Pluripotent stem cells	3rd
Others	Pluripotent stem cells	1st
	Tumor cells	2nd
	Fibroblasts	3rd

This table shows the top 3 cell types that are most strongly affected by each type of SASP.

Cathepsin An increased expression of cathepsin B and its nuclear translocation contribute to proinflammatory responses (13). Cathepsin D, an acidic protease active in intracellular protein breakdown, is significantly overexpressed during aging (14).

4.1.5 Lipid metabolites

Abnormal lipid accumulation induces pro-inflammatory genes activation and senescence phenotype (15). Ni et al. suggested that oxidized lipid mediators may serve as novel components of the SASP (16). Moreover, the levels of cyclooxygenase and its major product, prostaglandin E2 (PGE2), are increased in both replicative and premature senescence (17). Leukotriene D4 plays a role in cellular senescence (18).

4.1.6 Extracellular vesicles

EVs are small vesicles that contain proteins, lipids, and noncoding RNAs (19). An increased EV production is a common feature of senescence and senescent cells (20). Secreted EVs interact with or are internalized by recipient cells to transmit pro-senescence signals between cells and organs, and partially induce immune and inflammatory activation (21, 22).

4.1.7 Others

Additionally, the contributions of small molecules, such as ECM, miRNAs, and ROS, to SASP function remain understudied and may be considered an important future target.

4.2 Molecular mechanisms of SASP induction

Given the complexity and pleiotropic functionality of the SASP, we generalized the underlying mechanisms regulating it (Figure 2). The DNA damage response (DDR) is associated with SASP expression (23). The expression of some inflammatory SASP is regulated by NF-κB and C/EBPβ transcription cofactors by binding to SASP factor promoters. GATA-binding protein 4 transcription factor is responsible for upstream NF-κB signaling and, thus, regulates SASP factor expression (24).

Furthermore, many signaling pathways regulate SASP expression at the transcriptional level. For example, the Janus kinase signal transducer and activator of the transcription pathway participate in regulating SASP expression (25). Activation of p38 signaling also promotes SASP expression (26). More recently, the antiviral cyclic GMP-AMP synthase (cGAS)-stimulator of interferon genes (STING) pathway has been found to be important for SASP expression (27). Additionally, NOTCH signaling regulates the dynamic SASP transition (28).

SASP expression is transcriptionally regulated. mTOR pathway activation promotes the translation of SASP factors such as IL-1α. mTOR also stabilizes SASP mRNA transcripts by regulating MAPKAPK2 translation (29). Inflammasomes are key mediators of SASP induction; inflammasomes upstream of caspase-1 can activate the IL-1 inflammatory cascade during senescence (30).

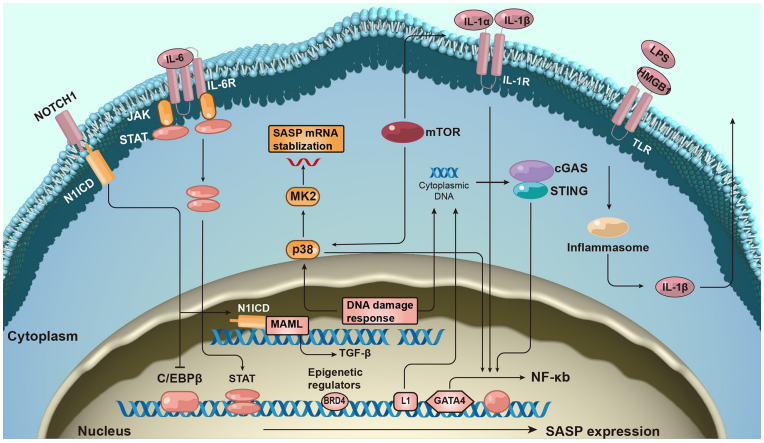

FIGURE 2
The molecular mechanism of senescence-associated secretory phenotype production. The formation of senescence-associated secretory phenotype (SASP) networks in cells is regulated by a complex molecular mechanism. The DNA damage response is related to senescence activation and SASP expression. SASP protein expression is regulated at both the transcription and post-transcriptional levels. Additionally, epigenetic changes regulate SASP gene expression.

