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Research Paper
Head and Neck Oncology

High level of *CD10* expression is associated with poor overall survival in patients with head and neck cancer

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Abstract. *CD10* is a common zinc-dependent metalloid protease that is expressed in numerous tissues, including malignant cells. Genomic alterations of *CD10* are frequently observed in haematopoietic and non-haematopoietic tumours. In the present study, we analysed the *CD10* expression in head and neck squamous cell carcinoma (HNSCC) and its association with tumour prognosis using bioinformatic analysis and explored the potential of a *CD10*-driven signalling pathway in a tumour-immune microenvironment. Briefly, data mining analysis showed strengthened *CD10* expression in HNSCC patients. High *CD10* expression was associated with unfavourable overall survival (OS) and recurrence-free survival (RFS). In addition, the correlation between *CD10* expression and interleukin (*IL*)-*6*/*IL*-8-mediated M1 macrophage activity could potentially explain the poor prognosis of HNSCC. Among 692 genes co-expressed with *CD10* in HNSCC, *Rap1* signalling pathway, regulation of actin cytoskeleton, protein digestion and absorption, proteoglycans in cancer, *PI3K-Akt* signalling pathway, focal adhesion and extracellular matrix–receptor interaction were the candidate signalling pathways driven by the *CD10* gene. Further investigation of immune-associated signalling pathways regulated by CD10 may be beneficial to improve the prognosis of HNSCC patients by immunotherapy.

Keywords: CD10; Head and neck cancer; Macrophage; Tumour microenvironment; Immune.

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Cancer is the leading cause of global death and the single most important barrier to increased life expectancy.[1] Despite advances in early detection, diagnosis, and treatment, the overall survival rate in patients with head and neck squamous cell carcinoma (HNSCC) remains poor.[2] High loco-regional recurrence, lymph node metastasis, and high-degree chemo-radio-resistance are considered to be the leading causes of poor prognosis in patients with HNSCC.[3,4] Thus, there is an urgent need to explore novel prognostic indicators, which could in turn improve treatment for patients with HNSCC.

CD10 is a common zinc-dependent metalloid protease or membrane metalloendopeptidase located on the surface

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of normal (haemopoietic and non-haemopoietic epithelial cells) and cancer cells,[5,6] which can inactivate various signalling peptides through enzymatic activity. Previous studies have shown that *CD10* expression is associated with tumour size, histological grade, vascular invasion, and overall survival rate within solid tumours.[7,8] In HNSCC, *CD10* has been associated with therapeutic resistance and cancer-stem-cell (CSC)-like properties and has been identified as a potential indicator of poor prognosis.[8,9] A CSC is a type of cell that possess unlimited self-renewal potential. Studies have suggested that CSCs may induce tumour regrowth and promote metastasis in HNSCC patients, if not eliminated by therapy.[10–12] Moreover, due to their ability to modulate and shape immune responses, CSC could lead to immunotherapy tolerance in head and neck cancer.[13] Thus, so far, no studies have investigated the relationship between CD10 and immune status in head and neck cancer and its impact on prognosis.

In this study, we explored the effect of *CD10* dysregulation on survival results in HNSCC patients by using bioinformatics analysis. Moreover, we analysed its correlation with macrophages in the tumour microenvironment, as well as the potential *CD10*-driven signalling pathway.

Materials and methods

CD10 expression within different types of malignancies

The expression of *CD10* in solid tumours and healthy tissues was analysed using data from The Cancer Genome Atlas (TCGA). Data analysis was performed using FireBrowse (http://firebrowse.org/).

CD10 expression analysis in different HNSCC cohorts

The gene expression profile data GSE58911 (15 healthy and 15 tumour tissues) and GSE107591 (23 healthy and 24 tumour tissues) were obtained from Gene Expression Omnibus (GEO). Limma package in R/Bioconductor software was applied to perform the normalization and log-2 conversion for the matrix data of each GEO dataset. *CD10* expression in each microarray was screened by the limma package.

Bioinformatic analysis

The level-three data of patients with primary HNSCC in TCGA-HNSCC were obtained via UCSC Xena browser (https://xenabrowser.net/). *CD10* mRNA expression of TCGA-HNSC data was extracted using the UCSC Xena browser. Receiver operating characteristic (ROC) curve analysis was used to determine the cut-off expression level of CD10. Kaplan–Meier curves of overall survival (OS) and recurrence-free survival (RFS) after initial therapy were generated by GraphPad Prism v7.00.

The correlation of mRNA expression and immune cell infiltration

A correlation analysis was performed for the mRNA expression data of *CD10* in TCGA HNSCC tumour samples and tumour infiltration of six immune cell types (CD8+ T cells, CD4+ T cells, B cells, neutrophils, dendritic cells and macrophages) using the online tool TIMER (Tumor IMmune Estimation Resource; https://cistrome.shinyapps.io/timer/). Furthermore, the associations (Spearman's correlation) between CD10 and IL-6 or IL-8 in HNSCC were analysed using TIMER.

