S100 proteins in head and neck squamous cell carcinoma (Review)

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Abstract. The most common tumor affecting the head and neck is head and neck squamous cell carcinoma (HNSCC). The characteristics of HNSCC include a rapid onset, a lack of early diagnosis, drug resistance, relapse and systemic adverse effects, leading to inadequate prevention, diagnosis and treatment. Notably, previous research suggests that there is an association between S100 proteins and HNSCC. S100A8, S100A9 and S100A14 interfere with tumor cell proliferation by blocking the cell cycle. The present review discusses this association. S100A4 enhances cancer stem cell properties, and interacts with actin and tropomyosin to promote tumor cell migration. S100A1, S100A8, S100A9, S100A10, S100A14 and S100P are involved in the initiation and progression of HNSCC via Hippo, nuclear factor κB, phosphatidylinositol kinase/protein kinase B/mammalian target of rapamycin and other signaling pathways. In addition, certain long non-coding RNAs and microRNAs are involved in regulating the expression of S100 proteins in HNSCC. Reducing the expression of certain members of the S100 protein family may enhance the chemosensitivity of HNSCC. Collectively, it is suggested that S100 proteins may function as markers and targets for the prevention, diagnosis and treatment of HNSCC.

Contents

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1. Introduction

The S100 protein family is the largest innate immune protein family containing EF-hand structural domains in vertebrates (1,2). As damage-associated molecular patterns, the expression of S100 proteins is increased to exert anti-infective effects following bacterial infection in epithelial cells (3,4). However, the overexpression of S100 protein family members leads to the secretion of numerous cytokines, chemokines or growth factors, such as tumor necrosis factor-α, C-C motif chemokine ligand (CCL)-2, CCL-20, C-X-C motif chemokine ligand 10, and interleukin (IL)-1, IL-6, IL-8, IL-10 and IL-12 (5-8). These factors cause tissue damage (9), and attract and activate immunosuppressive cells to alter the tumor microenvironment, promoting tumor growth and metastasis (10). The structure of S100 proteins is highly conserved in vertebrates (11), and is comprised of three exons. Thus, family members exhibit a number of similarities in both sequence and conformation (5); they all have hydrophobic amino acid domains in the N- and C-terminal regions (11). Notably, the first exon following the initiation codon of each member is unique, and each protein possesses specific functions. To date, ~25 family members have been identified (2,12-15). Among these, the S100A subfamily (*S100A1-S100A18*) is clustered in the epidermal differentiation complex region of 1q21; *S100B* is located on 21q22, *S100G* is located on Xp22, *S100P* is located on 4p16 and *S100Z* is located on 5q13 (2,5,12,16,17). While *S100S*, *S100T* and *S100U* are all orthologs in fish, no corresponding gene locus has been located in humans. The majority of the aforementioned family members exist as low molecular weight dimers of 9-14 kDa (5). In addition, some proteins form tetramers, while S100G exists as a monomer (12,13). The expression of S100 proteins is tissue-specific (12,16-18). S100A1 and S100A3 are predominantly expressed in heart and hair cuticle cells, respectively (12,16). S100A6 is preferentially expressed in fibroblasts (16). S100A8 and S100A9 are constitutively expressed in myeloid cells (16,17). S100B is normally expressed in the brain and can be used as a marker for brain injury (18). However, tumor progression can alter their expression and disrupt these specific expression patterns. The expression levels of S100A2, S100A4, S100A6,

S100A10, S100A14 and S100A16 are high in pancreatic ductal adenocarcinoma (19). The overexpression of S100A8, S100A9, S100A11 and S100P has been shown to be associated with a worse prognosis of patients with breast cancer (16); however, the expression of S100A1, S100A3, S100A5, S100A6, S100A13 and S100G is associated with improved outcomes of patients with ovarian cancer (17); these findings suggest that the S100 proteins play complex roles in the development of cancer.

The most common types of head and neck squamous cell carcinoma (HNSCC) are oral squamous cell carcinoma (OSCC), hypopharyngeal squamous cell carcinoma (HSCC), nasopharyngeal squamous cell carcinoma (NSCC) and papillary thyroid carcinoma (PTC) (20). Unlike other tumors, HNSCC has a complex anatomical structure at its site of origin, where numerous muscles, bones, blood vessels and nerves congregate, which protect the brain and participate in respiration, metabolism and other physiological processes (21). Due to the unique nature of the head and neck region, surgical resection may impede the ability of the patient to speak and swallow, which would significantly decrease their quality of life (22). Oncolytic virus therapy (23), chimeric antigen receptor T-cells (24), immune checkpoint inhibitor therapy (25) and other cutting-edge immunotherapy approaches, such as peptide vaccines, mRNA vaccines and adoptive cell therapy using T-cell receptor-engineered T cells, have made a significant impact in HNSCC recently (26-28). However, the use of these immunotherapy approaches is associated with high costs, and the treatment efficacy can be influenced by multiple factors of the tumor microenvironment (25); thus, the disease still has a 5-year overall survival rate of <50% (29). Currently, there is an urgent need for HNSCC prevention, early diagnosis and therapy. The results of a previous study demonstrated that HNSCC is closely associated with hypercalcemia (30). However, numerous members of the S100 protein family are calcium receptors, and the structure and function of their encoded proteins are regulated by Ca^{2+} (2). When Ca^{2+} is injected through voltage-gated or receptor-mediated channels, the conformation and hydrophobicity of S100 proteins are changed via converting or binding the signal of Ca^{2+} (2,14). This leads to interaction with the hydrophobic structural domains of target proteins, resulting in cell cycle dysregulation and apoptosis (2,14). In addition, S100 proteins are involved in the formation of the squamous epithelial keratinized envelope (31), which forms a keratinized layer to protect the skin barrier. Notably, the head and neck are covered by numerous squamous epithelial cells. Some studies have indicated that S100 proteins and HNSCC may be closely associated (31-34). An investigation of the association between S100 proteins and HNSCC is thus necessary. Therefore, the expression and mechanisms underlying S100 protein family members in the aforementioned subtypes of HNSCC are discussed in the present review. The present review also aimed to discuss whether S100 protein family members exhibit potential as therapeutic targets of HNSCC, and whether specific changes in S100 protein family members in HNSCC are caused by human papillomavirus (HPV).

2. S100 proteins in OSCC

Changes in the expression of S100 protein members differ in OSCC. The results of previous studies have demonstrated that the expression levels of S100A1, S100A3, S100A6, S100A11, S100A13, S100A14, S100A16 and S100Z are decreased in patients with OSCC, while the expression levels of S100A2, S100A4, S100A7, S100A8, S100A10, S100A12 and S100P are increased (15,35-46). In a previous study, the transcriptome analysis of 93 specimens from OSCC tissues and 87 specimens from adjacent tissues demonstrated that the *S100A1* and *S100A4* expression levels were significantly decreased in OSCC, while those of *S100A2*, *S100A3*, *S100A7* and *S100A11* were increased, compared with those in adjacent tissues (47). Moreover, it has been demonstrated that S100A9 expression is increased in OSCC (15,40,48), whereas the invasion of T1- and T2-stage OSCC mediated by S100A9 has been found to be reduced following treatment with an anti-CD147 antibody (49). However, The Cancer Genome Atlas (TCGA) demonstrated that the *S100A8* and *S100A9* expression levels are reduced in 90% of OSCC cases (50), and this reduction was associated with decreased tumor grading, a decreased expression of epidermal growth factor receptor, epithelial differentiation, the overexpression of apoptosis-related genes, and reduced tumor cell migration and invasion (51). These factors may be due to a wide range of sites involved in OSCC. Therefore, different tissue sites have been selected and different reagents have been used in different experiments. S100A7 expression was previously examined by the immunohistochemical staining of 41 samples from OSCC tissues (>45 years old), 36 samples from OSCC tissues (<45 years old), 40 samples from oral potentially malignant disorders (OPMD) and 36 samples from oral inflammatory lesions (45). Although S100A7 expression was significantly increased in the nuclear, cytoplasmic and membrane staining of OSCC tissues, it was significantly decreased in OPMD, and it was completely absent from oral inflammatory lesions (45). S100A7 may thus serve as a marker to distinguish between oral inflammatory lesions, OPMD and OSCC. The expression of S100A7 protein was not, however, associated with age in that study (45). Another study found a negative association between age and the overexpression of S100A7 mRNA in OSCC (38). The age groups used in the two studies differed, however; the first study (45) used 45 years as the grouping age, while the second study (38) used 65 years. Additionally, the assessment methods used in the two studies differed; immunohistochemical staining was used to detect S100A7 protein expression in the first study (45), while qPCR was used to detect S100A7 mRNA expression in the second study (38).