The expression of SASP also correlates with epigenetic changes. Recruitment of chromatin reader bromodomain-containing protein 4 (BRD4) leads to the remodeling of super-enhancer elements adjacent to SASP genes (31). Several ncRNAs can also affect SASP production at transcriptional and post-translational levels.

4.3 Source of oral SASP

4.3.1 Oral senescent cells with locally secreted SASP

Dental tissue-derived cells Long-term stress on teeth may induce the human dental pulp cells senescence and up-regulation of SASP factors in human dental pulp cells (32). In aging patients, dental pulp stem cells exhibit elevated expression of SASP factors (33). Dental follicle cells are positive for SA-β-gal staining in later stages of cell culture (34). Dental follicle stem cells secrete TGF-β3, TSP-1, and TGF-β2 to promote and relieve inflammation (35–37).

Periodontal-derived cells Senescent periodontal ligament cells express high levels of MMP2 (38). The induction of senescent periodontal ligament stem cell by TGF-β is accompanied by increased levels of certain SASP factors (39). *In vitro* experiments have shown that p16, p21, IL-6, and IL-8 mRNA expression in human gingival fibroblasts is upregulated after replicative senescence (40–42). Researchers have observed that liposaccharide (LPS) exposure causes osteocyte senescence and SASP expression by activating p53 (43).

Oral mucosa cells SASP secretion is significantly increased in senescent human oral keratinocytes (44–47), and IL-6, TNF-α, and IFN-γ levels are increased in the oral tongue tissues of the elderly (48). Tongue muscle stem cells and epithelial cells have been shown to degenerate with age, but the relevant SASP profile has not been tested (49, 50).

Cancer cells In head and neck squamous cell carcinoma, the number of SA-β-gal-positive aged cells and SASP factor levels are significantly increased after LY2835219 treatment (51). Moreover, an increased secretion of SASP has been observed in senescent cancer-associated fibroblasts (CAF). The senescent CAFs co-cultured with oral squamous cell carcinoma (OSCC) cells also exhibit higher levels of IL-6 and CXCL1 (52). In precancerous lesions, senescent oral submucosal fibroblasts accumulate and upregulate MMPs (53).

Immunocytes High-glucose induces macrophage senescence and increases IL-1, IL-6, TNF-α, MMP-2, and MMP-8 secretion (54). Periodontal pathogens induce monocyte activation and the up-regulation of multiple cytokines (55). Overactive neutrophils can release inflammatory molecules and MMPs (56). The SASP profile of B cells and plasma cells in the aging gingival tissue changes (57). The SASP profile varies with cell type; factors inducing senescence are shown in Table 2.

4.3.2 ARDs with increased circulating SASP

Circulating SASP is associated with aging and age-related diseases (ARDs) (64). Compared to that in young individuals, the proportion of senescent cells is increased in aging individuals; additionally, the levels of some SASP proteins (age-related SASP) increase significantly (65). Concurrently, the premature cells induced by ARDs can accelerate this process (ARD-related SASP) (66). Age-related versus ARD-related SASP and their effects on oral health are shown in Table 3.

4.4 Pleiotropic effects of SASP on oral immune homeostasis

The heterogeneity of SASP may partly account for its pleiotropic effects. Based on the mechanisms of SASP, we propose a four-step hypothetical framework by which oral disease progresses from the pathologic role of SASP to the destabilization of oral immune homeostasis. The pleiotropic effects of the SASP can be interpreted using the model depicted in Figure 3.

4.4.1 Step 1: Induction of cellular senescence in oral microenvironment

Replicative- and stress-induced senescence are the main patterns of cellular senescence in the oral microenvironment.

