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RESEARCH ARTICLE
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DECIPHERING THE MOLECULAR LANDSCAPE OF HEAD AND NECK SQUAMOUS CELL CARCINOMA: COMPREHENSIVE ANALYSIS OF SLC31A1 EXPRESSION AND REGULATORY MECHANISMS

Asim Gamaryani[1], Fadia Waheed[2], Ammar Nawazish[3], Sanobar Gull[4], Habibullah Janyaro[5], Bushra Parveen[6], Murtaza Haider[7], Momina Sami Khan[8], Syed Meesam Raza[7], Jamal Muhammad Khan[9]*

[1]School of Health and Society University of Wollongong, Australia,
Email: asimghayoorgamaryani@gmail.com
[2]Institute of Microbiology and Molecular Genetics Punjab University Lahore, Pakistan,
Email: fadia.phd.mmg@pu.edu.pk
[3]Hebei North University, Zhangjiakou, China, Email: ammar.nawazish.9@gmail.com
[4]Department of Biochemistry, Government College University Faisalabad, Faisalabad, Pakistan
Email: sanobargull@yahoo.com
[5]Department of Veterinary Surgery, Shaheed Benazir Bhutto University of Veterinary and Animal Sciences Sakrand, Sakrand, Pakistan, Email:Janyaroh@gmail.com
[6]Department of Chemistry, Government College University Faisalabad, Faisalabad, Pakistan,
Email: bushraparveen@gcuf.edu.pk
[7]Department of Microbiology, Quaid e Azam University, Islamabad, Pakistan,
Email:Murtazahaider4@outlook.com, 2019-cu-micro-005@cuvas.edu.pk
[8]Department of Microbiology, Shaheed Benazir Women University Peshawar. Peshawar, Pakistan,
Email:khanmominasami@gmail.com
[9]*Department of Parasitology, Cholistan University of Veterinary and Animal Sciences,
Bahawalpur, Pakistan, Email: jamalmkhan@cuvas.edu.pk

***Corresponding Author:** Jamal Muhammad Khan,
*Department of Parasitology, Cholistan University of Veterinary and Animal Sciences, Bahawalpur, Pakistan, Email: jamalmkhan@cuvas.edu.pk

Abstract

In the pursuit of understanding the intricate molecular landscape of Head and Neck Squamous Cell Carcinoma (HNSC), this study conducts a comprehensive analysis of the expression and regulatory mechanisms of SLC31A1. Initially focusing on overall expression, we uncover a significant up-regulation of SLC31A1 in HNSC tissues compared to normal controls, indicating its potential role in HNSC progression. Subsequent investigations, stratified based on various parameters, reveal consistent overexpression patterns across different cancer stages, patient ages, genders, and racial backgrounds, underscoring the substantial involvement of SLC31A1 in HNSC progression. Expanding our exploration into SLC31A1's regulatory mechanisms, we delve into its promoter methylation status. The analysis uncovers hypo-methylation in HNSC samples, suggesting a correlation with its overexpression in the disease context. Further investigations based on different parameters reveal persistent hypo-methylation patterns, emphasizing the pivotal role of SLC31A1 in

HNSC progression. In a critical survival analysis, higher expression levels of SLC31A1 are associated with poor prognosis in HNSC patients. This comprehensive examination provides valuable insights into the potential diagnostic and therapeutic implications of SLC31A1 in HNSC.

Keywords: SLC31A1; HNSC; Expression analysis; Biomarker

Introduction

Cancer stands as a significant global cause of mortality, claiming the lives of approximately 10 million individuals annually. Alarmingly, one in six deaths results from cancer, leading to substantial healthcare and socio-economic burdens. Head and Neck Cancer (HNC), ranking as the sixth most prevalent cancer type, records over 870,000 new cases and 440,000 deaths in 2022. HNC encompasses malignancies affecting the oral cavity, oropharynx, hypopharynx, and larynx. The inherent challenges of speaking, breathing, and swallowing contribute to the elevated mortality and morbidity associated with Head and Neck Cancer. Head and Neck Squamous Cell Carcinoma (HNSC) constitutes the most prevalent subtype within HNC, representing over 90% of cases. Projections indicate a potential surge to 1.8 million HNSC cases annually by 2030. Noteworthy risk factors for HNC include obesity, smoking, alcohol consumption, and human papillomavirus (HPV) infection, emphasizing the multifaceted nature of this health concern (1-6).