It was previously discovered that the overexpression of S100A16 not only inhibited the proliferation and invasion of OSCC cells, but also significantly reduced their ability of sphere formation (44). The sphere formation potency, which was used as a surrogate to isolate cancer stem cells (CSCs) from various tumors, can be used to evaluate the capability for self-renewal *in vitro* (52). It was also found that the expression of self-renewal markers, such as octamer-binding transcription factor 4A or B-cell-specific moloney murine leukemia virus integration site 1, was downregulated, confirming the results of sphere formation (44). Additionally, tumors with higher differentiation in mouse models caused by S100A16 overexpression may be related to the upregulation of the expression of involucrin, cytokeratin and other differentiation markers (44). The 10-year survival rate is poorer in patients

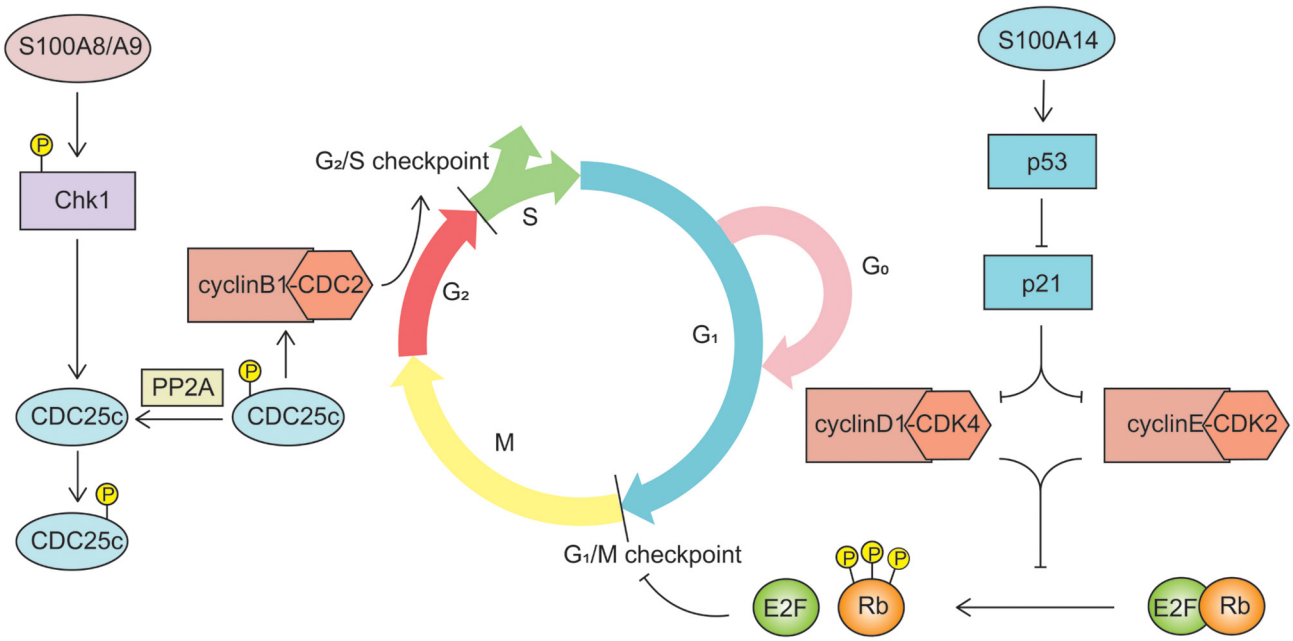

Figure 1. Mechanisms of S100A8/A9 and S100A14 in the OSCC cell cycle. PP2A activity and Chk1 phosphorylation are increased by S100A8/A9, thereby promoting the transition from CDC25c (Thr48) to CDC25c (Ser216), leading to cell cycle arrest in the G2 phase. p21 inhibits the activity of cyclin D1-CDK4 and cyclin E-CDK2. Cyclin-CDK complexes target Rb, for hyperphosphorylation and dissociation from E2F. When cyclin-CDK activity is inhibited, progression from G_1 phase to S phase is blocked, thus arresting cells in the G_1 phase. CDK, cyclin-dependent kinase; Rb, retinoblastoma protein; E2F, E2 transcription factor; OSCC, oral squamous cell carcinoma; PP2A, protein phosphatase 2; Chk1, checkpoint kinase 1; Thr, threonine; Ser, serine; CDC25c, cell division cycle 25c.

with OSCC, as OSCC has lower levels of S100A16 protein than normal tissue, which promotes tumor proliferation and metastasis (44). Sphere formation potency was also associated with drug resistance (52,53). Previously, the KB, SAS and DU145 cell lines with sphere formation ability were demonstrated to exhibit resistance to lapatinib, which was not favorable for the treatment of patients with OSCC (52,53); however, S100A16 was involved in this process required further investigation. OSCC recurrence is also closely linked to S100 proteins. In addition to downregulating E-cadherin expression (54) to facilitate tumor metastasis, the overexpression of S100A4 in OSCC can also regulate stem cells or CSCs, leading to OSCC recurrence through the phosphatase and tensin homolog (PTEN)/phosphorylated intracellular phosphatidylinositol kinase (PI3K)/protein kinase B (AKT) signaling pathway (42). In a previous study, 235 Indian patients with OSCC were divided into the high- and low-risk groups based on the degree of staining of S100A2 in the cytoplasm (43). Only 30% of patients in the low-risk group relapsed, compared to 86% of patients in the high-risk group who experienced recurrence or mortality within 1 year. This finding was confirmed in a cohort of Canadian patients with OSCC, where the rates of recurrence and mortality within 1 year were 81% in the high-risk group and 29% in the low-risk group, respectively (43). It is thus suggested that S100 proteins affect the prognosis of patients with OSCC.

The expression of p21 is increased by *S100A14* in a p53-dependent manner in CaLH3 and OSCC1 cell lines (1). Notably, p21 inhibits the activity of cyclin D1-cyclin-dependent kinase (CDK) 4 and cyclin E-CDK2 (1). Cyclin-CDK complexes target retinoblastoma protein (Rb), for hyperphosphorylation and dissociation from E2 transcription factor (E2F) (55). When cyclin-CDK activity is inhibited, progression from the G_1 phase to S phase is blocked; thus, cells are arrested in the G_1 phase (55) (Fig. 1). Moreover, *S100A8* and *S100A9* regulate the expression of cell division cycle (CDC)25c via phosphorylation at position threonine 48 (Thr48) (56). The activity of protein phosphatase 2 (PP2A), phosphorylation of checkpoint kinase 1 (Chk1) and the expression of CDC25c [phosphorylated at position serine 216 (Ser216)] are subsequently increased (56). Thus, CDC25c (Ser216) is unable to activate cyclin B1/CDC2 and the cell cycle is arrested in the G_2 phase (56) (Fig. 1). By contrast, S100A9 tends to promote cell proliferation in osteosarcoma, and the expression of p21 and p27 has been shown to be upregulated by the knockdown of S100A9, which also causes the inactivation of CDK2 and CDK4, resulting in the cell cycle being arrested in the G_0/G_1 phase (57). The transcription of *S100A13* is regulated via TEA domain transcription factor 4 (TEAD4) in CAL-27 and SCC-4 cell lines (58), in which TEAD4 forms a complex with yes-associated protein (YAP)1 (59). The transcription of G_1 phase-associated cell cycle proteins and CDKs are blocked via the inactivation of this complex or the knockdown of *TEAD4*, thus resulting in cell cycle arrest in the G_1 phase (59). However, whether this process involves S100A13 requires further investigation. Although a number of different mechanisms are involved, certain members of the S100 protein family may interfere with tumor cell proliferation by regulating the cell cycle, thus affecting the development and progression of OSCC.

DNA, RNA, proteins and other substances which are associated with the initiation and progression of OSCC may be secreted into saliva; thus, this is considered as a main source of biomarkers, which can be collected in a non-invasive manner (60). In a previous study, the proteomics analysis of

60 saliva samples from healthy individuals, and patients with OPMD and OSCC revealed that the level of S100A2 in saliva was significantly higher in patients with OSCC, which may be related to the invasion of OSCC (61), indicating that the detection of salivary S100 proteins may be helpful for diagnosis. The efficacy of salivary S100A2 alone in predicting OSCC was poor, and when combined with solute carrier family 3 member 2 and IL-1 receptor antagonist protein, the sensitivity of diagnosis was increased to 83.33% (61). Additionally, it was previously discovered that there was a high association between the expression levels of S100A7 and S100A8 in the saliva of patients with OSCC and the tumor clinical stage (41). The analysis of saliva samples from 100 patients with OSCC revealed that the S100A7 protein was substantially elevated in both T1 and T2 stages (41). The area under receiver-operating characteristic curves (AUROC) was used to rate the sensitivity and specificity of S100A7 protein in predicting T1 stage OSCC [0.71 (95% CI, 0.88-1.04)] and T2 stage OSCC [0.68 (95% CI, 0.89-1.01)], respectively (41). Therefore, salivary S100A7 protein is a possible marker for OSCC at the T1 and T2 stages. The salivary S100A8 protein may be used as a potential marker of OSCC at the T3 and T4 stages. In patients with T3 and T4 stage OSCC, S100A8 protein was detected in 92.9 and 100% of samples, respectively (41). In patients with T3 stage OSCC, the AUROC was 0.99 (95% CI, 0.58-1.00), while in those with T4 stage disease, it was 0.98 (95% CI, 0.63-1.06) (41). According to the aforementioned studies, salivary S100 proteins may prove helpful for the diagnosis of OSCC and even the clinical stage. However, the collection time, processing manner, measurement method and storage conditions of saliva remain to be standardized (62).

3. S100 proteins in HSCC

The prognosis of patients with HSCC may be affected by the expression of S100 proteins. Patients with HSCC with a low *S100A12* expression do not often respond to conventional treatment; thus, the prognosis of patients with HSCC who have a high expression of *S100A12* is improved, compared with that of those with low expression levels (51). However, the prognosis of patients with HSCC with high levels of S100A4 expression differs. The results of a previous study demonstrated that migration and invasion were inhibited following the knockdown of *S100A4* in FaDu cells, and HSCC metastasis in *Drosophila* and mouse models was reduced (63). On the one hand, S100A4 may inhibit the activation of p53 and reduce p53 binding to the promoter region of Nanog to upregulate its transcriptional activity (64). This may enhance the stem cell properties of cancer-initiating cells (64), thus promoting tumor proliferation (Fig. 2). On the other hand, S100A4 may interact with actin and tropomyosin to cause cell migration (12). Therefore, patients with HSCC with an elevated expression of S100A4 often exhibit a poor prognosis (Fig. 2). The expression levels of S100A9 and S100A11 in HSCC are markedly higher than those in adjacent healthy tissues (65,66). Moreover, the expression of S100A9 increases with the severity of clinical stages; its expression is higher in patients with stage III and IV HSCC than in those with stage I and II disease (65). Compared with patients with a low S100A9 expression, patients with high expression levels of S100A9 also exhibit a poor prognosis and a significantly reduced 5-year survival rate (65).