4.4.1.1 Replicative senescence

Serial cultivation of human diploid cells leads to indefinite cell division, which is currently defined as replicative senescence (72). Senescent cells arising from this physiological phenomenon are defined as primary senescent cells (73), which have a series of typical morphologies and biomarker alterations, including DDR (γ-H2AX and p53), cell cycle arrest ($p16^{INK4A}$ and $p21^{CDKN1A}$), anti-apoptotic genes (BCL-proteins), lysosomal content (SA-β-gal), and heterochromatin markers (H3K9me3 and HP1γ) (2, 74).

4.4.1.2 Stress-induced senescence

Due to various stressors, the stress in senescing cells can be classified as secondary senescent cells as follows: 1) DNA damage-induced senescence, which can lead to cellular senescence by inducing DNA damage (35, 75); 2) chemotherapy-induced senescence, in which chemotherapy and anti-resorptive agents have been shown to induce senescence in oral cells (76); 3) oxidative stress–induced senescence whereby H_2O_2 treatment increases the positive rate of SA-β-gal staining in human dental pulp cells; 4) oncogene-induced senescence wherein senescence markers are upregulated in oral premalignant lesions (77). 5) epigenetically induced senescence, which is characterized by the blockade of DNA

TABLE 2 Cellular senescence and SASP involved in the oral cavity.

Cell type		Senescence trigger	SASP factors involved	Ref.
Dental-derived cells	Human dental pulp cells	H_2O_2-induced	ICAM-1, VCAM-1, PPAR-g	(58)
	Human dental pulp stem cells	*p*-cresol-induced	IL-6	(33)
	Dental follicle stem cells	LPS-induced	TGF-β2, IL-6, IL-8, IL-1β	(37)
Periodontal -derived cells	Human periodontal ligament fibroblasts	Replicative and radiation-induced	MMP2	(38)
	human periodontal ligament stem cell	TGF-β-induced	IL-8, IL-18, IL-6	(39)
	Human gingival fibroblast	Replicative	IL-6, IL-8	(40)
		Replicative	IL-6, IL-8, TNF, TIMP-1	(41)
		Replicative	MMP3, MMP12, IL-1α	(42)
	Alveolar osteocyte	LPS-induced	ICAM-1, IL-1β, IL-6, IL-8, MCP-1, MMP12, MMP13	(43)
Oral mucosa cells	Human oral keratinocytes	Bisphosphonates-induced	IL-8, IL-6, MMP3	(59)
		Replicative	IL-1β, IL-1α, IL-8, IL-6	(46)
		Replicative	IL-1β, MMP3, PGF, CTGF, VEGF, MMP1, TIMP2, IL-8, MMP9,	(44)
		Replicative	IL-1β, IL-1α, IL-8, IL-6, TNF-α, G-CSF, GM-CSF, GROα	(47)
		High glucose-induced	IL-1β, IL-6, TNF-α	(60)
Cancer cells	Cal27, HSC3 and HSC6 cell lines	LY2835219-induced	IL6, IL8, MCP1, CXCL1, CXCL2, CXCL3	(51)
	CAF from OSCC	Cisplatin-induced	MCP-1, IL-6	(61)
		H_2O_2-induced	TGF-β, MMP2	(62)
		Co-culture with OSCC cells	IL-6, CXCL1	(52)
		The progression of oral submucous fibrosis	MMP1, MMP2	(53)
	Oral submucous fibroblasts	High glucose-induced	IL-1, IL-6, TNF-α, MMP-2, and MMP-8	(54)
Immunocytes	Macrophage	*Pg* and *Aa*-induced	IL-1β, TNF-α, IL-6, IL-23	(55)
	Monocytes	LPS, *Pg*, *Aa* and zymosan A-induced	IL-8, MMP-9	(56)
	Neutrophils	Replicative And Periodontal pathogens-induced	MMP2, MMP9, CTSK, TNF-α	(57)
	B cells/plasmacytes	*Pg*-induced	IL-17A, IFN-γ	(63)

