World Journal of

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ABOUT COVER

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AIMS AND SCOPE

The primary aim of *World Journal of Gastrointestinal Oncology* (*WJGO, World J Gastrointest Oncol*) is to provide scholars and readers from various fields of gastrointestinal oncology with a platform to publish high-quality basic and clinical research articles and communicate their research findings online.

   WJGO mainly publishes articles reporting research results and findings obtained in the field of gastrointestinal oncology and covering a wide range of topics including liver cell adenoma, gastric neoplasms, appendiceal neoplasms, biliary tract neoplasms, hepatocellular carcinoma, pancreatic carcinoma, cecal neoplasms, colonic neoplasms, colorectal neoplasms, duodenal neoplasms, esophageal neoplasms, gallbladder neoplasms, *etc*.

INDEXING/ABSTRACTING

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RESPONSIBLE EDITORS FOR THIS ISSUE

ORIGINAL ARTICLE

Basic Study

Potential of six-transmembrane epithelial antigen of the prostate 4 as a prognostic marker for colorectal cancer

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Abstract

BACKGROUND

Immune cells play a role in the regulation of tumor cell behavior, and accumulating evidence supports their significance in predicting outcomes and therapeutic efficacy in colorectal cancers (CRC). Human six-transmembrane epithelial antigen of the prostate (STEAP) proteins have been recognized and utilized as promising targets for cell- and antibody-based immunotherapy. One STEAP family member, STEAP4, is expected to be an attractive biomarker for the immunotherapy of prostate and breast cancer. However, the immunotherapeutic role of STEAP4 for colorectal carcinomas has not been demonstrated.

AIM

To explore the expression pattern of STEAPs in CRC and their relationship with immune infiltration, and investigate the potential utilization of STEAPs as novel prognostic indicators in colorectal carcinomas.

METHODS

The expression level of STEAPs in CRC was evaluated using various open-resource databases and online tools to explore the expression characteristics and prognostic significance of STEAPs, as well as their correlation with immune-related biomarkers, such as immune infiltration. Immunohistochemical (IHC) experiments were subsequently performed to verify the database conclusions.

RESULTS

The levels of STEAPs in CRC were inconsistent. The expression of STEAPs 1-3 in CRC was not significantly different from that in normal tissues. However,

STEAP4 mRNA levels were significantly lower in CRC than in normal tissue and were positively correlated with immune-related biomarkers, such as immune cell infiltration, immune stimulation, major histocompatibility complex levels, and chemokines. Interestingly, the expression of STEAP4 in microsatellite instability-high CRC subtype was higher than that in microsatellite stability subtype. IHC staining was performed on colon cancer tissue samples and showed that high expression of STEAP4 in adjacent tissues positively correlated with immune-related biomarkers, including MLH1, MLH6, and PMS2, but negatively correlated with programmed death ligand 1, to varying degrees.

CONCLUSION
Our results provide an analysis of the expression of STEAP family members in CRC. Among different STEAP family members, STEAP4 plays a different role in CRC compared to STEAPs 1-3. In CRC, STEAP4 expression is not only lower than that in normal tissues, but it is also positively correlated with immune infiltration and immune-related biomarkers. These findings suggest that STEAP4 may be a potential biomarker for predicting CRC immune infiltration status.

Key Words: Six-transmembrane epithelial antigen of the prostate; Colorectal cancer; Immunotherapy; Target; Survival

Core Tip: This study analyzed the expression levels of six-transmembrane epithelial antigen of the prostate (STEAP) family members in colorectal cancer (CRC) and explored the potential biological function of STEAP4. It was found that the expression of STEAP4 in CRC tissues had a positive correlation with immune infiltration and immune-related biomarkers, such as MLH1, MLH6, and PMS2, and a negative correlation with programmed death ligand 1. STEAP4 is expected to be a novel and potential prognostic biomarker for CRC.

INTRODUCTION

Colorectal cancer (CRC) ranks third in incidence and second in cancer mortality among malignant tumors worldwide[1]. Currently, large changes in lifestyle and dietary habits are thought to have contributed to the increased incidence and mortality of CRC. For example, in China, the annual average increase of new CRC cases was estimated to be 4.2%[2]. Although gender and regional differences are considered as the prognostic factors for patients with CRC[3], the etiology of CRC oncogenesis and development is still complex and unclear.

With the development of standardized treatment for patients with CRC, the prognosis of CRC has greatly improved. However, the control of progression of metastatic disease is still intractable. Immune cells play an important role in the regulation of tumor cell behavior, and accumulating evidence supports their significance in predicting outcomes and therapeutic efficacy in CRC patients[4]. In this regard, attention is currently being paid to the immune microenvironment and immunotherapy of CRC, mainly focusing on T cells and therapeutic response as related to promising treatment strategies[5].

Immunotherapy serves as an alternative treatment for cancer patients, especially for those whose tumors overexpress antigens recognized by immune cells. The human six-transmembrane epithelial antigen of the prostate (STEAP) family of proteins belongs to a class of cellular transmembrane proteins and has been used to derive epitope peptides that stimulate T lymphocytes in patients with renal cell or bladder cancer[6]. Importantly, STEAPs are present at the intercellular junctions of the prostate secretory epithelium, and are overexpressed in prostate cancer, serving as attractive targets for prostate cancer immunotherapy[7].

Although STEAPs have been reported to be overexpressed in CRC[8-11], research on STEAPs in CRC remains limited and the immunotherapeutic role of STEAPs in colorectal carcinomas has not been shown. This research investigated the biological function of STEAPs in CRCs, in addition to the relationship between STEAPs and immune infiltration, and demonstrated the potential of STEAPs to serve as novel and prognostic biomarkers for immunotherapy in colorectal carcinomas.

MATERIALS AND METHODS

Patient information and ethics statement

A tissue microarray with 87 matched primary CRC tissues and their corresponding adjacent normal colorectal tissue samples, and six extra samples of cancer cases without the corresponding paracancerous tissue were purchased from the Shanghai OUTDO Biotech Company, Shanghai, China (XT17-025, HColA180Su18). Pathological type was classified according to the prognostic degree of cancers. This study was approved by the Ethics Committee of Shantou University Medical College.