The expression levels of matrix metalloprotein (MMP)2, MMP7 and MMP9 have been found to be significantly decreased following the downregulation of *S100A9* and *S100A11* expression (65,66) (Fig. 2). MMPs belong to the zinc finger protein family that degrade almost all components of the extracellular matrix (67), and are thus considered essential proteases in the progression of epithelial-mesenchymal transition. MMPs also induce angiogenesis (68), to provide energy to support tumor cell diffusion. In addition, the downregulation of *S100A9* expression has been shown to significantly reduce the expression of nuclear factor κB (NF-κB), the phosphorylation of NF-κB and B-cell lymphoma 2 (Bcl-2) (65). Notably, restoring the expression of NF-κB reverses the inhibitory effects on cell proliferation and invasion (65). Bcl-2 is a well-established inhibitor of apoptosis, regulating apoptosis by controlling the release of mitochondrial cytochrome *c* (69). Apoptosis is also promoted via the reduction of mitochondrial cytochrome *c* phosphorylation (69). Thus, S100A9 may promote HSCC proliferation in this manner (Fig. 2). In addition, S100A9 may bind the receptor of advanced glycosylation end products (RAGE) to promote the phosphorylation of extracellular regulated protein kinases 1/2 (ERK1/2), stress-activated protein kinase/c-Jun amino-terminal kinase (JNK) and inhibitor of NF-κBα (IκBα) (70). In turn, this activates the mitogen-activated protein kinase (MAPK) signaling pathway and causes the nuclear translocation of NF-κB (70) (Fig. 2). Moreover, the expression levels of PI3K, AKT, mammalian target of rapamycin (mTOR) and Bcl-2 have been shown to be reduced following the knockdown of *S100A11* in FaDu cells, suggesting that S100A11 may affect migration via mediating the PI3K/Akt/mTOR signaling pathway (66) (Fig. 2). In conclusion, S100 proteins may affect downstream genes in HSCC through NF-κB or PI3K/Akt/mTOR signaling pathways, to promote the initiation and progression of HSCC.

4. S100 proteins in NSCC

The sustained upregulation of S100A4 expression is observed during dedifferentiation, using two-dimensional liquid chromatography and tandem mass spectrometry, combined with isotopic labeling, and relative and absolute quantification techniques (71). These methods have been used to analyze keratinizing NSCC, non-keratinizing NSCC and undifferentiated NSCC, and the results have demonstrated that S100A7, S100A8 and S100A9 expression levels are also significantly reduced (71). In a previous study, the S100A6 protein levels were higher in the poorly differentiated cell line, CNE2, than in the well-differentiated cell line, NP96, in NSCC, and S100A6 expression was also increased (72). The upregulation of S100A6 led to cell proliferation, which was decreased by SB20358 disrupting the p38/MAPK signaling pathway, which led to cell growth arrest and apoptosis (72). These findings suggest that S100A6 controls the proliferation of NSCC cells via the p38/MAPK signaling pathway (72). The growth of NSCC, as well as its differentiation are both enhanced by S100 proteins. *S100A8* and *S100A9* not only promote the activation of Thr308 and Ser473 through the PI3K/AKT pathway (73), following the control of the expression of effector targets in

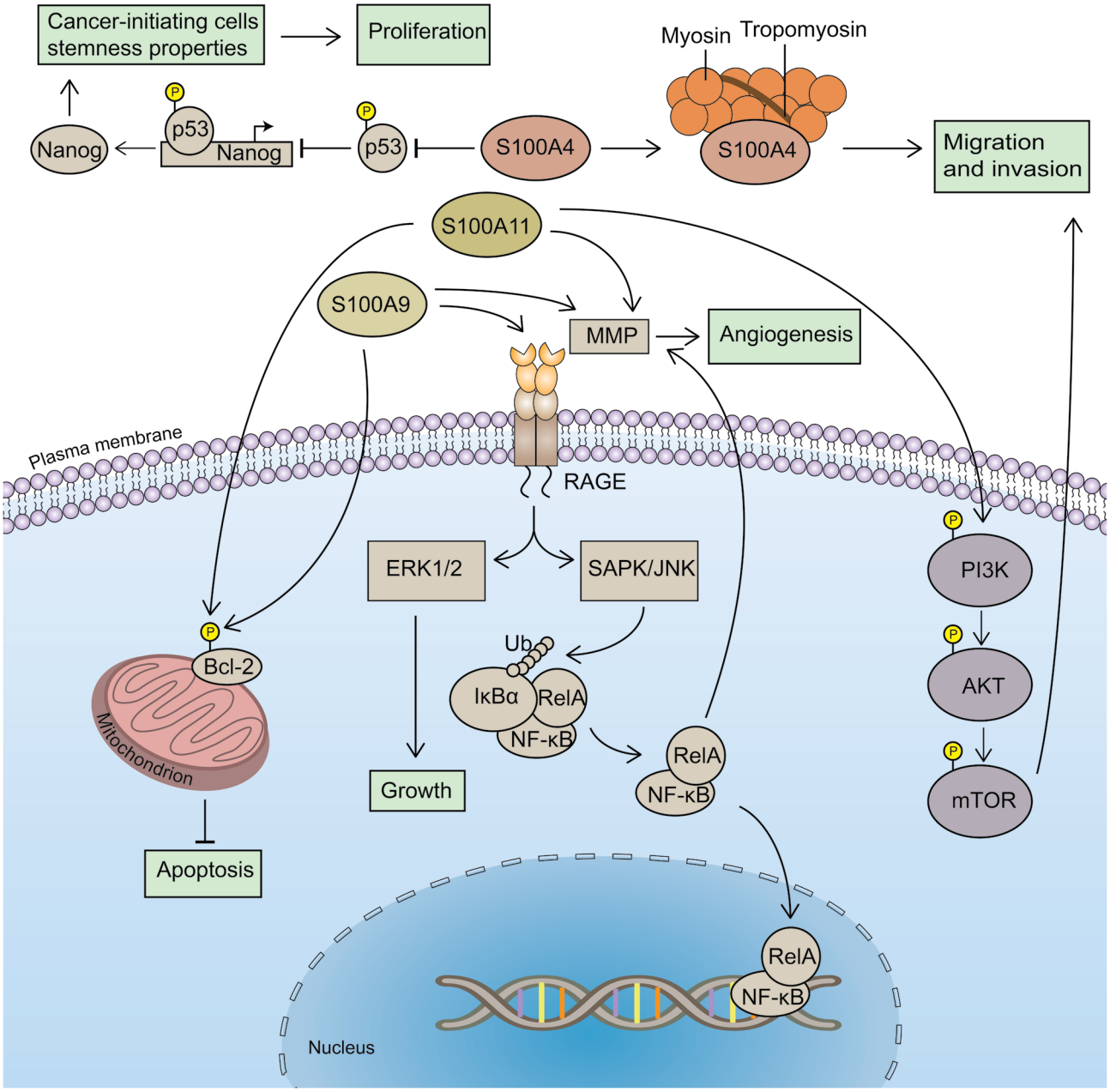

Figure 2. Mechanisms of action of S100A4, S100A9 and S100A11 in the development of HSCC. p53 binding to the Nanog promoter region may be inhibited by S100A4, promoting tumor proliferation. S100A4 may also act with actin and tropomyosin to promote cell migration. The expression of MMP and the phosphorylation of Bcl-2 may be upregulated by S100A9 and S100A11. In addition, S100A9 may activate ERK1/2 and SAPK/JNK through the binding of RAGE to promote nuclear translocation of NF-κB, and S100A11 may promote the expression of phosphorylated PI3K, AKT and mTOR. The aforementioned factors may promote the initiation and progression of HSCC. HSCC, hypopharyngeal squamous cell carcinoma; MMP, matrix metalloprotein; Bcl-2, bcelllymphoma-2; ERK, extracellular regulated protein kinases; SAPK, stress-activated protein kinase; JNK, c-Jun amino-terminal kinase; RAGE, receptor of advanced glycosylation end products; NF-κB, nuclear factor κB; PI3K, phosphatidylinositol kinase; Akt, protein kinase B; mTOR, mammalian target of rapamycin.

the nucleus to control the expression of effector targets in the nucleus and promote cell proliferation, but also increase the expression of MMP7, MMP9 and MMP12, which are involved in the metastasis of NSCC (67). The proliferation or migration of C666-1 cells is increased by S100P combined with RAGE, which also activates the NF-κB signaling pathway and MAPK (74).

At present, paclitaxel is one of the conventional treatment strategies used for NSCC (75); however, metastasis to the neck, local recurrence or drug resistance may lead to a decreased efficacy. The increased phosphorylation of IL-1 receptor associated kinase (IRAK1) was found in NSCC samples following metastasis and recurrence caused by paclitaxel resistance. Notably, the IRAK1/S100A9 axis is closely associated with drug resistance in NSCC (75) (Fig. 3). The sensitivity to paclitaxel was found to be increased in S26 and CNE2 cell lines following *S100A9* knockdown; however, the resistance to paclitaxel was restored following the addition of recombinant S100A9 (75). Notably, the deletion of *IRAK1* also significantly reduced the expression of S100A9 (75). *S100A14* promoted the degradation of IRAK1, and the expression and phosphorylation of IRAK1 were reduced following treatment with the IRAK1