For macrophage infiltration and mRNA expression of the 3 genes (*CD10, IL-6* and *IL-8*) in the TCGA HNSCC dataset, quanTIseq_lsei_TIL10 values for M1 and M2 macrophages were obtained from The Cancer Immunome Database (TCIA, https://tcia.at/home).[14] These values were then merged with RNA-seq data of the tumour samples. Overall, TCIA scores and RNA-seq data were available for 520 HNSCC samples.

Gene co-expression network analysis using cBioPortal for Cancer Genomics and ClueGo

The genes co-expressed with *CD10* in HNSCC ($|\text{Spearman's } r| \geq 0.4$) were identified using cBioPortal for Cancer Genomics (http://www.cbioportal.org/). The genes were then loaded into ClueGo in Cytoscape for Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis. Only pathways with $P < 0.05$ were included.

Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics for Windows, version 19.0 (IBM Corp., Armonk, NY, USA). The association between *CD10* mRNA expression and the clinicopathological features was evaluated using χ^2 tests. ROC curves for recurrence and death detection were constructed; the optimal cut-off value of *CD10* expression was determined based on the Youden index. A log-rank test was performed to assess the difference between the survival curves. Welch's *t*-test was conducted to compare *CD10* mRNA expression between normal and tumour tissues. A $P < 0.05$ was considered to be statistically significant.

Results

CD10 expression analysis in different cancer types

To determine changes in *CD10* gene expression in malignant tumours, we compared the expression of *CD10* in each solid tumour and their corresponding normal tissue of 37 different cancer types in TCGA. Data mining analysis by FireBrowse indicated that *CD10* gene expression was significantly upregulated in 25% (9/37) of solid malignancies compared with paired normal tissues. In addition, our data indicated that the expression was over twofold higher in colon adenocarcinoma, colorectal adenocarcinoma, oesophageal cancer, skin cutaneous melanoma, and HNSCC (Fig. 1).

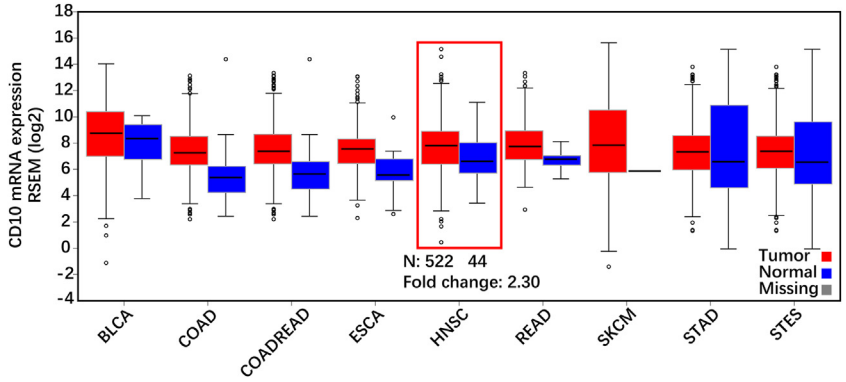

Fig. 1. CD10 expression in different solid tumour types and paired healthy tissues. (BLCA, bladder carcinoma; COAD, colon adenocarcinoma; COADREAD, colorectal adenocarcinoma; ESCA, oesophageal cancer; READ, rectum adenocarcinoma; SKCM, skin cutaneous melanoma; STAD, stomach adenocarcinoma; STES, stomach and oesophageal carcinoma).

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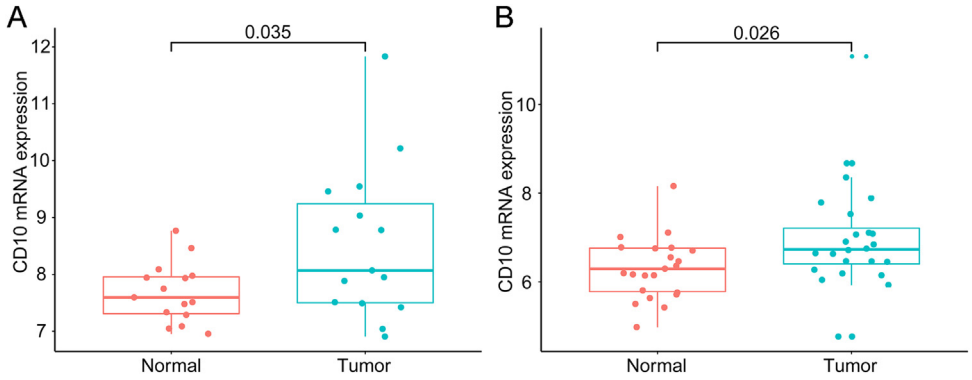

Fig. 2. Higher expression of CD10 mRNA was observed in two head and neck squamous cell carcinoma cohorts from the GSE database.