Comparison of STEAP expression in normal and cancerous tissues

TCGA datasets were used to evaluate the expression of STEAPs in normal and different cancerous tissues through the Tumor IMmune Estimation Resource (TIMER2.0) online source (http://timer.cistrome.org/)[12]. The UCSC Xena database (https://genome-cancer.ucsc.edu/)[13] was applied to analyze STEAP expression differences in colon adenocarcinomas (COAD) and rectal adenocarcinomas (READ) and related normal tissues. Regarding the different subtypes of CRC, namely, microsatellite instability-high (MSI-H), microsatellite instability-low (MSI-L), and microsatellite stable (MSS)[14], MSI is a biomarker for response to immune checkpoint inhibitors (ICIs); high disease control rates and good progression-free survival were observed in patients with MSI-H CRC[15]. MSI-L tumors are phenotypically indistinguishable from MSS tumors, and the biological significance of MSI-L is unclear[16], so emphasis has been placed on MSI-H and MSS. The expression of STEAPs in MSI-H and MSS was also evaluated in the GEPIA2 database (http://gepia2.cancer-pku.cn/)[17].

Relationship between STEAPs and immune infiltration in CRC

Immune cells involved in CRC development were evaluated using the TIMER2.0 database to predict the association between the expression of STEAPs and the abundance of immune cells, including B cells, CD4+ T cells, CD8+ T cells, neutrophils, macrophages, and dendritic cells, in the tumor microenvironment. In terms of immune characteristics of lymphocytes, immunostimulants, major histocompatibility complex (MHC), and chemokines, TISIDB, an integrated repository portal for tumor-immune system interactions (http://cis.hku.hk/TISIDB/)[18], was applied to explore the potential function of STEAPs in CRC immune infiltration. After being downloaded from the TISIDB, the dataset was analyzed and drawn with matrix2png (https://matrix2png.msl.ubc.ca/bin/matrix2png.cgi), an online mapping program[19].

Immunohistochemical staining

The immunohistochemical (IHC) staining for STEAP4 in tissue microarrays was conducted as described before[20]. The tissue microarray slide was dewaxed in xylene, hydrated in graded alcohols, and processed with 2% ethylenediamine tetraacetic acid antigen-repair solution (Fuzhou Maixin Biotechnology Development Co. LTD, Fuzhou, China) by microwave heating for epitope retrieval. After blocking endogenous peroxidase with 3% H_2O_2, the slide was incubated with anti-STEAP4 antibody (dilution: 1:1000, Proteintech 11944-AP) at 4 °C overnight. Stained tissues with DAB reagent were mounted and underwent nuclear counterstaining with hematoxylin for visualization.

Sections were visualized under a bright-field microscope (Axio Imager A2, Zeiss, Germany) and evaluated independently by two investigators with no prior knowledge of the CRC patient information. For tissue expression of STEAP4, staining intensity was scored as 0, 1, 2, and 3 for colorless, light yellow, brown yellow, and dark brown, respectively, while the percentage of positive cells, equaling 0%, 1%-25%, 26%-50%, 51%-75% and 76%-100%, was scored as 0, 1, 2, 3, and 4 points, respectively. The final staining score for STEAP4 expression was calculated as the sum of staining intensity and the percentage of positive cells, and divided into low expression (scores 0-4) and high expression (scores 5-7) groups. The expression levels of MLH1/2/6, PMS2, and programmed death ligand 1 (PDL1) were included in the patient information and the cutoffs were described before[21].

Statistical and survival analyses

SPSS 25.0 statistical software was used to analyze the results. Enumerated data are recorded as the number of cases (*n* = 93), and the relationship between STEAP4 and the clinicopathological parameters of CRC patients was analyzed by the χ^2 and Fisher's exact probability tests. The relationship between expression of STEAP4 in CRC and adjacent normal tissues (*n* = 87 cases) was examined by the χ^2 test. Likewise, the correlation between highly expressed STEAP4 in CRC and the immune-related factors MLH1, MLH2, MLH6, PMS2, and PDL1 was determined by the χ^2 test. To investigate the prognostic value of STEAP4 in CRC patients, the Kaplan-Meier survival curve and log-rank test were used to evaluate the association of STEAP4 expression with CRC patient prognosis by using SPSS 25.0 software. The difference was considered statistically significant at *P* < 0.05.

Table 1 Expression of six-transmembrane epithelial antigen of the prostate 4 in colorectal cancer and normal tissues

	Case (*n*)	STEAP4		χ^2	*P* value
		Low	High		
Tumor	87	42 (48.3%)	45 (51.7%)	8.866	0.003[a]
Normal	87	23 (26.4%)	64 (73.6%)		

[a]$P < 0.05$.

STEAP4: Six-transmembrane epithelial antigen of the prostate 4.

RESULTS

Expression of STEAPs in different types of malignant tumors

To determine the expression pattern of STEAPs in different types of malignant tumors, TIMER2.0 was used to analyze the difference between normal and cancerous tissues in TCGA. All STEAPs were found to be expressed at low levels in breast invasive carcinoma and kidney chromophobe compared to corresponding normal tissues, while in other types of malignancies, the expression patterns of STEAPs differed (Figure 1). Interestingly, in COAD, head and neck squamous cell carcinoma, lung adenocarcinoma, lung squamous cell carcinoma, and READ, the expression of STEAP4 was lower than that in normal tissues. However, in such cancerous tissues, the levels of STEAPs 1-3 were higher or not significantly different from those in normal tissues, suggesting that STEAP4 may perform a different function from STEAPs 1-3 in patients with such cancers.

CRC tissues have lower STEAP4 levels compared with normal tissues

To verify the expression pattern of STEAPs in CRC, another database, UCSC Xena, was analyzed to confirm the findings. Although expression of STEAPs 1 and 2 in COAD and READ were not different compared to their corresponding normal tissues, STEAP4 expression was lower, while STEAP3 expression was higher compared to that in corresponding normal tissues (Figure 2).

Low STEAP4 levels are associated with the MSS subtype of CRC

CRC is highly heterogeneous at the genetic and molecular levels, which affects the efficacy of clinical therapy. A subset of CRCs exhibit MSI, indicating defective DNA mismatch repair and high mutational burden, different from the majority of MSS subtypes[22]. CRC patients, especially those with MSI-H tumors, are more sensitive to ICIs than those with MSS tumors, and MSI-H tumors have greater infiltration of immune cells, higher expression of immune-related genes, and higher immunogenicity than MSS tumors[23]. Since the expression of STEAP4 is consistently low in CRC, different from the other three genes, we focused on STEAP4 for the remainder of this study. To explore STEAP4 expression levels in different subtypes of CRCs, the GEPIA2 database was used. Interestingly, the mRNA level of STEAP4 was high in MSI-H CRCs compared to MSI-L and MSS tumors (Figure 3).