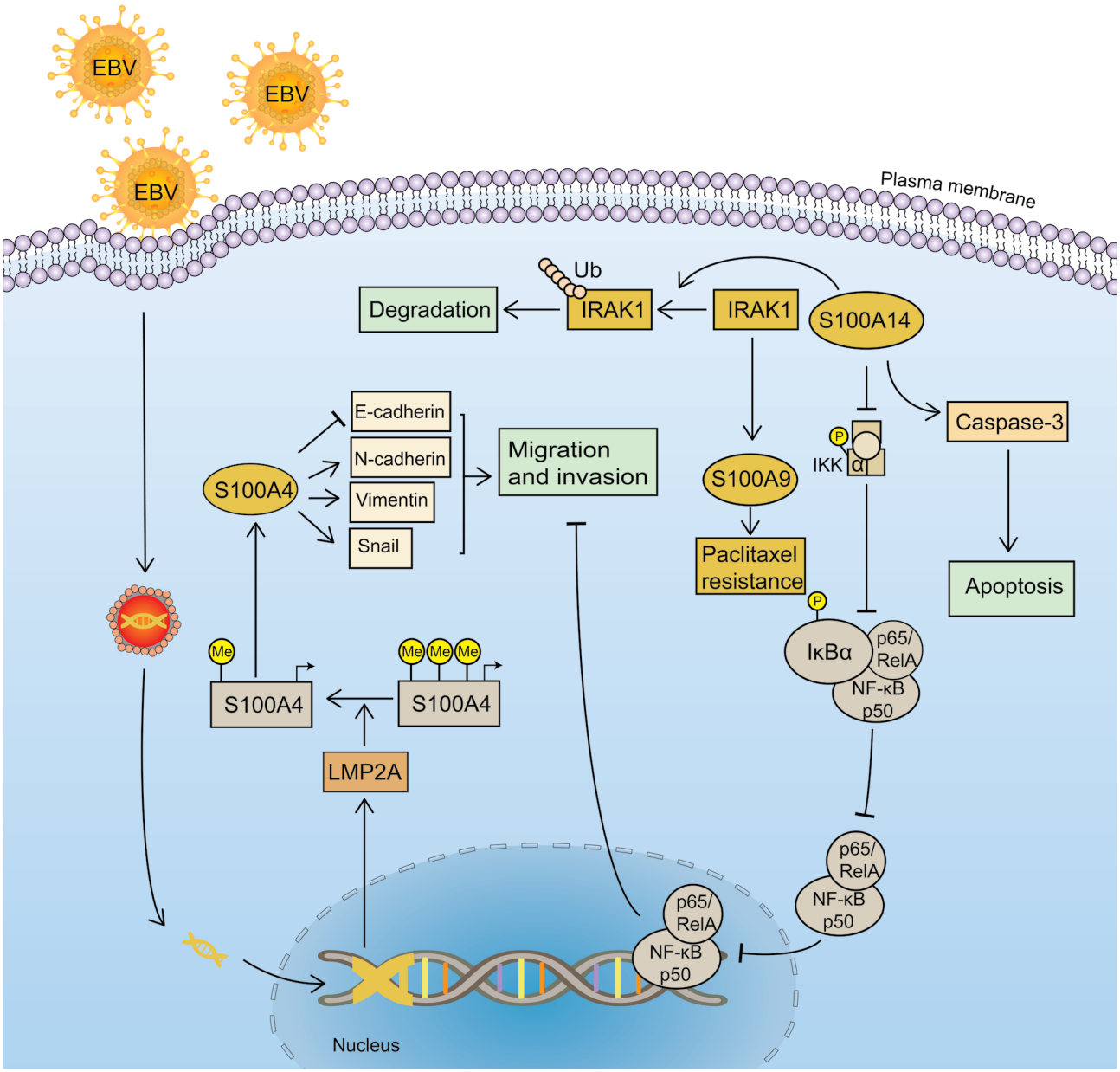

Figure 3. Biological functions of S100A4, S100A9 and S100A14 in NSCC. EBV encoding LMP2A promotes S100A4 demethylation, thereby promoting tumor migration and invasion. Expression of S100A9 is upregulated by the IRAK1/S100A9 axis to mediate paclitaxel resistance in NSCC. S100A14 increases the expression of caspase-3 to enhance apoptosis, and inhibit the nuclear translocation of p50 and p65. This impairs NSCC migration. NSCC, nasopharyngeal squamous cell carcinoma; EBV, Epstein-Barr virus; LMP2A, latent membrane protein 2A; IRAK1, interleukin-1 receptor associated kinase.

inhibitor, T2457, in CNE-1 and S26 cell lines (76). In addition, it was also demonstrated that the increased expression of *S100A14* and the inhibition of cell migration were reversed following treatment with IL-1β to induce IRAK1 phosphorylation (76). Following the overexpression of *S100A14* in S18 and 5-8F cell lines, the expression of caspase-3 and apoptosis were increased, and the phosphorylation of IκBα and inhibitor of kappa B kinase were reduced. This inhibited the nuclear translocation of p50 and p65, and impaired NSCC migration (76) (Fig. 3). Therefore, S100 proteins may improve chemosensitivity and reduce drug resistance in NSCC.

Epstein-Barr virus (EBV)-related NSCC is extremely common; there are ~78,100 new cases of EBV-related NSCC worldwide each year (77). EBV encodes latent membrane protein 2A (LMP2A) (78). The expression of S100A4 in CNE-1 cells with or without ectopic LMP2A expression was previously measured using a transfer gene array, and the results demonstrated that S100A4 expression was significantly upregulated in LMP2A-positive NSCC (78). Another previous study also reported that LMP2A was involved in gene methylation regulation in EBV-related gastric cancer, inducing signal transducer and activator of transcription 3 phosphorylation and activating DNA methyltransferase 1 (79). This led to the inhibition of PTEN deleted on chromosome 10 promoter methylation, reducing its transcription (79). Methylation levels of CNE-1 and CNE-1-LMP2A cells were also previously detected, to verify whether LMP2A exhibited a similar function in NSCC (78). The results demonstrated that LMP2A was significantly demethylated in CNE-1 cells without the ectopic expression of LMP2A (Fig. 3), and S100A4 expression was

increased following the inhibition of DNA methyltransferase via 5-aza-2-deoxycytidine (78). This resulted in the increased expression of N-cadherin, vimentin and Snail, and in the reduced expression of E-cadherin (78) (Fig. 3). These results suggested that the expression of S100 proteins may be regulated by methylation, and EBV-induced hypomethylation may reduce expression in NSCC.

5. S100 proteins in PTC

PTC is often associated with changes in 1q21, 1q42, 5p15 and 9q34 (80), while the majority of members of the S100 protein family are located on 1q21. This suggests that S100 proteins are associated with PTC. The expression of each member of the S100 protein family in 9 healthy thyroid tissues and 27 thyroid tumor tissues was previously analyzed using multiplex and targeted mass spectrometry (81). The results demonstrated that *S100A6*, *S100A11* and *S100A13* expression were upregulated in PTC, compared with healthy thyroid samples (81). Notably, S100A6 (82) and S100A11 may help to identify follicular and papillary tumors (81), and *S100A6* may function as a specific therapeutic target of PTC (83). Although the elevation of S100A13 expression is less prominent than that of S100A6 and S100A11, it may also function as a candidate biomarker for PTC (81). As previously demonstrated, the PTC cell cycle may be regulated by S100 proteins; following the knockdown of S100A12 in TPC1 or K1 cell lines, the expression of cyclin D1 and CDK4 was markedly downregulated, contributing to the accumulation of PTC cells in the G_0/G_1 phase (84). These findings demonstrate that certain members of the S100 protein family are strongly associated with the initiation and progression of PTC, and may function as early diagnostic markers of PTC.

In K1 and BCPAP cell lines transiently transfected with *S100A1*, nuclear and cytoplasmic YAP expression was found to be decreased following the knockdown of *S100A1*; however, the phosphorylation of YAP in the cytoplasm was significantly increased, suggesting that *S100A1* may promote the proliferation of PTC cells and inhibit apoptosis via the Hippo signaling pathway (85) (Fig. 4). The results of a previous study demonstrated that the *S100A1*-mediated Hippo signaling pathway was positively regulated by lncRNA FOXD2 adjacent opposite strand RNA 1 (FOXD2-AS1), and FOXD2-AS1 overexpression-mediated cell proliferation, migration and invasion were reversed following *S100A1* knockdown in breast cancer cells (86). Following the knockdown of lncRNA FOXD2-AS1, the expression of S100A1 was also suppressed (86). The high expression of FOXD2-AS1 was also observed in PTC, and this was associated with a poor prognosis (87). Therefore, PTC may also enhance tumor proliferation and invasion through the lncRNA FOXD2-AS1/S100A1/Hippo signaling pathway, resulting in decreased survival (Fig. 4). lncRNA HOXA cluster antisense RNA2 (HOXA-AS2) differs from lncRNA FOXD2-AS1. HOXA-AS2 binds to the 3'-untranslated region complementary binding site of microRNA (miRNA/miR)-520c-3p to reduce the expression of miR-520c-3p, and upregulate downstream gene *S100A4* to promote PTC metastasis (88,89) (Fig. 4). Moreover, the results of a previous study demonstrated that miR-181a binding to lysine demethylase 5C in TPC-1 cells increased the trimethylation of lysine 4 on histone H3 protein subunit (H3K4me3) on the *S100A2* promoter region, to promote S100A2 expression (90) (Fig. 4). This pathway also enhances the migration and invasion of PTC cells by promoting the expression of Ki-67, MMP2 and MMP9, and inhibits apoptosis through reducing caspase-3 (90). These results indicate that lncRNA and miRNA regulate the expression of S100 protein family members through different methods to promote the development of PTC.

6. S100 proteins in HPV-induced HNSCC

HPV is involved in the development of HNSCC (91). A previous retrospective analysis demonstrated that 34.5% of 4,852 patients with HNSCC worldwide were HPV-positive (92). Notably, HPV-induced HNSCC has continued to increase in recent years, and the prevalence of HPV-positive OSCC in the USA increased by 225% between 1975 and 2012 (91). In addition, HPV-16 is the most common genotype in HPV-positive OSCC (93), followed by HPV-18 (92). HPV-positive OSCC is caused by HPV-16 and HPV-18 at rates of 32.4 and 11.3%, respectively (92). Notably, HPV infection increases the prevalence of HNSCC; however, patients that are HPV-positive are more sensitive to treatment, and exhibit an improved prognosis and a higher survival rate than patients who are HPV-negative (94-96). The 3-year overall survival rates of patients with HNSCC who are HPV-positive and HPV-negative have been shown to be 82.4 and 57.1%, respectively (94,97). Due to the significant impact of HPV on prognosis, infection status assessment has been included in global treatment guidelines (91).

The clinical proteomic and immunohistochemical analysis of HPV-18-positive and -negative OSCC has indicated that S100A8 protein is upregulated >10-fold in HPV-18-positive OSCC (98). S100A8 and S100A9 heterodimers induce the transfer of myeloid differentiation factor adaptor proteins from the cytoplasm to the Toll-like receptor 4 (TLR4) receptor complex, activating extracellular signal-regulated kinases, NF-κB, p38 and JNK, to induce pro-inflammatory signal transduction, thus activating the innate immune system (99). In a previous study, the results obtained from TCGA demonstrated that *S100A8* and *S100A9* RNA expression levels in HPV-positive HNSCC were lower than those in HPV-negative HNSCC; however, the difference was not statistically significant (100). HPV-16 exerts no significant effect on *S100A8* mRNA expression levels; however, it has been shown to reduce *S100A8* promoter activity, suggesting that HPV may regulate the expression of S100 proteins at the post-transcriptional translation stage (101). In comparison to healthy skin, *S100A8* and *S100A9* are hypomethylated in HPV-induced warts (102). It is necessary to determine whether HPV may influence S100A8 expression throughout the post-transcriptional translation stage of HNSCC and how this regulation relates to methylation. It is also necessary to learn more about the S100 protein family members in HNSCC and how they relate to HPV.