Association of *CD10* expression with clinicopathological parameters

To further determine the *CD10* elevated expression in HNSCC, GEO human microarrays were extracted and analysed. *CD10* upregulation in tumour tissue of the two separate cohorts was compared to the normal tissue (Fig. 2A and B).

Among the total of 517 HNSCC patients enrolled for *CD10* gene expression analysis, 342 cases were classified as the high expression group and 175 cases as the low expression group. After removing the null data that had not been subjected to the p16 test, we found that *CD10* expression was significantly associated with human papillomavirus (HPV) and survival status, indicating a larger percentage of HPV-negative patients in the high *CD10* expression group compared with the low *CD10* expression group (61/68, 89.7% vs 11/42, 26.2%, $P < 0.01$) (Table 1).

Tumour staging and patient prognosis are closely related to the prognosis of HNSCC patients.[11] In this study, we analysed whether the *CD10* expression level

Table 1. The association between CD10 expression, the demographic and clinicopathological parameters in patients with primary head and neck squamous cell carcinoma in The Cancer Genome Atlas.

Parameters		CD10 expression	CD10 expression	χ^2	*P*
		High ($n = 342$)	Low ($n = 175$)		
Age (mean ± SD)		61.44 ± 11.83	59.97 ± 11.82		0.1817
Gender	Female	97	38	2.65	0.1034
	Male	245	137		
Smoking history	1	73	43	0.78	0.3765
	2/3/4/5	262	127		
	Null	7	5		
HPV status by p16 testing	Negative	61	11	46.32	<0.0001
	Positive	7	31		
	Null	274	133		
Clinical stage	I/II	80	37	0.32	0.5705
	III/IV	253	133		
	Discrepancy + null	9	5		
Pathologic stage	I/II	65	35	0.57	0.4515
	III/IV	238	107		
	Discrepancy + null	39	33		
Recurrence status	No	211	118	<0.0001	0.9974
	Yes	68	38		
	Null	63	19		
Radiation therapy	No	103	55	0.03	0.8607
	Yes	186	103		
	Null	53	17		
Living status	Living	182	115	7.40	0.0065
	Dead	160	60		

HPV, human papillomavirus; SD, standard deviation.
The data of smoking history comes from the TCGA database, which is classified into four levels without unit in itself. The content of the smoking history classification is described in detail below.
Lifelong Non-smoker (less than 100 cigarettes smoked in Lifetime) = 1.
Current smoker (includes daily smokers and non-daily smokers or occasional smokers) = 2.
Current reformed smoker for > 15 years (greater than 15 years) = 3.
Current reformed smoker for ≤15 years (less than or equal to 15 years) = 4.
Current reformed smoker, duration not specified = 5.
Smoker at Diagnosis = 6.
Smoking History not documented = 7.

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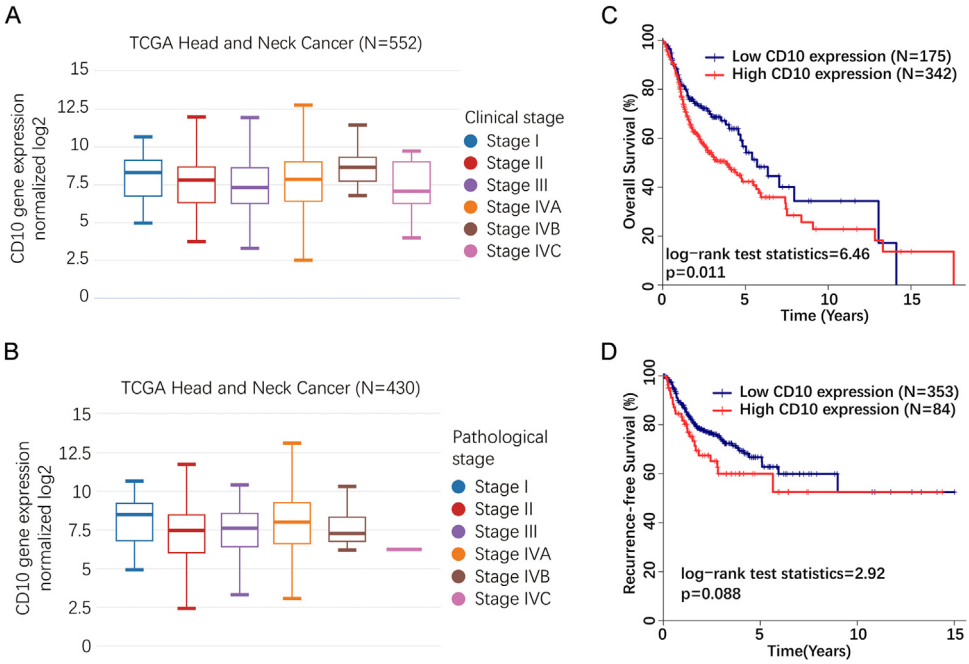

Fig. 3. The association between CD10 expression and survival in head and neck squamous cell carcinoma (HNSCC) from the GSE database. (A, B) CD10 expression in different clinical and pathological stages of HNSCC patients. (C, D) The association between CD10 expression and overall survival (C, cut-off level = 6.8905) or recurrence-free survival (D, cut-off level = 9.2064) in HNSCC patients. TCGA, The Cancer Genome Atlas.