STEAP4 is decreased in cancer tissues

As the expression pattern of STEAP4 seems to be different from those of the other STEAP members, paired normal and CRC tissues were used to evaluate the protein level of STEAP4. Representative images of STEAP4 expression are shown in Figure 4. The percentage of CRC tissues with high levels of STEAP4 was 51.7%, which was still lower than that in normal tissue (73.6%, $P = 0.003$) (Table 1).

Relationship between STEAP4 levels and clinicopathological parameters of patients with CRC

Clinicopathological analysis showed that STEAP4 expression was not associated with gender, primary tumor stage, lymph node status, American Joint Committee on Cancer (AJCC) stage, or pathological type (Table 2). The expression of STEAP4 decreased with the increase of primary tumor stage, lymph node status, and AJCC stage.

STEAP4 protein levels are positively associated with MLH1, MLH6, and PMS2, and negatively associated with PDL1

Based on the IHC results, the expression of STEAP4 and corresponding immune-related biomarkers in CRC was analyzed using χ^2 statistical analysis. As shown in Table 3, a low level of STEAP4 was positively related to low levels of MLH1, MLH6, and PMS2, but negatively associated with PDL1 level in CRC patients.

Expression level of STEAP4 is associated with the abundance of immune cell infiltration in CRC

Given the importance of antitumor immunity in the tumor microenvironment, the correlation between
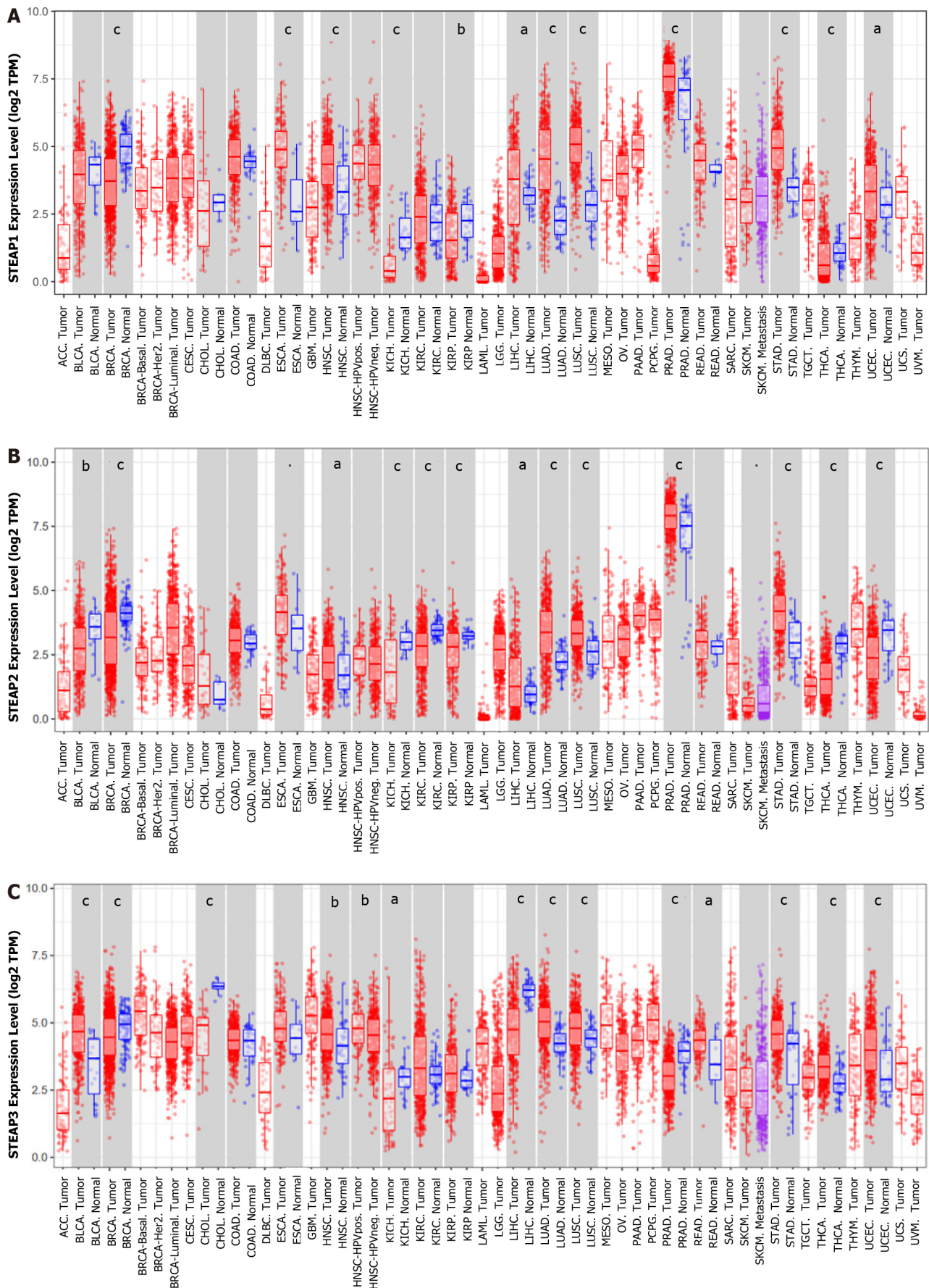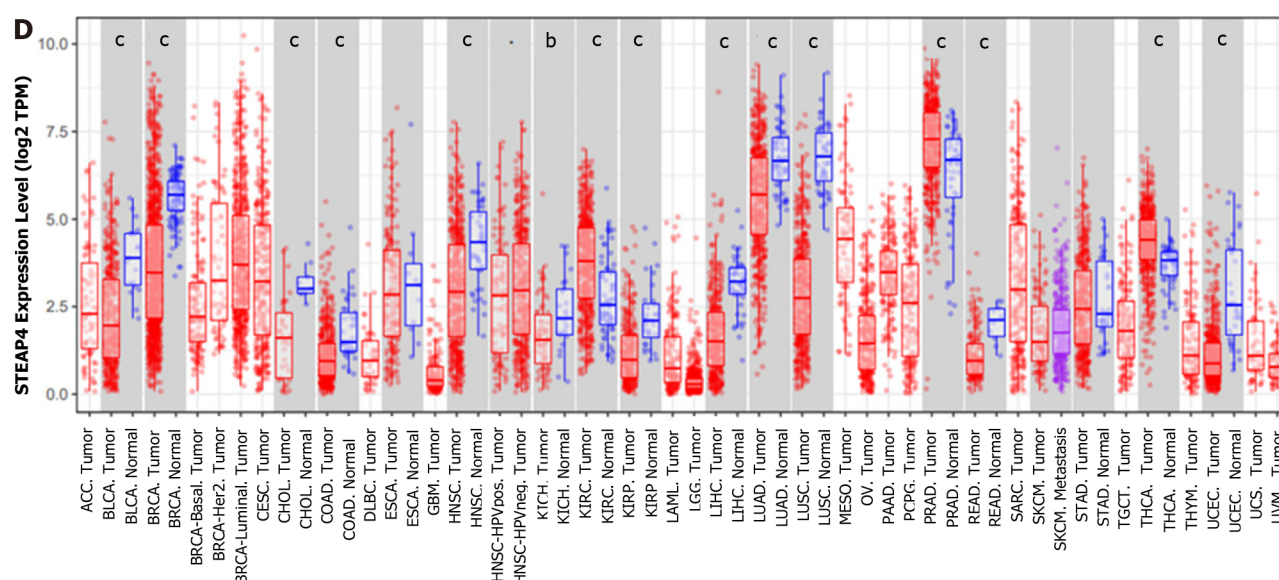
Figure 1 Expression of six-transmembrane epithelial antigen of the prostate in different normal and cancerous tissues from the TIMER2.0 database. A: Six-transmembrane epithelial antigen of the prostate (STEAP) 1; B: STEAP2; C: STEAP3; D: STEAP4. Student's *t*-test was used to estimate the significance of the differences in gene expression levels between groups. $^aP < 0.05$, $^bP < 0.01$, $^cP < 0.001$. STEAP: Six-transmembrane epithelial antigen of the prostate; ACC: Adrenocortical carcinoma; BLCA: Bladder urothelial carcinoma; BRCA: Breast invasive carcinoma; CESC: Cervical squamous cell carcinoma and endocervical adenocarcinoma; CHOL: Cholangiocarcinoma; COAD: Colon adenocarcinoma; DLBC: Lymphoid neoplasm diffuse large B-cell lymphoma; ESCA: Esophageal carcinoma; GBM: Glioblastoma multiforme; HNSC: Head and neck squamous cell carcinoma; KICH: Kidney chromophobe; KIRC: Kidney renal clear cell carcinoma; KIRP: Kidney renal papillary cell carcinoma; LAML: Acute myeloid leukemia; LGG: Brain lower grade glioma; LIHC: Liver hepatocellular carcinoma; LUAD: Lung adenocarcinoma; LUSC: Lung squamous cell carcinoma; MESO: Mesothelioma; OV: Ovarian serous cystadenocarcinoma; PAAD: Pancreatic adenocarcinoma; PCPG: Pheochromocytoma and paraganglioma; PRAD: Prostate adenocarcinoma; READ: Rectum adenocarcinoma; SARC: Sarcoma; SKCM: Skin cutaneous melanoma; STAD: Stomach adenocarcinoma; TGCT: Testicular germ cell tumors; THCA: Thyroid carcinoma; THYM: Thymoma; UCEC: Uterine corpus endometrial carcinoma; UCS: Uterine carcinosarcoma; UVM: Uveal melanoma.