7. Conclusions and future perspectives

Numerous previous studies have verified that members of the S100 protein family are closely associated with HNSCC (Table I). Certain members of the S100 protein family may

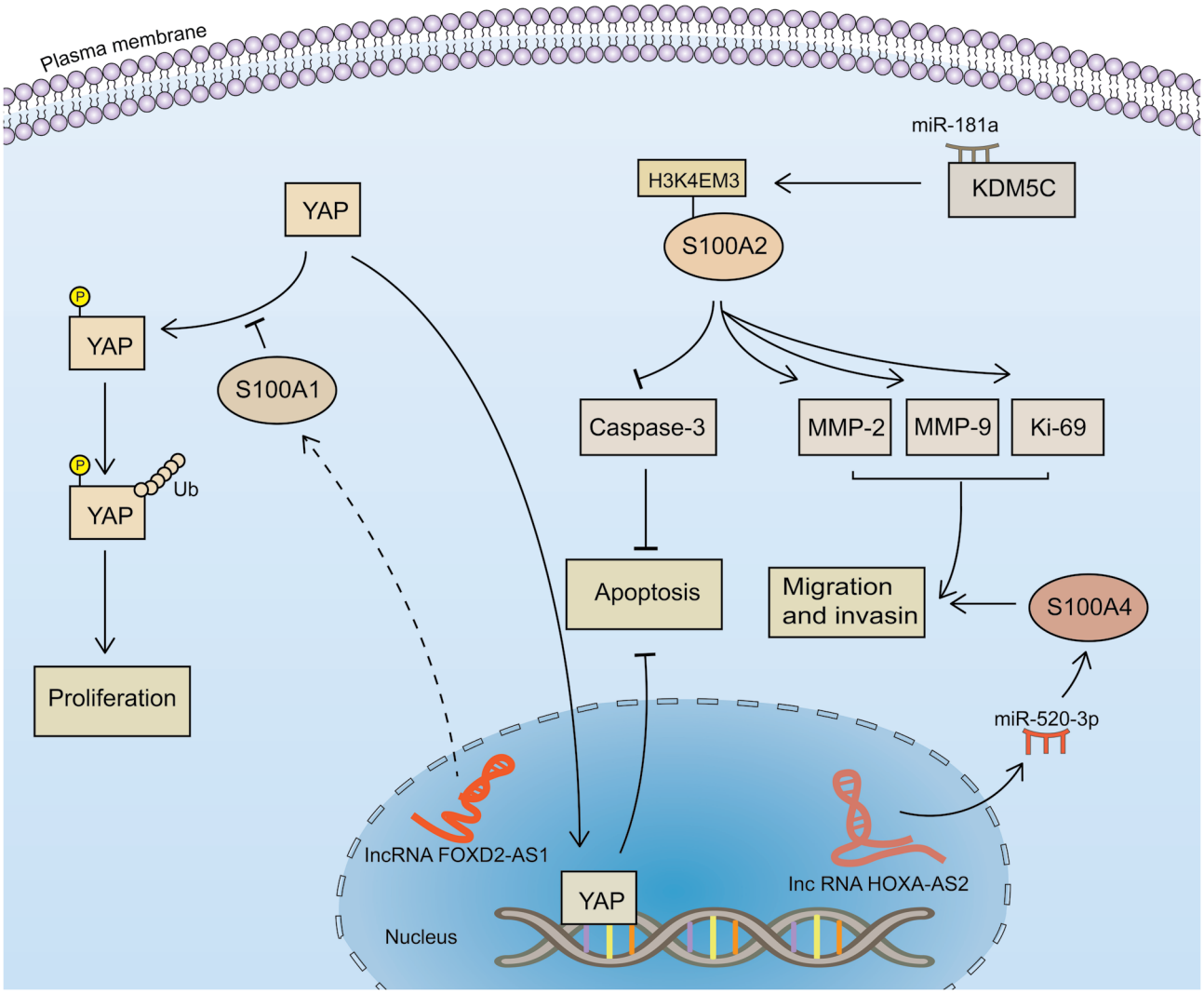

Figure 4. Mechanisms of the interactions of S100A1, S100A2 and S100A4 with lncRNA and miRNA in PTC. S100A1 inhibits YAP phosphorylation to promote PTC cell proliferation, which may be positively regulated by lncRNA FOXD2-AS1. The expression of S100A4 is increased to promote PTC metastasis, which is mediated by lncRNA HOXA-AS2 through miR-520c-3p. H3K4me3 modification on the S100A2 promoter region promotes S100A2 expression, and is increased by miR-181a binding to KDM5C. This enhances the migration and invasion of PTC cells, and decreases apoptosis. LncRNA, long non-coding RNA; miRNA, microRNA; YAP, yes-associated protein; PTC, papillary thyroid carcinoma; FOXD2-AS1, FOXD2 adjacent opposite strand RNA 1; HOXA-AS2, HOXA cluster antisense RNA2; H3K4me3, trimethylation of lysine 4 on histone H3 protein subunit; KDM5C, lysine demethylase 5C.

block the cell cycle and induce YAP phosphorylation to regulate cell proliferation (Table I). Moreover, these proteins mediate NF-κB, PI3K/AKT/mTOR and other signaling pathways to participate in HNSCC invasion, metastasis and recurrence (Table I). Thus, further investigations into the role of S100 proteins in the initiation and progression of HNSCC are required. Vertical flow microarrays based on gold nanoparticles and surface-enhanced Raman spectroscopy have been developed to reduce pain in patients, using detection in saliva, and to differentiate OSCC from healthy groups and quantify *S100P* mRNA expression levels (103). Thus, these methods may be useful mainstream tools for HNSCC detection due to their non-invasive, convenient and accurate nature. At present, there are no drugs available which can directly target this family that are used in clinical practice; however, numerous investigations are focused on increasing the sensitivity of tumors to chemotherapeutic drugs that regulate S100 proteins. Low expression levels of S100A11 in HSCC improve the sensitivity of FaDu cells to the thymidylate synthase inhibitor 5-fluorouracil (66), and the inhibition of the IRAK1-S100A9 axis may reduce the resistance of NSCC to paclitaxel (75). Moreover, AKT and ERK1/2 signaling inactivation via *S100A4* knockdown induces apoptosis, which reverses the resistance to vemurafenib and inhibits thyroid cancer cell invasion and proliferation (104). Blocking the binding of S100 proteins to target proteins has been attempted in order to cure tumors in clinical practice. Quinoline-3-carboxamide derivatives suppress tumors by interfering with the interaction of S100A8/S100A9 with TLR4 or RAGE (105). Amiloridezinol blocks the interaction of S100A13 with fibroblast growth factor 1 (FGF1), decreasing S100A13-FGF1 complex formation (106). In addition, the covalent modification of S100A7, S100A10 and S100A11 through transglutaminase, and the S-nitrosylation or phosphorylation of S100A1, S100A8, S100A9 and S100B also indirectly affects disease

Table I. Functions and mechanisms of action of S100 protein family members in the subtypes of HNSCC.

Cancer type	S100	Functions	(Refs.)
OSCC	S100A2	High expression often indicates a poor prognosis.	(43,61)
OSCC	S100A4	Regulates cancer stem cells. Associated with OSCC recurrence.	(42,54)
OSCC	S100A7	Distinguishes OSCC, OPMD and oral inflammatory lesions. Potential salivary marker for T1 and T2 stages of OSCC.	(41,45)
OSCC	S100A8	Arrests the cell cycle in the G_2 phase. Potential salivary marker for T3 and T4 stages of OSCC.	(41,56)
OSCC	S100A9	Arrests the cell cycle in the G_2 phase.	(56)
OSCC	S100A14	Arrests the cell cycle in the G_1 phase.	(1)
OSCC	S100A16	Regulates OSCC proliferation, metastasis and sphere formation ability.	(44)
HSCC	S100A4	Regulates HSCC cell migration and invasion. Enhances the stem cell properties of cancer-initiating cells.	(63,64)
HSCC	S100A9	Regulates MMP2 and MMP7 expression levels. Reduces the expression of NF-κB, the phosphorylation of NF-κB and Bcl-2.	(65)
HSCC	S100A11	Regulates the MMP9 expression level. Mediates the PI3K/Akt/mTOR signaling pathway to affect HSCC migration.	(66)
HSCC	S100A12	Alters sensitivity to conventional treatment.	(51)
NSCC	S100P	Combines with RAGE to activate the NF-κB signaling pathway and mitogen-activated protein kinase.	(74)
NSCC	S100A4	Associated with EBV-related NSCC.Increases N-cadherin, vimentin and Snail expression levels.Reduces the E-cadherin expression level.	(78)
NSCC	S100A6	Promotes cell proliferation through the p38/MAPK signaling pathway.	(72)
NSCC	S100A8	Activation of Thr308 and Ser473 through the PI3K/AKT pathway. Increases the expression of MMP7, MMP9 and MMP12 to promote NSCC metastasis.	(67,73)
NSCC	S100A9	Activation of Thr308 and Ser473 through the PI3K/AKT pathway Increases the expression of MMP7, MMP9 and MMP12 to promote NSCC metastasis.Closely associated with paclitaxel resistance.	(67,73,75)
NSCC	S100A14	Promotes the degradation of IRAK1. Increases the expression of caspase-3 and apoptosis. Reduces the phosphorylation of IκBα and inhibitor of κB kinase to impair NSCC migration.	(76)
PTC	S100A1	Enhances proliferation and invasion, and inhibits apoptosis.	(85)
PTC	S100A2	Promotes the expression of Ki-67, MMP-2 and MMP-9. Inhibits apoptosis by reducing the caspase-3 expression level.	(90)
PTC	S100A4	Promotes PTC metastasis.	(88,89)
PTC	S100A6	Helps to identify follicular and papillary tumors.	(81-83)
PTC	S100A11	Helps to identify follicular and papillary tumors.	(81)
PTC	S100A12	Regulates the cell cycle.	(84)
PTC	S100A13	A candidate biomarker for PTC.	(81)

OSCC, oral squamous cell carcinoma; HSCC, hypopharyngeal squamous cell carcinoma; NSCC, nasopharyngeal squamous cell carcinoma; PTC, papillary thyroid carcinoma; OPMD, oral potentially malignant disorders; MMP, matrix metalloprotein; NF-κB, nuclear factor κB; PI3K, phosphatidylinositol kinase; Bcl-2, bcelllymphoma-2; Akt, protein kinase B; mTOR, mammalian target of rapamycin; RAGE, receptor of advanced glycosylation end products; EBV, Epstein-Barr virus; MAPK, mitogen-activated protein kinase; Thr308 phosphorylation at position threonine 308; Ser473, phosphorylated at position serine 473; IRAK1, interleukin-1 receptor associated kinase; IκBα, inhibitor of NF-κBα.

progression (2). Although the association between other family members and HNSCC requires further research, the present review provides an overview of the mechanisms underlying S100 proteins in HNSCC, and may provide novel insight into the prevention, diagnosis and treatment of HNSCC.