Table 2. Univariate and multivariate analysis of overall survival (OS) and recurrence-free survival (RFS) in patients with primary head and neck squamous cell carcinoma in The Cancer Genome Atlas.

Parameters	Univariate analysis				Multivariate analysis			
	P	HR	95% CI		*P*	HR	95% CI	
			Lower	Upper			Lower	Upper
OS								
Age >65 years vs ≤65 years	0.0134	1.4077	1.0736	1.8459	0.0468	1.3536	1.0043	1.8243
Female vs male	0.0401	1.3493	1.0136	1.7961	0.1298	1.2744	0.9313	1.7440
Smoking history 2/3/4/5 vs 1	0.4978	1.1233	0.8026	1.5723				
HPV status by p16 testing Positive vs negative	0.0498	0.3453	0.1193	0.9993	0.1369	0.2219	0.0305	1.6133
Clinical stage III/IV vs I/II	0.3070	1.1838	0.8564	1.6364				
Pathological stage III/IV vs I/II	0.0035	1.7539	1.2025	2.5582	0.0013	1.8727	1.2772	2.7459
CD10 expression high vs low	0.0116	1.4669	1.0896	1.9748	0.0927	1.3257	0.9544	1.8415
RFS								
Age >65 years vs ≤65 years	0.2082	1.2842	0.8699	1.8958				
Female vs male	0.5553	0.8717	0.5523	1.3758				
Smoking history 2/3/4/5 vs 1	0.9963	0.9989	0.6378	1.5646				
HPV status by p16 testing Positive vs negative	0.3843	0.6059	0.1960	1.8734				
Clinical stage III/IV vs I/II	0.3364	1.2778	0.7751	2.1065				
Pathological stage III/IV vs I/II	0.0093	2.2541	1.2220	4.1579	0.0104	2.2290	1.2071	4.1160
CD10 expression high vs low	0.0725	1.5004	0.9636	2.3363	0.1211	1.4501	0.9064	2.3201

CI, confidence interval; HR, hazard ratio; NA, not applicable.
The data of smoking history comes from the TCGA database, which is classified into four levels without unit in itself. The content of the smoking history classification is described in detail below.
Lifelong Non-smoker (less than 100 cigarettes smoked in Lifetime) = 1.
Current smoker (includes daily smokers and non-daily smokers or occasional smokers) = 2.
Current reformed smoker for > 15 years (greater than 15 years) = 3.
Current reformed smoker for ≤15 years (less than or equal to 15 years) = 4.
Current reformed smoker, duration not specified = 5.
Smoker at Diagnosis = 6.
Smoking History not documented = 7.

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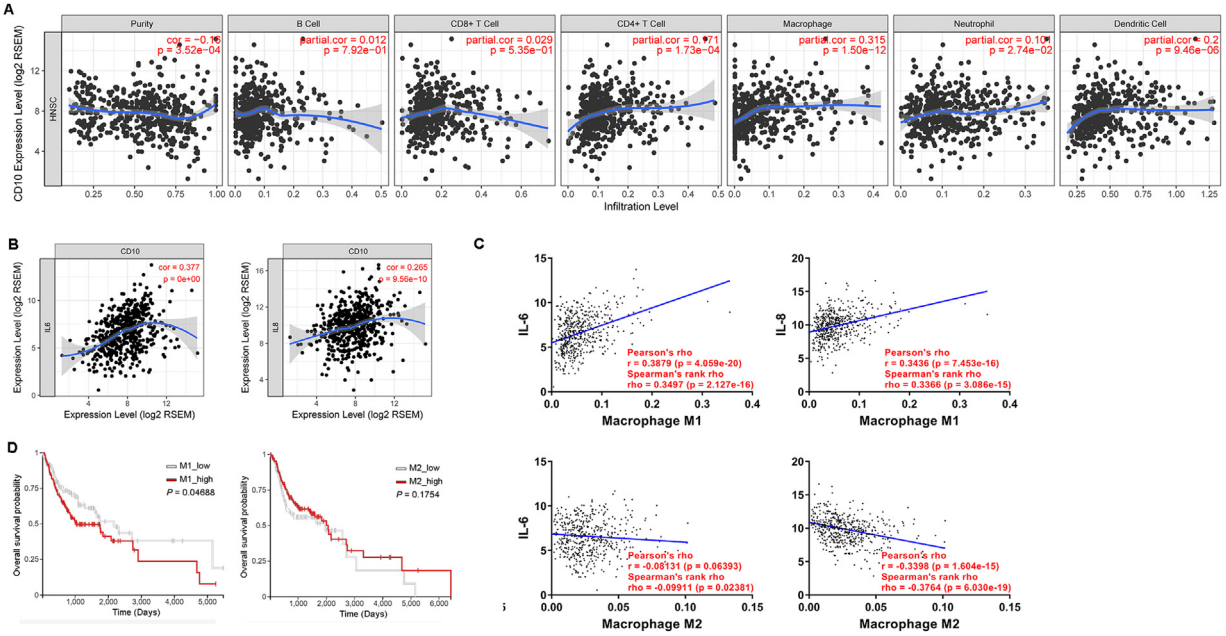