CAF, cancer-associated fibroblast; OSCC, oral squamous cell carcinoma; LPS, lipopolysaccharide; ICAM, intercellular adhesion molecule; VCAM, vascular cell adhesion molecule; PPAR, peroxisome proliferator-activated receptor; IL, interleukin; MCP, monocyte chemoattractant protein; MMP, matrix metalloproteinase; TNF, tumor necrosis factor; PGF, placental growth factor; CTGF, connective tissue growth factor; VEGF, vascular endothelial-derived growth factor; TIMP, tissue inhibitor of metalloproteinases; G-CSF, granulocyte colony-stimulating factor; GM-CSF, granulocyte-macrophage colony-stimulating factor; GROα(CXCL1), C-X-C motif chemokine ligand 1; Pg, Porphyromonas gingivalis; Aa, Aggregatibacter actinomycetemcomitans.

TABLE 3 Age-related versus ARD-related circulating SASP and effects on oral health.

Age-related or ARD-related		Study type	Circulating increased SASP factors	Effects on oral health
Age-related		Cross-sectional study (65)	IL-6, TNF-α, CCL3, CCL4, GDF15, ACTIVIN A, TNFR1, FAS	1. Recruitment of immune cells: chemokines (CCL3, CCL4) 2. Matrix remodeling: MMPs (MMP9) 3. Fibrosis: growth factors (TGF-β) 4. Senescence re-enforcement: cytokines (IL-6, TNF-α, IL-10)
ARDs	Diabetes	Case-control study (67)	IL-6	
		Cohort study (68)	IL-6, TNF-α	
	Cancer	Systematic review (69)	IL-6, TGF-β, IL-10	
	Cardiovascular disease	Case-control study (70)	MMP9	
		Systematic review (71)	IL-6	

ARD, age-related disease; SASP, senescence-associated secretory phenotype; GDF, Growth/differentiation factor; TNFR, tumor necrosis factor receptor; CCL, CC chemokine ligand; TNF, tumor necrosis factor; TGF, transforming growth factor; MMP, matrix metalloproteinase.