expression of STEAP family members and immune cells in CRC was analyzed. Based on the TIMER database, it was found that the expression of STEAP1, STEAP2, and STEAP4 was negatively correlated with tumor purity and positively correlated with six types of immune cells, specifically, B cells, CD8$^+$ T cells, CD4$^+$ T cells, neutrophils, macrophages, and dendritic cells, in COAD and READ (Figures 5A, B, and D). However, the expression of STEAP3 was positively correlated with tumor purity, CD4$^+$ T cells, neutrophils, macrophages, and dendritic cells, and negatively correlated with B cells and CD8$^+$ T cells in COAD and READ (Figure 5C).

STEAP4 is positively correlated with immune characteristics in CRC

According to the TISIDB database, Spearman correlation analysis showed that the expression of STEAP1, STEAP2, and STEAP4 was positively correlated, but STEAP3 was negatively correlated with lymphocytes, immunostimulants, MHCs, and chemokines (Figures 6A-F), which is consistent with the results obtained based on the TIMER database (Figures 6C and G).

Low expression of STEAP4 in cancer tissues tends to predict poor overall survival in CRC patients

Based on the IHC results, the protein expression of STEAP4 was analyzed by the Kaplan-Meier method with log-rank test, which demonstrated that STEAP4 expression was not significantly associated with the overall survival (OS) (*P* > 0.05, Figure 7). Although the difference did not meet the statistical criteria, it was found that high expression of STEAP4 tended to predict a longer OS for CRC patients, suggesting that the protein level of STEAP4 could be a predictor of the survival of CRC patients.

DISCUSSION

Members of the STEAP family were originally identified as metalloreductases *in vivo*, playing an important role in maintaining iron homeostasis[24]. Abnormal accumulation of iron caused by unbalanced iron metabolism has been reported to lead to the occurrence, progression, and invasion of tumors[25]. Thus, the STEAP family bridges iron homeostasis and cancer[26]. As potential biomarkers and therapeutic targets of tumors, the STEAP family members play an important role in tumor therapy.