Fig. 4. Association between mRNA expression of three genes and macrophage infiltration in head and neck squamous cell carcinoma (HNSCC) tumours. (A) Immune cell landscape of HNSCC patients compared with CD10 gene expression. (B) Correlation between CD10 mRNA and mRNA levels for interleukin (IL)-6, IL-8 from The Cancer Genome Atlas (TCGA) HNSCC dataset. (C) Scatter plots show the correlation between CD10, IL-6, and IL-8 mRNA expression (log-2 scale) and M1 or M2 macrophages infiltration scores (obtained from The Cancer Immunome (TCIA) database) in tumour samples from the TCGA HNSCC dataset. Each circle represents a single tumour sample. The regression lines are shown in blue. (D) Kaplan–Meier plots of overall survival between M1/M2 low and high infiltrated HNSCC patients ($n = 520$) of the TCGA HNSCC dataset. Categorized Pearson's product-moment correlation of immune cell landscape of HNSCC compared with TCGA gene expression of CD10, IL-6 and IL-8 (TIMER). r, categorized Pearson's correlation coefficient; (−−), −0.5 to −0.3, weak negative association; (−−−), −1.0 to −0.5, strong negative association; (+), +0.1 to 0.3, little association; (−), −0.3 to 0.1, little association; (++) +0.3 to +0.5, weak positive association; (+++), +0.5 to +1.0, strong positive association. RSEM, RNA-seq by expectation-maximization.

was related to clinical and pathological staging. Interestingly, no association between *CD10* expression and clinical or pathological stages was found (Table 1 and Fig. 3A and B).

Association of *CD10* expression with patient survival

It has been reported that patients with different expression levels of *CD10* have different survival status.[15–17] We found a reduced survival status in the high *CD10* expression group compared with low *CD10* expression (182/342, 53.2s% vs 115/175, 65.7%, $P < 0.01$; Table 1). Moreover, HNSCC patients with high *CD10* expression had significantly poorer overall survival compared with patients with low *CD10* expression ($P = 0.011$, Fig. 3C). Although the analysis for recurrence-free survival (RFS) showed a trend to worse survival for high levels of CD-10, this was not statistically significant ($P = 0.088$, Fig. 3D).

Multivariate and univariate analyses were performed using the HNSCC patients' data from TCGA. Univariate analysis revealed that age older than 65 years, gender, HPV status, pathological staging, and *CD10* expression levels were all associated with OS. Meanwhile, the pathological staging was associated with RFS. According to multivariate analysis, age older than 65 years and pathological staging were associated with survival, while *CD10* expression was not an independent prognostic factor for poor OS and RFS (Table 2).

CD10 expression is associated with IL-6/8-driven tumour immunology

Due to their importance in cancer in general, tumour immunology and cancer immunotherapy in particular have been in the focus of theoretical investigators.[18] Next, we investigated the immune status of tumour microenvironment based on the different *CD10* expressions in HNSCC. We found that macrophages were closely associated with the expression of *CD10* (Fig. 4A). Tumour-associated macrophages (TAMs) can promote cancer progression and metastasis through the release of a variety of cytokines, such as *IL-6* and *IL-8*[19]. Thus, we explored the correlation between CD10 and macrophage-associated IL-6 and IL-8. Our correlation analysis revealed a close association between *IL-6 or IL-8* and CD10 (Fig. 4B).

To gain a better understanding of the tumour-immune microenvironment, we analysed associations between CD10/IL-6/IL-8 and polarized M1 and M2 macrophages from the TCIA database. The comparison revealed association between CD10 and M1 or M2 macrophages. Nevertheless, both IL-6 and IL-8 showed weak positive associations with M1 macrophages, and low or weak negative associations with M2 macrophages (Fig. 4C). Besides, we assessed the prognostic association of M1 and M2 macrophages in HNSCC by stratifying the population for M1 and M2 high/low. As expected, high numbers of M1 macrophages predicted worse outcomes compared with M2 macrophages (Fig. 4D). These findings suggest that CD10 might be correlated with macrophages in the tumour microenvironment, possibly through IL-6/IL-8-mediated M1 macrophages.