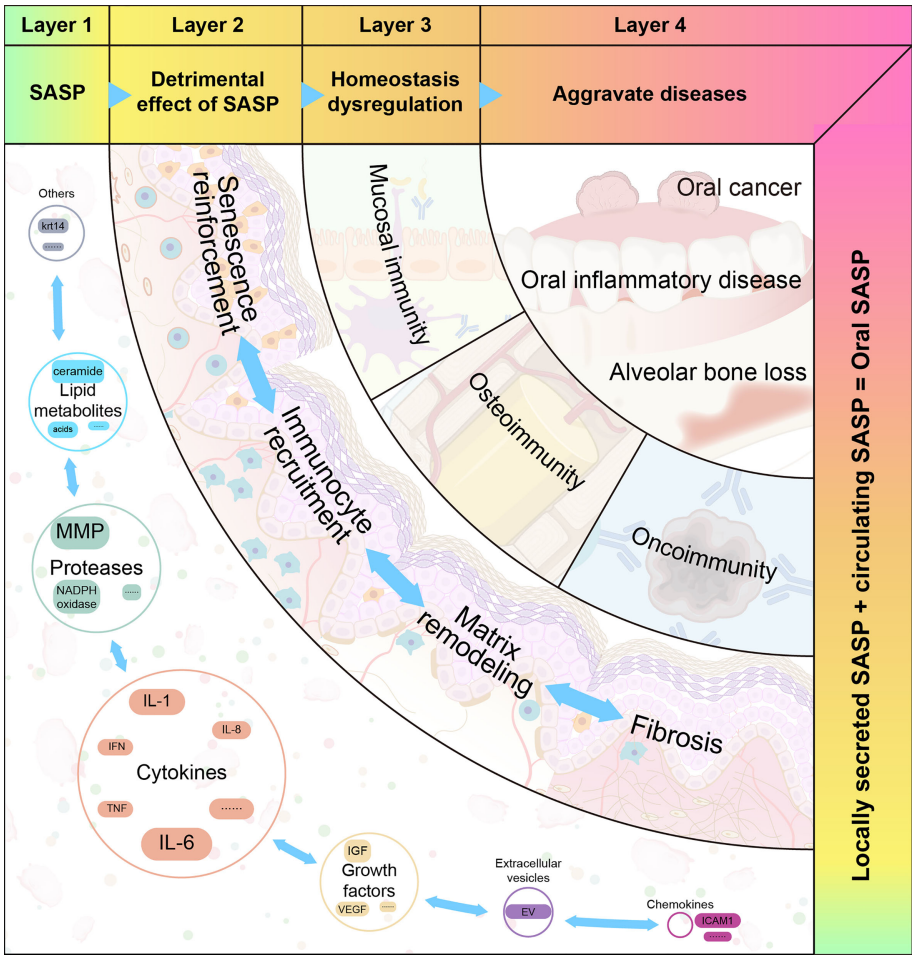

FIGURE 3
The four proposed layers of how senescence-associated secretory phenotype impacts oral diseases. Senescence-associated secretory phenotype (SASP) (including cytokines, chemokines, growth factors, and proteases) secreted by oral senescent cells, as well as circulating SASP, constitute the aging microenvironment of oral cavity. As an important mediator, SASP accelerates oral pathological alterations including senescence re-enforcement, recruitment of immune cells, matrix remodeling and fibrosis. Then, the SASP-induced dysregulation of immune homeostasis can be divided into three categories: mucosal immunity, bone immunity, and tumor immunity. These destroy the structure and function of different oral tissues. When age-related tissue damage accumulates, it manifests as age-related diseases.

Methyltransferase 1 (DNMT1) and activation of histone acetylation in oral cells (78); and 6) paracrine senescence wherein the SASP produced by primary senescent cells initiates senescence in surrounding cells.

4.4.1.3 Distinguishing senescence and inflammation

The inflammatory cytokines secreted by activated immune cells overlap with SASP factors. Some cytokines are unique to inflammation (such as IL-22), while others are unique to senescence (such as TIMP). By definition, SASP is downstream of cellular senescence. Notably, the senescence process is generally accompanied by sterile, chronic, low-level inflammation, termed inflamm-aging (79). Chronic inflammation may occur due to age-related immune dysregulation or decreased resistance to challenges, which can induce tissue pathology. The frameworks for aging, inflammation, and cellular senescence are shown in Figure 4.

4.4.2 Step 2: SASP accelerates oral pathologic alterations

The SASP is beneficial for maintaining homeostasis and regeneration at a moderate level. However, when SASP is expressed continuously, it induces pathological alterations and disrupts the immune homeostasis of the oral microenvironment.

4.4.2.1 Amplifying the immune cascade

Secreted SASP activates proximal and distant immunocytes in autocrine and paracrine manners. IL-1β is known to induce