Various cancer treatment modalities exist, with our emphasis on leveraging immunotherapy through cuproptosis. Cuproptosis manifests in cells when the induction of copper leads to targeted cell death. Solute Carrier Family 31 Member 1 (SLC31A1) takes the forefront as a primary contributor to cellular copper uptake in mammalian cells and tissues (7).

Mutations and methylations in the SLC31A1 gene have been recognized across 33 different cancers. Comprehensive in silico analyses point towards SLC31A1 as a potential gene associated with cuproptosis in HNSC cancer. Copper, an essential trace element, plays a crucial role in the biochemical activity of at least 30 enzymes in human cells (8). Copper (Cu) serves as an essential micronutrient linked to tumor metastasis, acting as a crucial cofactor for various metalloenzymes involved in numerous biological processes. Solute Carrier Family 31 Member 1 (SLC31A1) plays a vital role in maintaining intracellular copper homeostasis (9). Recent research indicates that elevated intracellular copper levels can lead to cuproptosis, and dysregulated copper levels are implicated in oncogenesis and the progression of cancer (10, 11). SLC31A1, as a strong-affinity copper carrier, is indispensable for mitochondrial function (12, 13). Excessive and upregulated expression of SLC31A1 accelerates intracellular copper buildup, contributing to cancer progression and a more unfavorable prognosis.

In this study, various bioinformatics methods were employed to elucidate the mechanistic role of SLC31A1 in predicting the prognosis of HNSC. The functions of the SLC31A1 gene associated with HNSC were delineated by analyzing its clinical and molecular mechanisms. Utilizing the Cancer Genome Atlas (TCGA) and UALCAN databases, we investigated the expression of SLC31A1 to glean insights into its potential benefits for HNSC patients.

Material and Methods

Analysis of gene expression of SLC31A1 in HNSC

To compare the expression levels between normal cells and cancer cells, the TIMER 2.0 database was employed (8, 14). TIMER 2.0 offers an expanded range of tumor profiles by utilizing six state-of-the-art algorithms (15). The expression analysis of SLC31A1 in HNSC was conducted using GEPIA 2.0. The accessibility of cancer research data is enhanced, making it readily available for analysis.

Expression analysis of SLC31A1 across different stages of HNSC

The UALCAN database was employed for the expression analysis of SLC31A1 across different stages of HNSC (16). UCLCAN, being publicly available, proves to be a user-friendly resource for

researchers. UCLCAN facilitates the analysis of gene expression within various subgroups, allowing the utilization of different parameters. The user-friendly interface of UCLCAN makes data easily accessible for download (17). In addition, this database was utilized to assess the dysregulation and correlation of SLC31A1 with overall survival (OS) in HNSC patients.

Survival analysis of SLC31A1

Kaplan-Meier (KM) statistics were employed to estimate overall survival (OS) analysis (18, 19). This platform utilizes extensive clinical data to assess the impact of specific genes on the OS of cancer patients. In this study, the Kaplan-Meier (KM) plotter was utilized to investigate the prognostic value of SLC31A1 gene expression. Additionally, it was used to analyze the effect of SLC31A1 dysregulation on the overall survival (OS) of cancer patients.

Results

Expression analysis of SLC31A1 in HNSC and normal control samples

Initially, we focused on the expression of SLC31A1 in HNSC and normal control samples. Utilizing the UALCAN database for expression analysis, we identified variations in SLC31A1 expression. The results revealed a significant up-regulation of SLC31A1 expression in HNSC tissues compared to controlled normal symbols (Figure 1). The statistical significance observed, with a value of 1.56E-01 between normal and HNSC, led us to believe that SLC31A1 plays a significant role in the progression of HNSC.