All four STEAP proteins are increased in prostate cancer and play important roles in the development and progression of prostate cancer[27]. Interestingly, although the structure of STEAP4 is similar to that
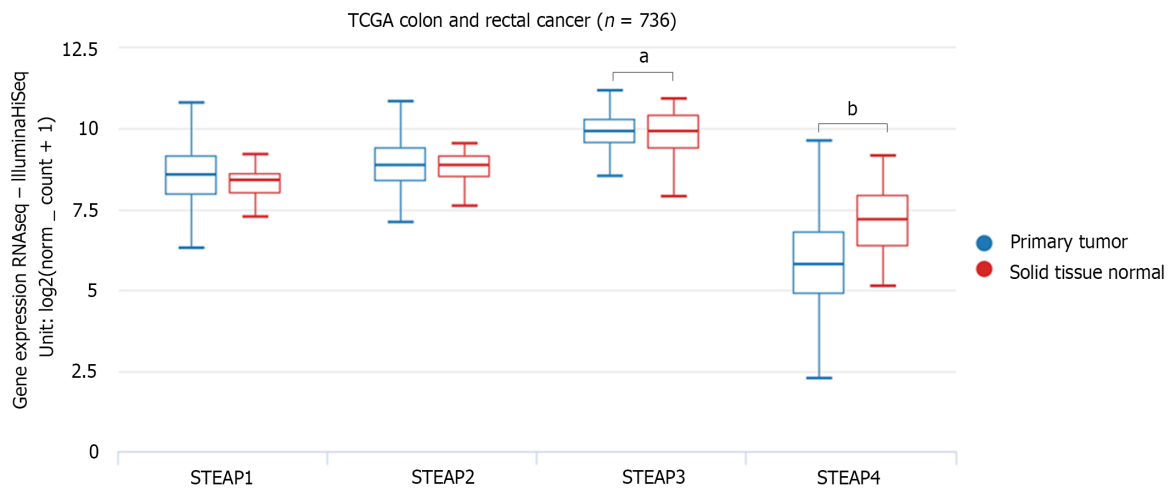
Figure 2 Six-transmembrane epithelial antigen of the prostate mRNA expression in colon adenocarcinoma and rectal adenocarcinoma tissues in the UCSC Xena database. Red represents normal tissue and blue is cancerous tissue. One-way ANOVA was utilized to estimate the significance of differences in six-transmembrane epithelial antigen of the prostate expression levels between groups. [a]$P < 0.01$, [b]$P < 0.001$. STEAP: Six-transmembrane epithelial antigen of the prostate.
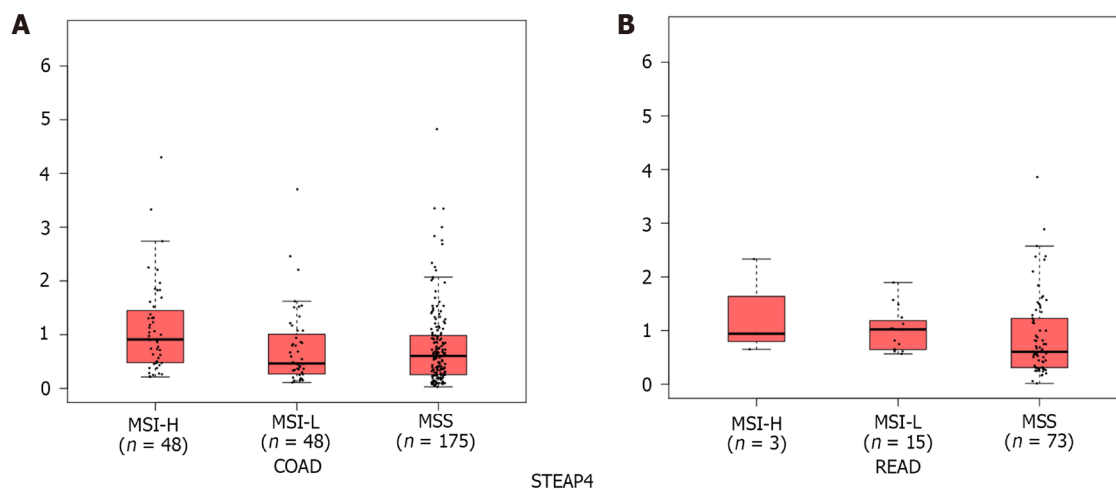
Figure 3 Expression patterns of six-transmembrane epithelial antigen of the prostate 4 in different subtypes of colorectal cancer. A: Colon adenocarcinoma; B: Rectal adenocarcinoma. MSI-H: Microsatellite instability-high; MSI-L: Microsatellite instability-low; MSS: Microsatellite stable; STEAP4: Six-transmembrane epithelial antigen of the prostate 4; COAD: Colon adenocarcinoma; READ: Rectal adenocarcinoma.
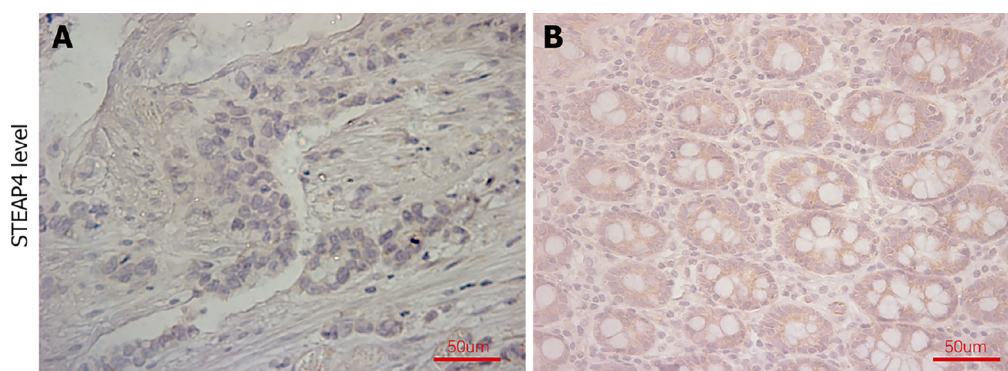
Figure 4 Representative images of six-transmembrane epithelial antigen of the prostate 4 expression in colorectal cancer and normal tissues. A: Six-transmembrane epithelial antigen of the prostate 4 (STEAP4) expression in colorectal cancer tissues; B: STEAP4 expression in adjacent normal tissues. Scale bar, 50 μm. STEAP4: Six-transmembrane epithelial antigen of the prostate 4.
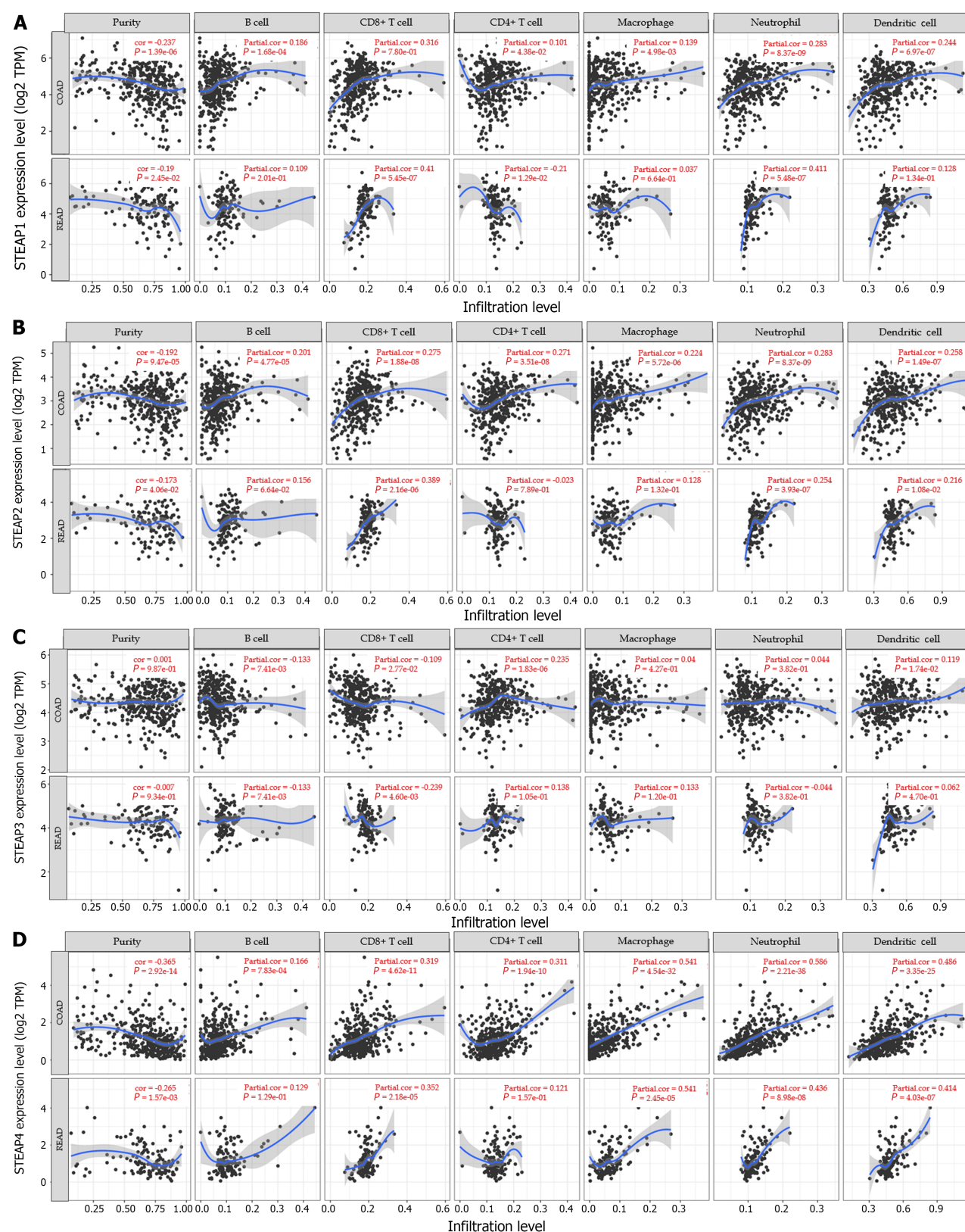
Figure 5 Relationship between six-transmembrane epithelial antigen of the prostate family member expression levels and immune infiltrates validated by the TIMER database. A: Six-transmembrane epithelial antigen of the prostate (STEAP1); B: STEAP2; C: STEAP3; D: STEAP4. STEAP: Six-transmembrane epithelial antigen of the prostate.