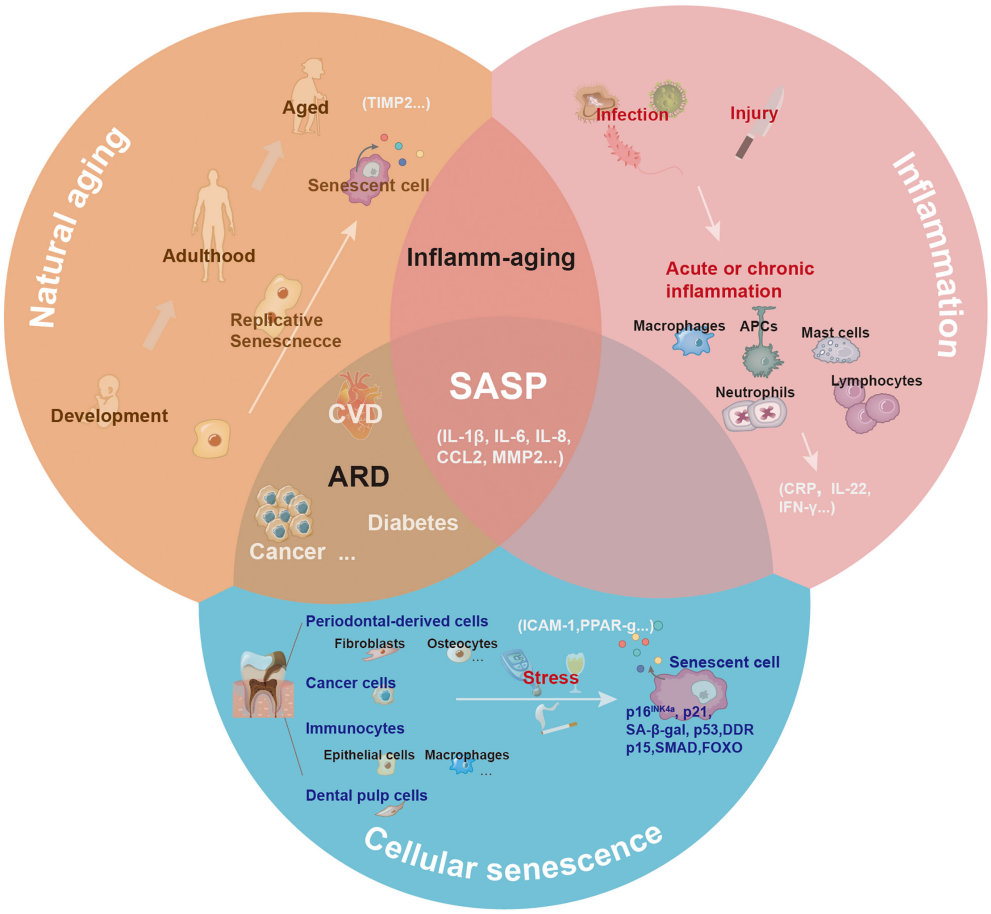

FIGURE 4
The central role of senescence-associated secretory phenotype in natural aging, inflammation, and cellular senescence. With natural aging, there is a progressive loss in tissue and organ functions, and accumulated senescence-associated secretory phenotype (SASP) can contribute to this process. Depending on the triggers, including infection or injury, the inflammatory response has different pathological consequences. The inflammatory response can be amplified *via* the secretion of inflammatory cytokines. Various cells (such as epithelial cells, dental pulp cells, fibroblasts, and macrophages) in the oral cavity undergoing senescence release SASP to the local microenvironment under the induction of natural aging and stress.

$CD4^+$ T cell proliferation in response to challenges associated with cognate antigens (80). IL-8 induces the migration of activated immune cells to gingival tissue and promotes tissue remodeling and angiogenesis (81). Elevated CCL2 and CCL4 levels are responsible for macrophage recruitment in periodontal lesions (82). The CXCL1 secreted by tumor cells promotes tumor growth by recruiting tumor-associated neutrophils (83).

4.4.2.2 Supporting senescence reinforcement

Certain key SASP factors, such as IL-6, IL-8, GROα, and IGFBP-7, act in an autocrine feedback loop. Non-senescent human oral epithelial keratinocytes cultured with senescent cell supernatants exhibit increased SA-β-gal activity and SASP expression (84). The IL-1β expressed by tumor cells can significantly increase CXCL1 production in CAFs *via* paracrine signaling (85). Moreover, IL-1β induces significant IL-6 production in human gingival fibroblasts and promotes cellular responsiveness to IL-6 through an autocrine loop (86). IL-6 induces normal fibroblast senescence by establishing a senescence induction circuit (87).

4.4.2.3 Remodeling the extracellular matrix

MMPs constitute an important proteolytic pathway that affects tissue remodeling and ECM structure. MMP expression reduces the ability of tissues to maintain homeostasis (88). Specifically, MMP-1 destroys the periodontal connective tissue by directly degrading collagen or activating the fibrinolytic protease cascade, leading to tooth loss (89). The proinflammatory factors IL-1β and IL-6 aggravate tissue destruction by increasing MMP-1 in periodontal tissue (90, 91). Furthermore, TNF-α is important for osteoclast formation

and bone resorption in mice and suppresses ECM protein expression (92, 93).