CD10 was involved in different signalling pathways in HNSCC

Using data mining and cBioPortal for Cancer Genomics, we identified 691 genes

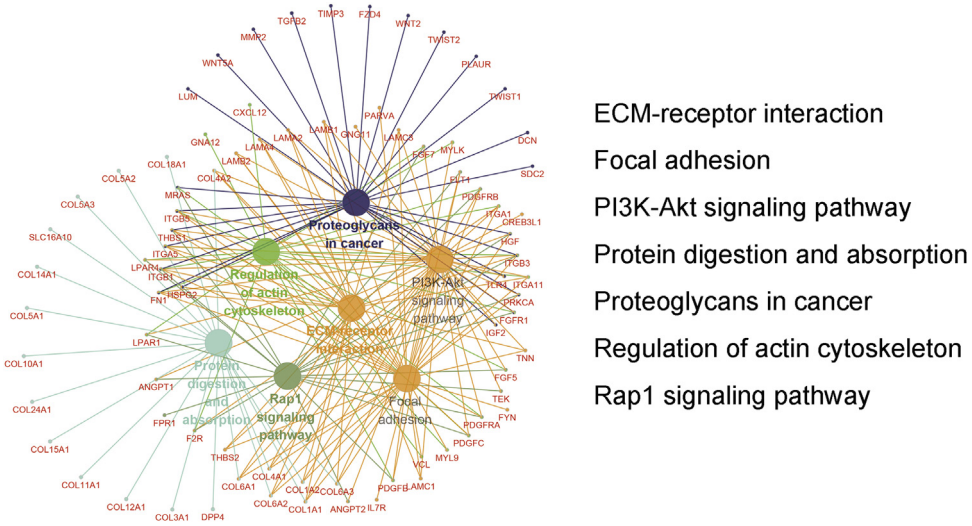

Fig. 5. Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis of the genes co-expressed with CD10 in head and neck squamous cell carcinoma. ECM, extracellular matrix.

that were co-expressed with *CD10* in HNSCC (|Spearman's $r| \leq 0.4$) (Supplementary Table S1). To further investigate the possible signalling pathways of *CD10*, *CD10* co-expressed genes in HNSCC were subjected to KEGG pathway analysis. Results showed the genes were enriched in *Rap1* signalling pathway, regulation of actin cytoskeleton, protein digestion and absorption, proteoglycans in cancer, *PI3K-Akt* signalling pathway, focal adhesion and ECM-receptor interaction (Fig. 5 and Supplementary Table S2).

Discussion

Identification of new prognostic indicators is essential for the development of a more personalized treatment for cancer. In this study, we found that *CD10* expression based on data in TCGA-HNSCC was upregulated in HNSCC compared with normal tissues. Previous studies have indicated that aberrant *CD10* expression is a tumour-specific antigen of leukaemia cells; also, dysregulation of CD10 has been found in a variety of cancers, including gastric, lung, breast and colorectal cancer.[20] In patients with lung adenocarcinoma, *CD10* is considered an adverse prognostic factor.[21] Moreover, increased stromal *CD10* expression is significantly related to an increasing tumour grade in breast cancer.[16] In this study, we observed that high *CD10* expression predicted poor prognosis, thus suggesting that surface glycoprotein of the peptidase M13 family is an essential mechanism of dysregulated phosphoramidon in HNSCC.

Our data indicated a significantly higher proportion of HPV-negative cases in patients with high *CD10* expression compared with those with low *CD10* expression. Besides, patients with high *CD10* expression had a reduced survival status. Age, gender, and CD10 expression are probably co-variables linked to HPV which is a disease of younger males. Improved outcomes in HPV-positive HNSCC patients have been consistently reported. For example, studies have reported that patients with oropharyngeal carcinoma positive for HPV or p16 live longer after locoregional failure compared with those without HPV.[2,4,22] This may be due to fewer genetic alterations, increased sensitivity to therapy or enhanced antitumour immunity.[23,24] This suggests that *CD10* and HPV may have an antagonistic relationship in tumourigenesis and development, which in turn affects the prognosis of HNSCC patients.