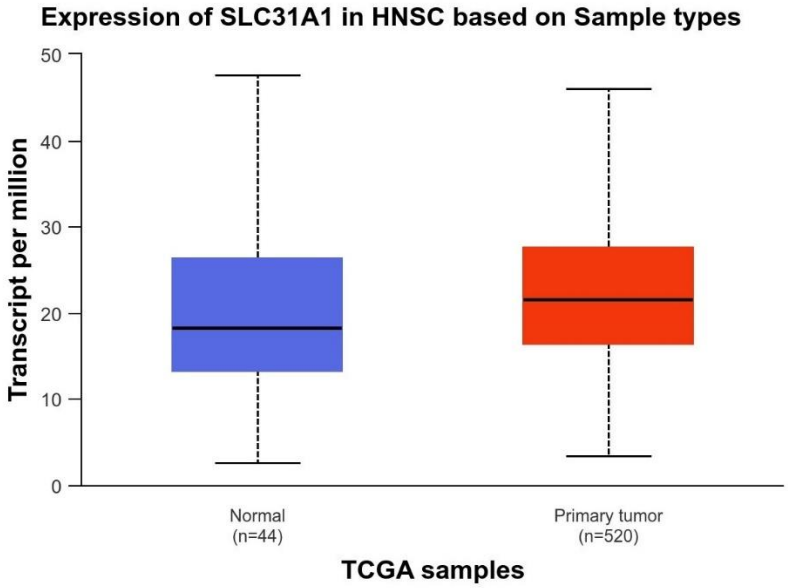

Figure 1: Expression pattern of SLC31A1 in HNSC and normal control samples.

Expression analysis of SLC31A1 in HNSC sample divided based on different clinical parameters

Following the analysis of SLC31A1 expression in both HNSC and normal samples, we further investigated its expression in HNSC samples with varying parameters, including individual cancer stages, patient's age, gender, and race. The examination of SLC31A1 expression at different cancer stages revealed a significant overexpression at various stages (Figure 2A). Subsequently, when considering patient race as a parameter, we observed variations in expression, with significant overexpression in samples of different races compared to normal samples (Figure 2B). Further exploration into SLC31A1 expression based on gender showed overexpression in both male and female HNSC samples (Figure 2C). Additionally, setting patient's age as a parameter revealed significant overexpression in HNSC samples across different age groups (Figure 2D). This comprehensive analysis underscores the considerable role of SLC31A1 in the progression of HNSC.