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Availability of data and materials

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Authors' contributions

YHu and XZ conceived and designed the study. YHu wrote the manuscript. YHa, MH and YZ participated in revising and proofreading the manuscript. All authors have read and approved the final manuscript. Data authentication is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Sapkota D, Costea DE, Blo M, Bruland O, Lorens JB, Vasstrand EN and Ibrahim SO: S100A14 inhibits proliferation of oral carcinoma derived cells through G1-arrest. Oral Oncol 48: 219-225, 2012.
2. Gonzalez LL, Garrie K and Turner MD: Role of S100 proteins in health and disease. Biochim Biophys Acta Mol Cell Res 1867: 118677, 2020.
3. Zaia AA, Sappington KJ, Nisapakultorn K, Chazin WJ, Dietrich EA, Ross KF and Herzberg MC: Subversion of antimicrobial calprotectin (S100A8/S100A9 complex) in the cytoplasm of TR146 epithelial cells after invasion by Listeria monocytogenes. Mucosal Immunol 2: 43-53, 2009.
4. Zou X, Sorenson BS, Ross KF and Herzberg MC: Augmentation of epithelial resistance to invading bacteria by using mRNA transfections. Infect Immun 81: 3975-3983, 2013.
5. Singh P and Ali SA: Multifunctional role of S100 protein family in the immune system: An Update. Cells 11: 2274, 2022.
6. Crowe LAN, McLean M, Kitson SM, Melchor EG, Patommel K, Cao HM, Reilly JH, Leach WJ, Rooney BP, Spencer SJ, *et al*: S100A8 & S100A9: Alarmin mediated inflammation in tendinopathy. Sci Rep 9: 1463, 2019.
7. Schenten V, Plançon S, Jung N, Hann J, Bueb JL, Bréchard S, Tschirhart EJ and Tolle F: Secretion of the phosphorylated form of S100A9 from neutrophils is essential for the proinflammatory functions of extracellular S100A8/A9. Front Immunol 9: 447, 2018.
8. De Veirman K, De Beule N, Maes K, Menu E, De Bruyne E, De Raeve H, Fostier K, Moreaux J, Kassambara A, Hose D, *et al*: Extracellular S100A9 protein in bone marrow supports multiple myeloma survival by stimulating angiogenesis and cytokine secretion. Cancer Immunol Res 5: 839-846, 2017.
9. Wu Z, Jiang D, Huang X, Cai M, Yuan K and Huang P: S100A8 as a promising biomarker and oncogenic immune protein in the tumor microenvironment: An integrative pancancer analysis. J Oncol 2022: 6947652, 2022.
10. Basso D, Fogar P and Plebani M: The S100A8/A9 complex reduces CTLA4 expression by immature myeloid cells: Implications for pancreatic cancer-driven immunosuppression. Oncoimmunology 2: e24441, 2013.
11. Kligman D and Hilt DC: The S100 protein family. Trends Biochem Sci 13: 437-443, 1988.
12. Allgower C, Kretz AL, von Karstedt S, Wittau M, Henne-Bruns D and Lemke J: Friend or Foe: S100 proteins in cancer. Cancers (Basel) 12: 2037, 2020.
13. Delangre E, Oppliger E, Berkcan S, Gjorgjieva M, Correia de Sousa M and Foti M: S100 proteins in fatty liver disease and hepatocellular carcinoma. Int J Mol Sci 23: 11030, 2022.
14. Austermann J, Spiekermann C and Roth J: S100 proteins in rheumatic diseases. Nat Rev Rheumatol 14: 528-541, 2018.
15. Raffat MA, Hadi NI, Hosein M, Mirza S, Ikram S and Akram Z: S100 proteins in oral squamous cell carcinoma. Clin Chim Acta 480: 143-149, 2018.
16. Zhang S, Wang Z, Liu W, Lei R, Shan J, Li L and Wang X: Distinct prognostic values of S100 mRNA expression in breast cancer. Sci Rep 7: 39786, 2017.
17. Bai Y, Li LD, Li J and Lu X: Prognostic values of S100 family members in ovarian cancer patients. BMC Cancer 18: 1256, 2018.
18. Kleindienst A, Hesse F, Bullock MR and Buchfelder M: The neurotrophic protein S100B: Value as a marker of brain damage and possible therapeutic implications. Prog Brain Res 161: 317-325, 2007.
19. Li HB, Wang JL, Jin XD, Zhao L, Ye HL, Kuang YB, Ma Y, Jiang XY and Yu ZY: Comprehensive analysis of the transcriptional expressions and prognostic value of S100A family in pancreatic ductal adenocarcinoma. BMC Cancer 21: 1039, 2021.
20. Arantes L, De Carvalho AC, Melendez ME and Lopes Carvalho A: Serum, plasma and saliva biomarkers for head and neck cancer. Expert Rev Mol Diagn 18: 85-112, 2018.
21. Chi H, Xie X, Yan Y, Peng G, Strohmer DF, Lai G, Zhao S, Xia Z and Tian G: Natural killer cell-related prognosis signature characterizes immune landscape and predicts prognosis of HNSCC. Front Immunol 13: 1018685, 2022.
22. Xu J, Lai F, Liu Y, Tan Z, Zheng C, Wang J, Guo H, Jiang L, Ge X, Lan X, *et al*: Novel computer-aided reconstruction of soft tissue defects following resection of oral and oropharyngeal squamous cell carcinoma. World J Surg Oncol 20: 196, 2022.
23. Runge A, Mayr M, Schwaiger T, Sprung S, Chetta P, Gottfried T, Dudas J, Greier MC, Glatz MC, Haybaeck J, *et al*: Patient-derived head and neck tumor slice cultures: A versatile tool to study oncolytic virus action. Sci Rep 12: 15334, 2022.
24. Mei Z, Zhang K, Lam AK, Huang J, Qiu F, Qiao B and Zhang Y: MUC1 as a target for CAR-T therapy in head and neck squamous cell carinoma. Cancer Med 9: 640-652, 2020.
25. Chen Y, Li ZY, Zhou GQ and Sun Y: An Immune-related gene prognostic index for head and neck squamous cell carcinoma. Clin Cancer Res 27: 330-341, 2021.
26. Chen Y, Jiang N, Chen M, Sui B and Liu X: Identification of tumor antigens and immune subtypes in head and neck squamous cell carcinoma for mRNA vaccine development. Front Cell Dev Biol 10: 1064754, 2022.
27. Wei T, Leisegang M, Xia M, Kiyotani K, Li N, Zeng C, Deng C, Jiang J, Harada M, Agrawal N, *et al*: Generation of neoantigen-specific T cells for adoptive cell transfer for treating head and neck squamous cell carcinoma. Oncoimmunology 10: 1929726, 2021.
28. Massarelli E, William W, Johnson F, Kies M, Ferrarotto R, Guo M, Feng L, Lee JJ, Tran H, Kim YU, *et al*: Combining immune checkpoint blockade and tumor-specific vaccine for patients with incurable human papillomavirus 16-related cancer: A phase 2 clinical trial. JAMA Oncol 5: 67-73, 2019.
29. Sasahira T, Kurihara-Shimomura M, Shimojjukoku Y, Shima K and Kirita T: Searching for new molecular targets for oral squamous cell carcinoma with a view to clinical implementation of precision medicine. J Pers Med 12: 413, 2022.
30. Sundriyal D, Arya L, Saha R, Walia M and Nayak PP: Hypercalcemia of malignancy: Time to pull the brakes. Indian J Surg Oncol 13: 28-32, 2022.