Studies have suggested that tumour-associated inflammatory cells, especially macrophages, may enhance tumour progression.[25] *IL-6* is a potent pleiotropic cytokine produced by monocytes and macrophages involved in tumour progression and metastasis through *STAT3* signalling pathways.[26] M2 macrophage is a type of TAM that participates in the progression of colorectal cancer through the tumour necrosis factor alpha-mediated secretion of *IL-6* and *IL-8*.[19] Moreover, a recent study demonstrated that *CD10* +*GPR77*+ cancer-associated fibroblasts could induce cancer stem cell enrichment and chemoresistance by secreting *IL-6* and *IL-8*.[27] In this study, we found a high correlation between *CD10* expression and IL-6, which demonstrated that CD10-IL6-mediated tumour-associated macrophages activity might be the potential mechanism for the poor prognosis of HNSCC. Considering the negative correlation between *CD10* expression and HPV, HPV-positive HNSCC patients express lower levels of IL6 and IL8 than HPV-negative ones, which may be an underlying reason for the improved prognosis.

The exact mechanisms through which *CD10* participates in HNSCC remain unclear. One plausible explanation is through the enhanced accumulation of peptides that are cleaved by *CD10*, which leads to alteration of associated signalling pathways or biological behaviour of undifferentiated cells. *CD10* is also involved in the activation of focal adhesion kinase (FAK)-promoted cell adhesion.[28] Indeed, *CD10* in GPI-microdomains coimmunoprecipitate with Lyn and p85. This protein complex blocks PI3K (phosphatidylinositol 3-kinase) interaction with FAK by competitive binding, leading to decreased FAK phosphorylation and cell migration in the prostatic epithelial model.[29,30] By KEGG analysing of over 600 *CD10* co-expressed genes in HNSCC, we found that *Rap1* signalling pathway, regulation of actin cytoskeleton, protein digestion and absorption, proteoglycans in cancer, *PI3K-Akt* signalling pathway, focal adhesion and ECM-receptor interaction are potential signalling candidates driven by *CD10*. Therefore, future studies should

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further investigate the involvement of these pathways in HNSCC.

In conclusion, both genetic and epigenetic alterations contribute to dysregulated *CD10* in HNSCC. High *CD10* is an important prognostic factor for poor OS and RFS in HNSCC, though it is not an independent factor. Patients with high *CD10* expression are usually HPV-negative and have a poor prognosis. Besides, the *CD10* expression was associated with enhanced TAMs by *IL6* activity, which may be the potential mechanism for the poor prognosis of HNSCC. The altered pathways derived from KEGG analysis should be addressed by future studies.

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Ethical approval

Not applicable.

Patient consent

Not applicable.

Competing interests

None declared.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at https://doi.org/10.1016/j.ijom.2020.07.037.