4.4.2.4 Promoting fibrosis

TGF-β is the primary factor driving fibrosis. TGF-β activation in epithelial cells can interact with fibroblast cells and induce the expression of other profibrotic cytokines (e.g., endothelin and CTGF) (94). Areca nut alkaloids induce senescence in oral fibroblasts and TGF-β production, which is favorable for the development of oral submucosal fibrosis (95). TIMP-1 and -2 have also been proven to be early indicators of oral submucous fibrosis and aging (96). Additionally, the MMP-1 and MMP-3 secreted by senescent cells during oral submucosal fibrosis have been shown to promote fibrosis in the advanced stages (97).

4.4.3 Step 3: SASP disrupts oral immune homeostasis

4.4.3.1 Effects on mucosal immunity

SASP challenges mucosal epithelial homeostasis by undermining the physical barrier. MMP-2 cleaves cell–cell adhesion molecules, thus disrupting epithelial adhesion (62). An increased MMP-1 expression in inflamed tissues directly leads to the degradation of collagen, thereby causing tissue destruction (89). TNF modulates the apoptosis of epithelial cells and fibroblasts and suppresses ECM proteins. These results indicate that TNF—in the senescence process—can damage the epithelial barrier (93). Some SASP factors participate in the recruitment and activation of immune cells. IL-1β greatly induces the proliferation and activation of Th1 and Th2 cells (80). IL-8 regulates neutrophil activation and migration in inflamed tissues (98). Additionally, IL-6 significantly increases the production of VEGF, bFGF, and cathepsin B in human gingival fibroblasts and synergistically induces angiogenesis in periodontitis lesions (86, 99). Senescent macrophages in the gingiva contribute to SASP release and inflammatory response, which indicates that senescence may also play an important role.

4.4.3.2 Effect on osteoimmunity

The secretome produced *via* innate host responses facilitates communication between immune cells and bone cells. Senescent immune cells regulate bone homeostasis through immune mediators that involve the SASP. For instance, IL-17, IL-1, and IL-6, as well as low levels of IFN-γ secreted by Th17 cells, promote osteoclastogenesis (100, 101). TNF-α has also been shown to strengthen osteoclastogenesis by synergizing with RANKL (102). In contrast, the bone-senescent microenvironment further enhances alveolar bone ageing. SASP factors released extracellularly from osteocytes accelerate the senescence of bone marrow (BM) (103). Selected SASP markers secreted by senescent osteocytes from alveolar bones promote inflammation and alveolar bone loss (43). In senescent fibroblasts, IL-1β increases the production of chemokines, including PGE2, an important chemical mediator of alveolar bone resorption (104). Additionally, senescent osteocytes develop a unique SASP signature composed of upregulated MMPs (105). MMPs can degrade ECM proteins, including sulfated proteoglycans, collagen, and fibronectin, in cartilage. Moreover, insulin-like growth factor-binding protein 4 (IGFBP-4) are upregulated in senescent osteocytes and myeloid cells, leading to deficiency in bone formation (22, 106).

4.4.3.3 Effect on oncoimmunity

Senescent cells in the tumor microenvironment (TME) may play roles in tumor progression and metastasis. CAFs are the most prominent stromal cells in TME. CAFs are senescent cells that actively communicate with other cells in the TME by secreting the SASP. TGF-β levels are upregulated by senescent oral CAFs and synergize with MMP-2 to reduce the expression of cell adhesion molecules and promote epithelial invasion (107). CAFs also modulate the epithelial-mesenchymal transition (EMT) by secreting TGF-β (108). Moreover, activated CAFs secrete proinflammatory factors that recruit and activate infiltrating immune cells (IICs). IICs provide mitogenic growth factors that stimulate the proliferation of tumor cells and other nearby stromal cells (109). IICs also express multiple proteolytic enzymes that selectively modify ECM structure and composition (110). Additionally, Park et al. proposed that the serum levels of IL-6 may be a serum biomarker for OSCC diagnosis (111). IL-6 promotes the invasion of cancer cells through the epithelial–mesenchymal transition (112). However, the immune cell subtype and its mechanism in the TME require further elucidation; further research is necessary to determine the specific roles of these factors in oral cancer.