31. Roesch Ely M, Nees M, Karsai S, Magele I, Bogumil R, Vorderwulbecke S, Ruess A, Dietz A, Schnolzer M and Bosch FX: Transcript and proteome analysis reveals reduced expression of calgranulins in head and neck squamous cell carcinoma. Eur J Cell Biol 84: 431-444, 2005.
32. Argyris PP, Slama Z, Malz C, Koutlas IG, Pakzad B, Patel K, Kademani D, Khammanivong A and Herzberg MC: Intracellular calprotectin (S100A8/A9) controls epithelial differentiation and caspase-mediated cleavage of EGFR in head and neck squamous cell carcinoma. Oral Oncol 95: 1-10, 2019.
33. Oliveira MV, Fraga CA, Barros LO, Pereira CS, Santos SH, Basile JR, Gomez RS, Guimaraes AL and De-Paula AM: High expression of S100A4 and endoglin is associated with metastatic disease in head and neck squamous cell carcinoma. Clin Exp Metastasis 31: 639-649, 2014.
34. Tripathi SC, Matta A, Kaur J, Grigull J, Chauhan SS, Thakar A, Shukla NK, Duggal R, DattaGupta S, Ralhan R and Siu KW: Nuclear S100A7 is associated with poor prognosis in head and neck cancer. PLoS One 5: e11939, 2010.
35. Sapkota D, Bruland O, Bøe OE, Bakeer H, Elgindi OA, Vasstrand EN and Ibrahim SO: Expression profile of the S100 gene family members in oral squamous cell carcinomas. J Oral Pathol Med 37: 607-615, 2008.
36. Driemel O, Murzik U, Escher N, Melle C, Bleul A, Dahse R, Reichert TE, Ernst G and von Eggeling F: Protein profiling of oral brush biopsies: S100A8 and S100A9 can differentiate between normal, premalignant, and tumor cells. Proteomics Clin Appl 1: 486-493, 2007.
37. Hu S, Arellano M, Boontheung P, Wang J, Zhou H, Jiang J, Elashoff D, Wei R, Loo JA and Wong DT: Salivary proteomics for oral cancer biomarker discovery. Clin Cancer Res 14: 6246-6252, 2008.
38. Kesting MR, Sudhoff H, Hasler RJ, Nieberler M, Pautke C, Wolff KD, Wagenpfeil S, Al-Benna S, Jacobsen F and Steinstraesser L: Psoriasin (S100A7) up-regulation in oral squamous cell carcinoma and its relation to clinicopathologic features. Oral Oncol 45: 731-736, 2009.
39. Sapkota D, Bruland O, Costea DE, Haugen H, Vasstrand EN and Ibrahim SO: S100A14 regulates the invasive potential of oral squamous cell carcinoma derived cell-lines in vitro by modulating expression of matrix metalloproteinases, MMP1 and MMP9. Eur J Cancer 47: 600-710, 2011.
40. Roman E, Lunde ML, Miron T, Warnakulasauriya S, Johannessen AC, Vasstrand EN and Ibrahim SO: Analysis of protein expression profile of oral squamous cell carcinoma by MALDI-TOF-MS. Anticancer Res 33: 837-845, 2013.
41. Jou YJ, Hua CH, Lin CD, Lai CH, Huang SH, Tsai MH, Kao JY and Lin CW: S100A8 as potential salivary biomarker of oral squamous cell carcinoma using nanoLC-MS/MS. Clin Chim Acta 436: 121-129, 2014.
42. Natarajan J, Hunter K, Mutalik VS and Radhakrishnan R: Overexpression of S100A4 as a biomarker of metastasis and recurrence in oral squamous cell carcinoma. J Appl Oral Sci 22: 426-433, 2014.
43. Kumar M, Srivastava G, Kaur J, Assi J, Alyass A, Leong I, MacMillan C, Witterick I, Shukla NK, Thakar A, *et al*: Prognostic significance of cytoplasmic S100A2 overexpression in oral cancer patients. J Transl Med 13: 8, 2015.
44. Sapkota D, Bruland O, Parajuli H, Osman TA, Teh MT, Johannessen AC and Costea DE: S100A16 promotes differentiation and contributes to a less aggressive tumor phenotype in oral squamous cell carcinoma. BMC Cancer 15: 631, 2015.
45. Sood A, Mishra D, Kharbanda OP, Chauhan SS, Gupta SD, Deo SSV, Yadav R, Ralhan R, Kumawat R and Kaur H: Role of S100 A7 as a diagnostic biomarker in oral potentially malignant disorders and oral cancer. J Oral Maxillofac Pathol 26: 166-172, 2022.
46. Pandey S, Osman TA, Sharma S, Vallenari EM, Shahdadfar A, Pun CB, Gautam DK, Uhlin-Hansen L, Rikardsen O, Johannessen AC, *et al*: Loss of S100A14 expression at the tumor-invading front correlates with poor differentiation and worse prognosis in oral squamous cell carcinoma. Head Neck 42: 2088-2098, 2020.
47. Tyszkiewicz T, Jarzab M, Szymczyk C, Kowal M, Krajewska J, Jaworska M, Fraczek M, Krajewska A, Hadas E, Swierniak M, *et al*: Epidermal differentiation complex (locus 1q21) gene expression in head and neck cancer and normal mucosa. Folia Histochem Cytobiol 52: 79-89, 2014.
48. Qu ZF, Ma H, Duan XF, Wu R and Zou Y: The expression and significance of S100A9 in oral squamous cell carcinoma. Lin Chuang Er Bi Yan Hou Tou Jing Wai Ke Za Zhi 31: 219-222, 2017 (In Chinese).
49. Suzuki S, Honda K, Nanjo H, Iikawa N, Tsuji T, Kawasaki Y, Yamazaki K, Sato T, Saito H, Shiina K and Ishikawa K: CD147 expression correlates with lymph node metastasis in T1-T2 squamous cell carcinoma of the tongue. Oncol Lett 14: 4670-4676, 2017.
50. Argyris PP, Slama ZM, Ross KF, Khammanivong A and Herzberg MC: Calprotectin and the initiation and progression of head and neck cancer. J Dent Res 97: 674-682, 2018.
51. Mints M, Landin D, Nasman A, Mirzaie L, Ursu RG, Zupancic M, Marklund L, Dalianis T, Munck-Wikland E and Ramqvist T: Tumour inflammation signature and expression of S100A12 and HLA class I improve survival in HPV-negative hypopharyngeal cancer. Sci Rep 11: 1782, 2021.
52. Ohnishi Y, Yasui H, Kakudo K and Nozaki M: Lapatinib-resistant cancer cells possessing epithelial cancer stem cell properties develop sensitivity during sphere formation by activation of the ErbB/AKT/cyclin D2 pathway. Oncol Rep 36: 3058-3064, 2016.
53. Ohnishi Y, Yasui H, Nozaki M and Nakajima M: Molecularly-targeted therapy for the oral cancer stem cells. Jpn Dent Sci Rev 54: 88-103, 2018.
54. Moriyama-Kita M, Endo Y, Yonemura Y, Heizmann CW, Miyamori H, Sato H, Yamamoto E and Sasaki T: S100A4 regulates E-cadherin expression in oral squamous cell carcinoma. Cancer Lett 230: 211-218, 2005.
55. Sun Q, Wang R, Wang Y, Luo J, Wang P and Cheng B: Notch1 is a potential therapeutic target for the treatment of human hepatitis B virus X protein-associated hepatocellular carcinoma. Oncol Rep 31: 933-939, 2014.
56. Khammanivong A, Wang C, Sorenson BS, Ross KF and Herzberg MC: S100A8/A9 (calprotectin) negatively regulates G2/M cell cycle progression and growth of squamous cell carcinoma. PLoS One 8: e69395, 2013.
57. Cheng S, Zhang X, Huang N, Qiu Q, Jin Y and Jiang D: Down-regulation of S100A9 inhibits osteosarcoma cell growth through inactivating MAPK and NF-κB signaling pathways. BMC Cancer 16: 253, 2016.
58. Li R, Li W, He F, Zhang M, Luo H and Tang H: Systematic screening identifies a TEAD4-S100A13 axis modulating cisplatin sensitivity of oral squamous cell carcinoma cells. J Oral Pathol Med 50: 882-890, 2021.
59. Takeuchi S, Kasamatsu A, Yamatoji M, Nakashima D, Endo-Sakamoto Y, Koide N, Takahara T, Shimizu T, Iyoda M, Ogawara K, *et al*: TEAD4-YAP interaction regulates tumoral growth by controlling cell-cycle arrest at the G1 phase. Biochem Biophys Res Commun 486: 385-390, 2017.
60. Sivadasan P, Gupta MK, Sathe G, Sudheendra HV, Sunny SP, Renu D, Hari PS, Gowda H, Suresh A, Kuriakose MA and Sirdeshmukh R: Salivary proteins from dysplastic leukoplakia and oral squamous cell carcinoma and their potential for early detection. J Proteomics 212: 103574, 2020.
61. Shan J, Sun Z, Yang J, Xu J, Shi W, Wu Y, Fan Y and Li H: Discovery and preclinical validation of proteomic biomarkers in saliva for early detection of oral squamous cell carcinomas. Oral Dis 25: 97-107, 2019.
62. Cheng YS, Rees T and Wright J: A review of research on salivary biomarkers for oral Clin Transl Med 3: 3, 2014.
63. Xu J, Gross N, Zang Y, Cao S, Yang F, Yang Z, Yu W, Lei D and Pan X: Overexpression of S100A4 predicts migration, invasion, and poor prognosis of hypopharyngeal squamous cell carcinoma. Mol Diagn Ther 23: 407-417, 2019.
64. Cheng LH, Hung KF, Huang TF, Hsieh HP, Wang SY, Huang CY and Lo JF: Attenuation of cancer-initiating cells stemness properties by abrogating S100A4 calcium binding ability in head and neck cancers. Oncotarget 7: 78946-78957, 2016.
65. Wu P, Quan H, Kang J, He J, Luo S, Xie C, Xu J, Tang Y and Zhao S: Downregulation of Calcium-Binding Protein S100A9 Inhibits Hypopharyngeal Cancer Cell Proliferation and Invasion Ability Through Inactivation of NF-kappaB Signaling. Oncol Res 25: 1479-1488, 2017.
66. Wang C, Lin C, Tao Q, Zhao S, Liu H and Li L: Evaluation of calcium-binding protein A11 promotes the carcinogenesis of hypopharygeal squamous cell carcinoma via the PI3K/AKT signaling pathway. Am J Transl Res 11: 3472-3480, 2019.
67. Hu W, Tao Z, Zhou Q, Zhao D, Gu L, Zhu S and Chen J: Effects of S100 calcium-binding protein A8 (S100A8) and S100 calcium-binding protein A9 (S100A9) on matrix metalloproteinase (MMP) expression in nasopharyngeal carcinoma CNE-2 cells. Transl Cancer Res 10: 1874-1884, 2021.