References

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin* 2018;**68**:394–424.
2. Machiels JP, Lambrecht M, Hanin FX, Duprez T, Gregoire V, Schmitz S, et al. Advances in the management of squamous cell carcinoma of the head and neck. *F1000Prime Rep* 2014;**6**:44.
3. Mannelli G, Gallo O. Cancer stem cells hypothesis and stem cells in head and neck cancers. *Cancer Treat Rev* 2012;**38**:515–39.
4. Leeman JE, Li JG, Pei X, Venigalla P, Zumsteg ZS, Katsoulakis E, et al. Patterns of treatment failure and postrecurrence outcomes among patients with locally advanced head and neck squamous cell carcinoma after chemoradiotherapy using modern radiation techniques. *JAMA Oncol* 2017;**3**:1487–94.
5. Cutrona G, Tasso P, Dono M, Roncella S, Ulivi M, Carpaneto EM, et al. CD10 is a marker for cycling cells with propensity to apoptosis in childhood ALL. *Br J Cancer* 2002;**86**:1776–85.
6. Louhichi T, Saad H, Dhiab MB, Ziadi S, Trimeche M. Stromal CD10 expression in breast cancer correlates with tumor invasion and cancer stem cell phenotype. *BMC Cancer* 2018;**18**:49.
7. Kim HS, Kim GY, Kim YW, Park YK, Song JY, Lim SJ. Stromal CD10 expression and relationship to the E-cadherin/beta-catenin complex in breast carcinoma. *Histopathology* 2010;**56**:708–19.
8. Fukusumi T, Ishii H, Konno M, Yasui T, Nakahara S, Takenaka Y, et al. CD10 as a novel marker of therapeutic resistance and cancer stem cells in head and neck squamous cell carcinoma. *Br J Cancer* 2014;**111**:506–14.
9. Piattelli A, Fioroni M, Iezzi G, Perrotti V, Stellini E, Piattelli M, et al. CD10 expression in stromal cells of oral cavity squamous cell carcinoma: a clinic and pathologic correlation. *Oral Dis* 2006;**12**:301–4.
10. Yanamoto S, Yamada S, Takahashi H, Naruse T, Matsushita Y, Ikeda H, et al. Expression of the cancer stem cell markers CD44v6 and ABCG2 in tongue cancer: effect of neoadjuvant chemotherapy on local recurrence. *Int J Oncol* 2014;**44**:1153–62.
11. Peitzsch C, Nathansen J, Schniewind SI, Schwarz F, Dubrovska A. Cancer stem cells in head and neck squamous cell carcinoma: identification, characterization and clinical implications. *Cancers (Basel)* 2019;**11**.
12. Xiao M, Liu L, Zhang S, Yang X, Wang Y. Cancer stem cell biomarkers for head and neck squamous cell carcinoma: a bioinformatic analysis. *Oncol Rep* 2018;**40**:3843–51.
13. Maccalli C, Rasul KI, Elawad M, Ferrone S. The role of cancer stem cells in the modulation of anti-tumor immune responses. *Semin Cancer Biol* 2018;**53**:189–200.
14. Zins K, Heller G, Mayerhofer M, Schreiber M, Abraham D. Differential prognostic impact of interleukin-34 mRNA expression and infiltrating immune cell composition in intrinsic breast cancer subtypes. *Oncotarget* 2018;**9**:23126–48.
15. Jang TJ, Park JB, Lee JI. The expression of CD10 and CD15 is progressively increased during colorectal cancer development. *Korean J Pathol* 2013;**47**:340–7.
16. Jana SH, Jha BM, Patel C, Jana D, Agarwal A. CD10 – a new prognostic stromal marker in breast carcinoma, its utility, limitations and role in breast cancer pathogenesis. *Indian J Pathol Microbiol* 2014;**57**:530–6.
17. Sasaki T, Kuniyasu H, Luo Y, Fujiwara R, Kitayoshi M, Tanabe E, et al. Serum CD10 is associated with liver metastasis in colorectal cancer. *J Surg Res* 2014;**192**:390–4.
18. Charoentong P, Angelova M, Efremova M, Gallasch R, Hackl H, Galon J, et al. Bioinformatics for cancer immunology and immunotherapy. *Cancer Immunol Immunother* 2012;**61**:1885–903.
19. Xu H, Lai W, Zhang Y, Liu L, Luo X, Zeng Y, et al. Tumor-associated macrophage-derived IL-6 and IL-8 enhance invasive activity of LoVo cells induced by PRL-3 in a KCNN4 channel-dependent manner. *BMC Cancer* 2014;**14**:330.
20. Mishra D, Singh S, Narayan G. Role of B cell development marker CD10 in cancer progression and prognosis. *Mol Biol Int* 2016;**2016**:4328697.
21. Leithner K, Wohlkoenig C, Stacher E, Lindenmann J, Hofmann NA, Galle B, et al. Hypoxia increases membrane metallo-endopeptidase expression in a novel lung cancer ex vivo model – role of tumor stroma cells. *BMC Cancer* 2014;**14**:40.
22. Lohaus F, Linge A, Tinhofer I, Budach V, Gkika E, Stuschke M, et al. HPV16 DNA status is a strong prognosticator of loco-regional control after postoperative radiochemotherapy of locally advanced oropharyngeal carcinoma: results from a multicentre explorative study of the German Cancer Consortium Radiation Oncology Group (DKTK-ROG). *Radiother Oncol* 2014;**113**:317–23.
23. Kimple RJ, Harari PM. The prognostic value of HPV in head and neck cancer patients undergoing postoperative chemoradiotherapy. *Ann Transl Med* 2015;**3**:S14.
24. Nichols AC, Chan-Seng-Yue M, Yoo J, Xu W, Dhaliwal S, Basmaji J, et al. A pilot study comparing HPV-positive and HPV-negative head and neck squamous cell carcinomas by whole exome sequencing. *ISRN Oncol* 2012;**2012**:809370.
25. Mantovani A. Molecular pathways linking inflammation and cancer. *Curr Mol Med* 2010;**10**:369–73.
26. Schafer ZT, Brugge JS. IL-6 involvement in epithelial cancers. *J Clin Invest* 2007;**117**:3660–3.
27. Su S, Chen J, Yao H, Liu J, Yu S, Lao L, et al. CD10(+)GPR77(+) cancer-associated fibroblasts promote cancer formation and chemoresistance by sustaining cancer stemness. *Cell* 2018;**172**. 841-56 e816.
28. Sumitomo M, Shen R, Nanus DM. Involvement of neutral endopeptidase in neoplastic progression. *Biochim Biophys Acta* 2005;**1751**:52–9.

29. Iijima-Ando K, Hearn SA, Granger L, Shenton C, Gatt A, Chiang HC, et al. Overexpression of neprilysin reduces alzheimer amyloid-beta42 (Abeta42)-induced neuron loss and intraneuronal Abeta42 deposits but causes a reduction in cAMP-responsive element-binding protein-mediated transcription, age-dependent axon pathology, and premature death in Drosophila. *J Biol Chem* 2008;**283**:19066–76.
30. Maguer-Satta V, Besancon R, Bachelard-Cascales E. Concise review: neutral endopeptidase (CD10): a multifaceted environment actor in stem cells, physiological mechanisms, and cancer. *Stem Cells* 2011;**29**:389–96.

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