of the three other STEAP family members, the function of STEAP4 may diverge in different types of cancers[24]. STEAP1 antibody can effectively activate CD8+ T cells, natural killer cells, and other immune-related factors against a broad spectrum of tumors[28]. In this study, there was a great difference between STEAP4 and STEAP1-3; STEAP4 promoted androgen receptor (AR)-positive prostate cancer (PC) and inhibited AR-negative PC, while it was very different from STEAP1-3 in CRC, so there

Table 2 Correlation between six-transmembrane epithelial antigen of the prostate 4 expression and clinicopathological parameters in colorectal cancer patients

Clinical parameter	STEAP4		P value
	Low (%)	High (%)	
Gender			
Female	29 (59.2)	20 (40.8)	0.123
Male	19 (43.2)	25 (56.8)	
Primary tumor stage[1]			
T1-T3	36 (48.0)	39 (52.0)	0.213
T4a-T4b	11 (64.7)	6 (35.3)	
Lymph node status			
N0-N1	41 (48.8)	43 (51.2)	0.160
N2	7 (77.8)	2 (22.2)	
AJCC stage			
Phase 1-2	28 (43.1)	30 (56.9)	0.407
Phase 3	20 (57.1)	15 (42.9)	

[1]One colorectal cancer patient with undetected primary tumor (Tx) was excluded from the primary tumor stage.
STEAP4: Six-transmembrane epithelial antigen of the prostate 4; AJCC: American Joint Committee on Cancer.

Table 3 Correlation of six-transmembrane epithelial antigen of the prostate 4 expression with MLH1/2/6, PMS2, and programmed death ligand 1 expression in colorectal cancer

		STEAP4		X^2	P value
		Low	High		
MLH1	Low	6	5	18.42	0.000004[a]
	High	33	36		
MLH2	Low	18	10	3.90	0.071
	High	21	30		
MLH6	Low	8	7	13.44	0.0003[a]
	High	29	33		
PMS2	Low	12	8	10.31	0.017[b]
	High	27	30		
PDL1	Low	43	41	35.37	0.0019[c]
	High	2	4		

[a]$P < 0.001$.
[b]$P < 0.05$.
[c]$P < 0.01$.
PDL1: Programmed death ligand 1; STEAP4: Six-transmembrane epithelial antigen of the prostate 4.