68. Isaksen B and Fagerhol MK: Calprotectin inhibits matrix metalloproteinases by sequestration of zinc. Mol Pathol 54: 289-292, 2001.
69. Carlsson H, Yhr M, Petersson S, Collins N, Polyak K and Enerback C: Psoriasin (S100A7) and calgranulin-B (S100A9) induction is dependent on reactive oxygen species and is downregulated by Bcl-2 and antioxidants. Cancer Biol Ther 4: 998-1005, 2005.
70. Ichikawa M, Williams R, Wang L, Vogl T and Srikrishna G: S100A8/A9 activate key genes and pathways in colon tumor progression. Mol Cancer Res 9: 133-148, 2011.
71. Xiao Z, Li M, Li G, Fu Y, Peng F, Chen Y and Chen Z: Proteomic characterization reveals a molecular portrait of nasopharyngeal carcinoma differentiation. J Cancer 8: 570-577, 2017.
72. Li A, Shi D, Xu B, Wang J, Tang YL, Xiao W, Shen G, Deng W and Zhao C: S100A6 promotes cell proliferation in human nasopharyngeal carcinoma via the p38/MAPK signaling pathway. Mol Carcinog 56: 972-984, 2017.
73. Wen L, Ding Y, Chen X, Tian K, Li D, Liang K and Yue B: Influences of S100A8 and S100A9 on proliferation of nasopharyngeal carcinoma cells through PI3K/Akt signaling pathway. Biomed Res Int 2021: 9917365, 2021.
74. Wang C, Wang X, Han A, Wang Y and Jiang H: Proof-of-concept study investigating the role of S100P-RAGE in nasopharyngeal carcinoma. Exp Ther Med 21: 470, 2021.
75. Liu L, Liu S, Deng P, Liang Y, Xiao R, Tang LQ, Chen J, Chen QY, Guan P, Yan SM, *et al*: Targeting the IRAK1-S100A9 axis overcomes resistance to paclitaxel in nasopharyngeal carcinoma. Cancer Res 81: 1413-1425, 2021.
76. Meng DF, Sun R, Liu GY, Peng LX, Zheng LS, Xie P, Lin ST, Mei Y, Qiang YY, Li CZ, *et al*: S100A14 suppresses metastasis of nasopharyngeal carcinoma by inhibition of NF-kB signaling through degradation of IRAK1. Oncogene 39: 5307-5322, 2020.
77. Png YT, Yang AZY, Lee MY, Chua MJM and Lim CM: The role of NK cells in EBV infection and EBV-associated NPC. Viruses 13: 300, 2021.
78. Lin Z, Deng L, Ji J, Cheng C, Wan X, Jiang R, Tang J, Zhuo H, Sun B and Chen Y: S100A4 hypomethylation affects epithelial-mesenchymal transition partially induced by LMP2A in nasopharyngeal carcinoma. Mol Carcinog 55: 1467-1476, 2016.
79. Hino R, Uozaki H, Murakami N, Ushiku T, Shinozaki A, Ishikawa S, Morikawa T, Nakaya T, Sakatani T, Takada K and Fukayama M: Activation of DNA methyltransferase 1 by EBV latent membrane protein 2A leads to promoter hypermethylation of PTEN gene in gastric carcinoma. Cancer Res 69: 2766-2774, 2009.
80. Chakladar J, Li WT, Bouvet M, Chang EY, Wang-Rodriguez J and Ongkeko WM: Papillary thyroid carcinoma variants are characterized by Co-dysregulation of immune and cancer associated genes. Cancers (Basel) 11: 1179, 2019.
81. Martinez-Aguilar J, Clifton-Bligh R and Molloy MP: A multiplexed, targeted mass spectrometry assay of the S100 protein family uncovers the isoform-specific expression in thyroid tumours. BMC Cancer 15: 199, 2015.
82. Nipp M, Elsner M, Balluff B, Meding S, Sarioglu H, Ueffing M, Rauser S, Unger K, Höfler H, Walch A and Zitzelsberger H: S100-A10, thioredoxin, and S100-A6 as biomarkers of papillary thyroid carcinoma with lymph node metastasis identified by MALDI imaging. J Mol Med (Berl) 90: 163-174, 2012.
83. Zhao M, Wang KJ, Tan Z, Zheng CM, Liang Z and Zhao JQ: Identification of potential therapeutic targets for papillary thyroid carcinoma by bioinformatics analysis. Oncol Lett 11: 51-58, 2016.
84. Wang X, Sun Z, Tian W, Piao C, Xie X, Zang J, Peng S, Yu X and Wang Y: S100A12 is a promising biomarker in papillary thyroid cancer. Sci Rep 10: 1724, 2020.
85. Wang G, Li HN, Cui XQ, Xu T, Dong ML, Li SY and Li XR: S100A1 is a potential biomarker for papillary thyroid carcinoma diagnosis and prognosis. J Cancer 12: 5760-5771, 2021.
86. Huang P and Xue J: Long non-coding RNA FOXD2-AS1 regulates the tumorigenesis and progression of breast cancer via the S100 calcium binding protein A1/Hippo signaling pathway. Int J Mol Med 46: 1477-1489, 2020.
87. Tian J and Luo B: Identification of three prognosis-related differentially expressed lncRNAs driven by copy number variation in thyroid cancer. J Immunol Res 2022: 9203796, 2022.
88. Xia F, Chen Y, Jiang B, Du X, Peng Y, Wang W, Huang W, Feng T and Li X: Long Noncoding RNA HOXA-AS2 promotes papillary thyroid cancer progression by regulating miR-520c-3p/S100A4 pathway. Cell Physiol Biochem 50: 1659-1672, 2018.
89. Cheon MG, Son YW, Lee JH, Jang HH and Chung YS: Mts1 Up-regulation is associated with aggressive pathological features in thyroid cancer. Cancer Genomics Proteomics 16: 369-376, 2019.
90. Wang Y, Ye H, Yang Y, Li J, Cen A and Zhao L: microRNA-181a promotes the oncogene S100A2 and enhances papillary thyroid carcinoma growth by mediating the expression of histone demethylase KDM5C. J Endocrinol Invest 45: 17-28, 2022.
91. Economopoulou P, Kotsantis I and Psyrri A: Special issue about head and neck cancers: HPV positive cancers. Int J Mol Sci 21: 3388, 2020.
92. Tokuzen N, Nakashiro KI, Tojo S, Goda H, Kuribayashi N and Uchida D: Human papillomavirus-16 infection and p16 expression in oral squamous cell carcinoma. Oncol Lett 22: 528, 2021.
93. Mork J, Lie AK, Glattre E, Hallmans G, Jellum E, Koskela P, Møller B, Pukkala E, Schiller JT, Youngman L, *et al*: Human papillomavirus infection as a risk factor for squamous-cell carcinoma of the head and neck. N Engl J Med 344: 1125-1131, 2001.
94. Ang KK, Harris J, Wheeler R, Weber R, Rosenthal DI, Nguyen-Tân PF, Westra WH, Chung CH, Jordan RC, Lu C, *et al*: Human papillomavirus and survival of patients with oropharyngeal cancer. N Engl J Med 363: 24-35, 2010.
95. Wuerdemann N, Wittekindt C, Sharma SJ, Prigge ES, Reuschenbach M, Gattenlöhner S, Klussmann JP and Wagner S: Risk factors for overall survival outcome in surgically treated human papillomavirus-negative and positive patients with oropharyngeal cancer. Oncol Res Treat 40: 320-327, 2017.
96. Granata R, Miceli R, Orlandi E, Perrone F, Cortelazzi B, Franceschini M, Locati LD, Bossi P, Bergamini C, Mirabile A, *et al*: Tumor stage, human papillomavirus and smoking status affect the survival of patients with oropharyngeal cancer: An Italian validation study. Ann Oncol 23: 1832-1837, 2012.
97. Paver EC, Currie AM, Gupta R and Dahlstrom JE: Human papilloma virus related squamous cell carcinomas of the head and neck: Diagnosis, clinical implications and detection of HPV. Pathology 52: 179-191, 2020.
98. Lo WY, Lai CC, Hua CH, Tsai MH, Huang SY, Tsai CH and Tsai FJ: S100A8 is identified as a biomarker of HPV18-infected oral squamous cell carcinomas by suppression subtraction hybridization, clinical proteomics analysis, and immunohistochemistry staining. J Proteome Res 6: 2143-2151, 2007.
99. Liu S, Xie Y, Luo W, Dou Y, Xiong H, Xiao Z and Zhang XL: PE_PGRS31-S100A9 interaction promotes mycobacterial survival in macrophages through the regulation of NF-κB-TNF-α signaling and arachidonic acid metabolism. Front Microbiol 11: 845, 2020.
100. Khammanivong A, Sorenson BS, Ross KF, Dickerson EB, Hasina R, Lingen MW and Herzberg MC: Involvement of calprotectin (S100A8/A9) in molecular pathways associated with HNSCC. Oncotarget 7: 14029-14047, 2016.
101. Gyöngyösi E, Szalmás A, Ferenczi A, Póliska S, Kónya J and Veress G: Transcriptional regulation of genes involved in keratinocyte differentiation by human papillomavirus 16 oncoproteins. Arch Virol 160: 389-398, 2015.
102. Alghamdi MA, Al-Eitan LN, Tarkhan AH and Al-Qarqaz FA: Global gene methylation profiling of common warts caused by human papillomaviruses infection. Saudi J Biol Sci 28: 612-622, 2021.
103. Han S, Locke AK, Oaks LA, Cheng YL and Coté GL: Nanoparticle-based assay for detection of S100P mRNA using surface-enhanced Raman spectroscopy. J Biomed Opt 24: 1-9, 2019.
104. Jiao X, Zhang H, Xu X, Yu Y, Zhang H, Zhang J, Ning L, Hao F, Liu X, Niu M, *et al*: S100A4 knockout sensitizes anaplastic thyroid carcinoma cells harboring BRAFV600E/Mt to Vemurafenib. Cell Physiol Biochem 49: 1143-1162, 2018.
105. Björk P, Björk A, Vogl T, Stenström M, Liberg D, Olsson A, Roth J, Ivars F and Leanderson T: Identification of human S100A9 as a novel target for treatment of autoimmune disease via binding to quinoline-3-carboxamides. PLoS Biol 7: e97, 2009.
106. Mouta Carreira C, LaVallee TM, Tarantini F, Jackson A, Lathrop JT, Hampton B, Burgess WH and Maciag T: S100A13 is involved in the regulation of fibroblast growth factor-1 and p40 synaptotagmin-1 release in vitro. J Biol Chem 273: 22224-22231, 1998.