4.4.4 Step 4: Aging and SASP in oral diseases

4.4.4.1 SASP in oral inflammatory disease

The SASP may be responsible for chronic oral inflammation, as it disrupts mucosal homeostasis through matrix degradation, senescence reinforcement, and immune cell recruitment. Compared to their young counterparts, old mice suffer frequent spontaneous periodontitis, and the expression of IL-1β and TNF-α in the gingiva is significantly elevated (113). Increased levels of IL-6 and MMP-8 have been observed in the saliva of patients with chronic periodontitis (114). Enhanced senescence and increased SASP are observed after ligation and *P. gingivalis* infection–induced periodontitis *in vivo* (115, 116). Additionally, hyperglycemia can increase the burden of senescence in the gingival tissue (54). Senescent cells accumulate in aged and diseased oral tissues, and this accumulation is associated with severe tissue destruction.

4.4.4.2 SASP in alveolar bone loss

Bone integrity and quality undergo differential changes in various oral diseases (117). Animal studies have shown that aging is positively correlated with alveolar bone loss. Old mice have poorer alveolar bone quality, lower alveolar bone crest height, and more active bone resorption (118). Senescence-associated distension of satellites (an early and consistent marker of senescence) and p16 mRNA expression are increased in old alveolar bone samples (119). Moreover, senescent osteocytes show changes in cell phenotype and diminished osteocyte density during age-related skeletal changes. This may further damage the mechanical conduction, impair nutrient access, influence signal transduction, and ultimately result in significant bone loss (120). Senescent bone cells exacerbate chronic inflammation through SASP accumulation, leading to deterioration of the periodontal environment (119). The SASP factor secreted by LPS-induced senescent osteocytes promotes the proliferation of some oral pathogens. These pathogens produce more LPS, thereby exacerbating the senescence of alveolar osteocytes and resulting in alveolar bone loss (43).

4.4.4.3 SASP in oral cancer

Cell senescence occurs throughout life and plays dual roles in modulating the progression and suppression of oral cancers (121). The number of SA-β-Gal-positive cells is higher in OSCC specimens than in tumor-free marginal tissues (52). Senescent fibroblasts also accumulate in precancerous lesions *in vivo* (53). Senescent cells secrete many SASP factors into the TME, which may support cell proliferation, EMT, and angiogenesis, thereby promoting tumor growth and invasion. MMP-1, -2, -10, and-12 levels in the saliva of OSCC patients increase significantly (122). In OSCC, the expression of MMP-11 is associated with an increased lymph node metastasis and a low survival rate (123). MMP-7 is mainly expressed in the invasive portion of oral cancer, whereas MMP-8 and MMP-9 are mainly detected in peritumoral inflammatory cells (124). This evidence suggests that senescent cells and the SASP are key factors in the onset and progression of oral cancer.

5 Concluding remarks

SASP, derived from senescent cells, includes secreted factors that may alter the extracellular environment (proteases), mediators that transmit and amplify senescence signals (cytokines, chemokines, bioactive lipids, and EVs), and proteins that influence cancer behavior (growth factors). The composition of SASP in the oral environment consists of two parts: local SASP and circulating SASP. Local SASP is secreted by oral senescent cells undergoing primary or secondary senescent patterns while the circulating SASP is closely associated with chronological age and ARD. As an important bridge for intercellular communication, the SASP communicates with different immune cells and is the key to securing oral homeostasis. Conversely, the SASP-induced dysregulation of immune homeostasis leads to intrinsically complex phenotypes in oral pathology. A better understanding of the relationship between SASP and the immune system is necessary for developing therapies to prevent or treat various ARDs in the oral cavity.

Author contributions

ZY and LN contributed to design, drafted and revised the manuscript; PZ contributed to design, and critically revised the manuscript; NJ and GL. assisted data analysis. QW, contributed to conception, design, and critically revised the manuscript. All authors gave final approval and agree to be accountable for all aspects of the work.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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