might be different mechanisms. Interestingly, we found dual anti-STEAP1 antibody targeting T cells for cancer immunotherapy[29]. Combined with our study, it is suggested that STEAP4 can be developed as a new therapeutic strategy. Therefore, this study explored the role of STEAP4 at the mRNA and protein levels. STEAP4 was rarely studied in CRC. The current study used clinical tissues from CRC patients and characterized the mRNA and protein levels of STEAP4 to determine the expression of STEAP4 in CRC. Not surprisingly, low expression of STEAP4 was found in CRC tissues compared with adjacent tissues, which is consistent with the low mRNA expression of STEAP4 in CRC and a potential tumor suppressor role for STEAP4 in CRC patients.
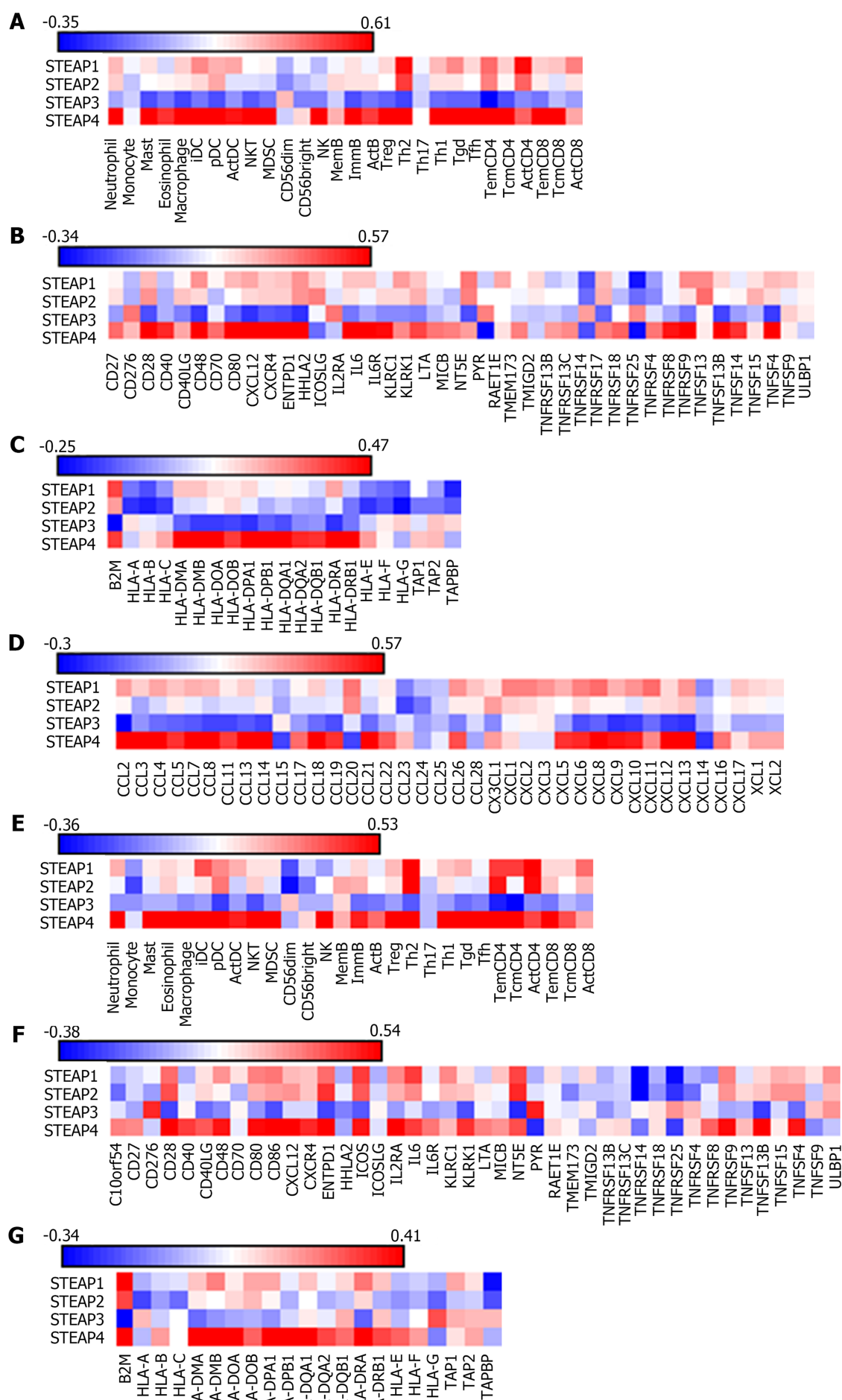
Figure 6 Spearman correlation between six-transmembrane epithelial antigen of the prostate family member expression and immune factors in colon adenocarcinoma and rectal adenocarcinoma. A-D: Colon adenocarcinoma; E-H: Rectal adenocarcinoma. STEAP: Six-transmembrane epithelial antigen of the prostate; iDC: Immature dendritic cell; pDC: Plasmacytoid dendritic cell; ActDC: Act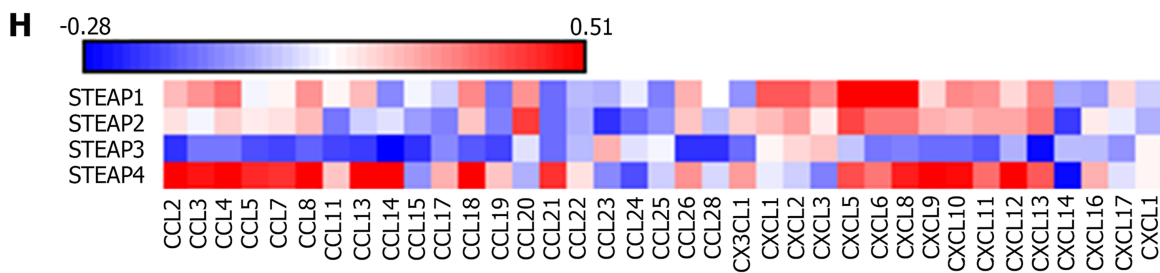ive dendritic cells; NKT: Natural killer T cell; MDSC: Myeloid derived suppressor cell; NK: Natural killer cell; Th: T helper cell; Tgd: Gamma delta T cell; Tfh: T follicular helper cell; B2M: Beta-2-microglobulin; HLA: Major histocompatibility complex; HLA-DOA: Major histocompatibility complex, class II, DO alpha; HLA-DOB: Major histocompatibility complex, class II, DO beta; HLA-DPA1: Major histocompatibility complex, class II, DP alpha 1; HLA-DPB1: Major histocompatibility complex, class II, DP beta 1; HLA-DQA1: Major histocompatibility complex, class II, DQ alpha 1; HLA-DRA: Major histocompatibility complex, class II, DR alpha; TAP: Transporter; TAPBP: Transporter Binding Protein; CD: Cluster of differentiation; CXCL: C-X-C motif chemokine ligand; ENTPD: Ectonucleoside triphosphate diphosphohydrolase; HHLA: HERV-H LTR-associating; ICOS: Inducible T cell costimulator; ICOSLG: Inducible T cell costimulator ligand; IL2RA: Interleukin 2 receptor subunit alpha; IL: Interleukin; IL6R: Interleukin 6 receptor; KIRC1: KIR3DL3, Homo sapiens killer cell immunoglobulin-like receptor, three domains, long cytoplasmic tail, 3; KIRK1: Killer cell lectin like receptor subfamily K, member 1; LTA: Lymphotoxin alpha; MICB: MHC class I polypeptide-related sequence B; NT5E: 5'-nucleotidase ecto; PVR: Poliovirus receptor; RAET1E: Retinoic acid early transcript 1E; TMEM173: STING, stimulator of interferon response CGAMP interactor; TMIGD: Transmembrane and immunoglobulin domain containing; TNFRSF: TNF receptor superfamily member; ULBP: UL16 binding protein; CCL: C-C motif chemokine ligand; CX3CL: C-X3-C motif chemokine ligand; XCL: X-C motif chemokine ligand.

To uncover the potential function of STEAP4 in CRC, clinicopathological parameters were analyzed. However, no statistical significance was found between the protein level of STEAP4 and primary tumor stage, lymph node metastasis, or AJCC stage. Reduced STEAP4 expression tended to be associated with advanced CRC stage and increased lymph node metastases, suggesting that the suppression of STEAP4 could play a role in promoting the progression and metastasis of CRC.

Recently, Xue *et al*[11] investigated the molecular mechanism of STEAP4 involvement in the hypoxic metabolism of inflammatory bowel disease and the linkage with mitochondrial dysfunction in colon cancer. Increasingly, high levels of STEAP4 result from increased levels of hypoxia and are associated with colitis in mouse models and inflammatory bowel disease patients. Inflammatory factors were not examined in the current study, which may influence STEAP4 levels in different types of CRC. Based on the inflammatory environment, hypoxic conditions can be associated with mitochondrial iron dysfunction caused by increased STEAP4[30]. However, in the absence of inflammatory infiltration, the inhibition of STEAP4 was reversed to be a tumor suppressor through interactions with protein kinases [31].

Further analyses were performed to uncover the potential relationship of STEAP4 with immune infiltration. As expected, both the protein and mRNA levels of STEAP4 are associated with immune-related factors, predicting a potential role of STEAP4 in stimulating immune infiltration. MLH1 deficiency has been shown to be associated with cetuximab resistance in CRC[21]. The positive relationship between STEAP4 and MLH1 in CRC suggests an involvement of STEAP4 in the immune response in the tumor microenvironment. Recently, Ijsselsteijn *et al*[32] reported that, for a cell-based model for Lynch syndrome, DNA mismatch repair deficiency was related to the core DNA mismatch repair genes MSH2, MSH6, MLH1, and PMS2[32]. The protein level of STEAP4 was found to be positively correlated with such DNA mismatch repair genes in the current study. The above results support a role for STEAP4 in the immune response to CRC to prevent further development and metastasis.

CONCLUSION

In the current study, STEAP4 was found to be a protective factor in the intestinal tract and could be used as a prognostic indicator for patients with CRC. CRC patients with high STEAP4 expression tend to have a longer survival.
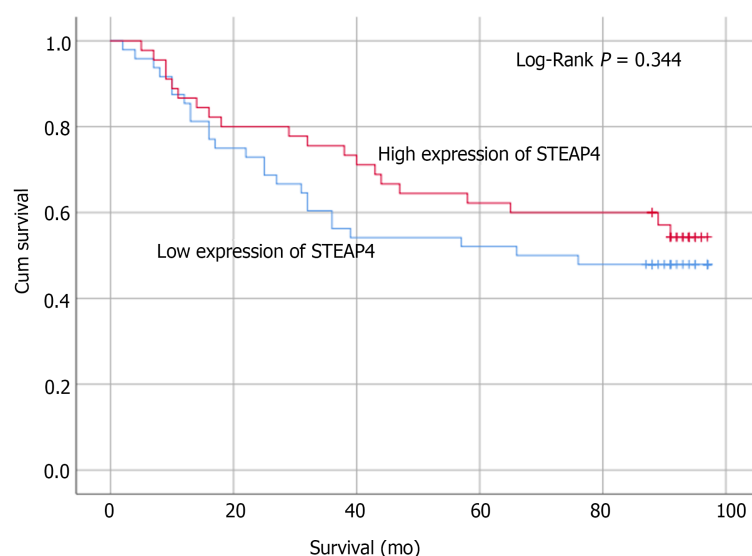
Figure 7 Low six-transmembrane epithelial antigen of the prostate 4 protein level tends to predict a poor overall survival in colorectal cancer patients. STEAP4: Six-transmembrane epithelial antigen of the prostate 4.

ARTICLE HIGHLIGHTS

Research background

Six-transmembrane epithelial antigen of the prostate 4 (STEAP4) is an attractive biomarker for the immunotherapy of prostate and breast cancers. However, the immunotherapeutic role of STEAP4 for colorectal carcinomas has not been demonstrated.

Research motivation

Immunotherapy emerges with predicting outcomes and therapeutic efficacy in colorectal cancers (CRC).

Research objectives

To explore the expression pattern of STEAPs in CRCs and their relationship with immune infiltration, and investigate the potential utilization of STEAPs as novel prognostic indicators in colorectal carcinomas.

Research methods

CRC patients' tissues and online datasets were used to analyze the expression level of STEAP4 in different types of CRC and their relationship with immune characteristics.

Research results

The expression of STEAP4 was significantly decreased in CRC tissues compared with adjacent normal ones, and was related to immune-related biomarkers. Low STEAP4 level predicted a poor overall survival of CRC patients.

Research conclusions

STEAP4 was found to be a protective factor in the intestinal tract and could be used as a prognostic indicator for patients with CRC.

Research perspectives

STEAP4 may be a potential biomarker for predicting CRC immune infiltration status.

FOOTNOTES

Author contributions: Liu J and Wu HT designed the research study; Fang ZX performed the research; Fang ZX, Li CL, Chen WJ, and Wu HT analyzed the research and wrote the manuscript; Liu J revised the manuscript critically; and all authors have read and approved the final manuscript.

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