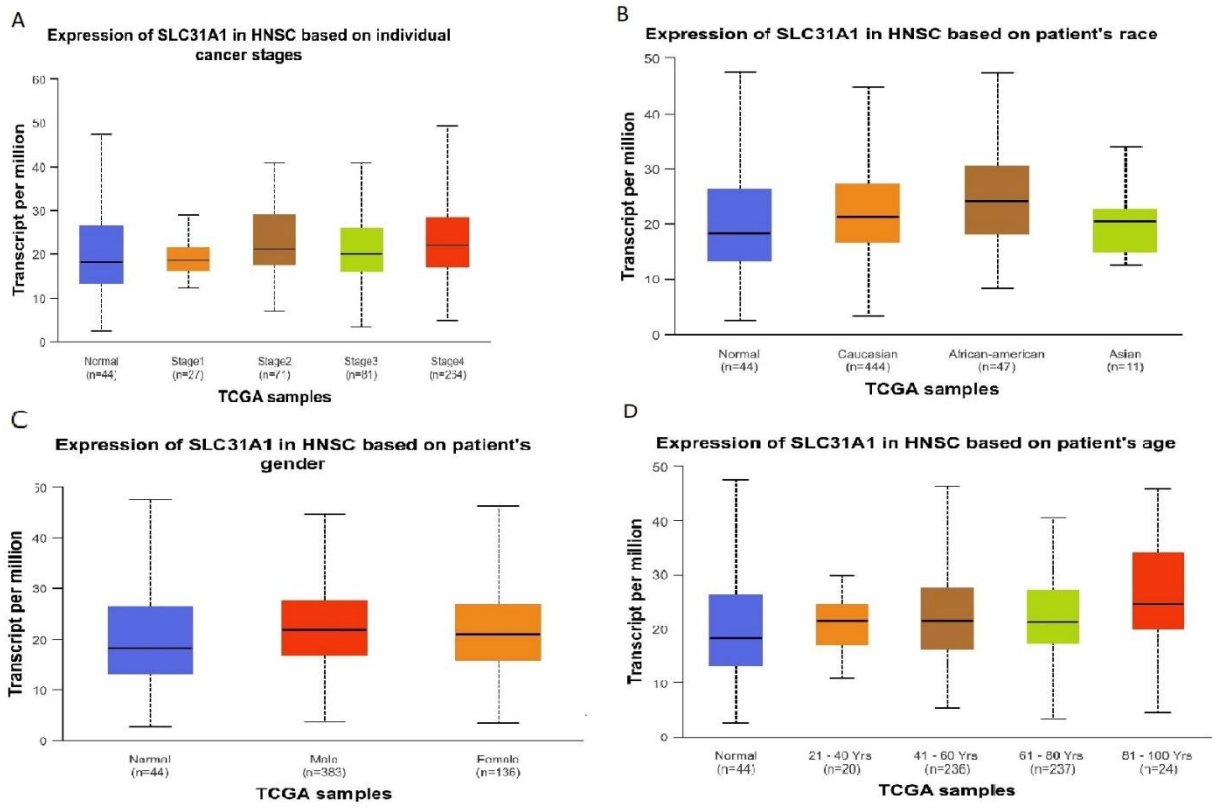

Figure 2: Expression pattern of SLC31A1 in normal control samples and HNSC samples stratified by different clinical variables.

Promoter methylation of SLC31A1 in HNSC and normal control samples

Research indicates that promoter methylation plays a crucial role in gene expression regulation (20). Therefore, we conducted an analysis of promoter methylation in HNSC and normal control samples utilizing the UALCAN database. Our findings revealed that the SLC31A1 level was hypo-methylated in HNC samples compared to normal controlled samples (Figure 3). This hypo-methylation suggests an association with the overexpression of SLC31A1 in HNSC.

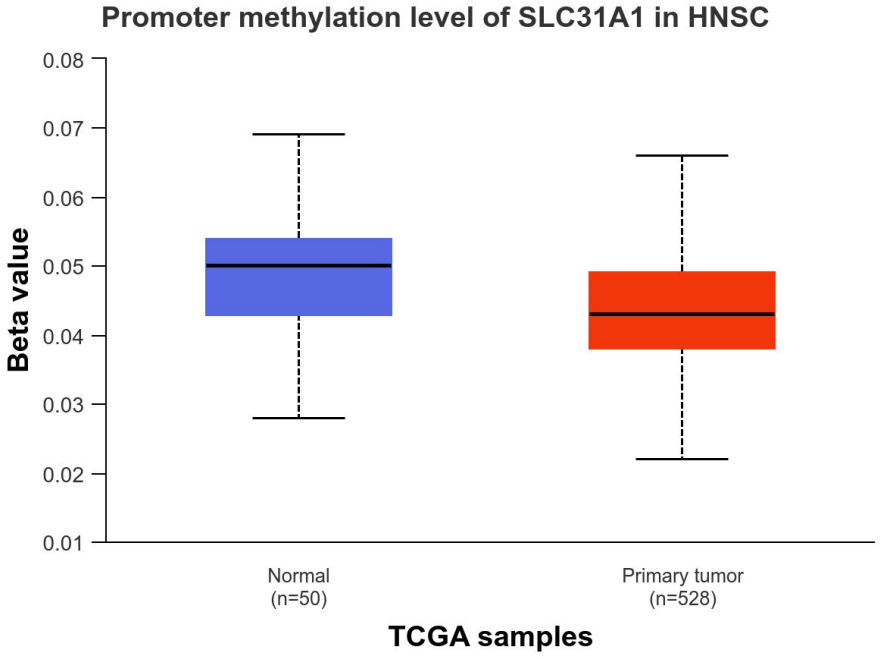

Figure 3: Promoter methylation pattern of SLC31A1 in HNSC and normal control samples.

Promoter methylation of SLC31A1 in HNSC sample divided based on different clinical parameters

Our investigation extended to the analysis of Promoter methylation of SLC31A1 within HNSC samples, considering various parameters such as Individual Cancer Stages, Patient's age, Patient's Gender, and Patient's race. Examining Promoter methylation of SL31A1 across different stages in patients with HNSC revealed Hypo-methylation at various stages (Figure 4A). Subsequently, we explored Promoter methylation of SLC31A1 in HNSC samples across different races, noting hypo-methylation in SLC31A1 (Figure 4B). Further analysis of SLC31A1 promoter methylation in HNSC samples across different genders indicated hypo-methylation in both male and female samples (Figure 4C). Continuing our parameter analysis, we focused on patient's age, revealing hypo-methylation in SLC31A1 Promoter at different ages in patients with HNSC (Figure 4D). This comprehensive analysis underscores the significant role of SLC31A1 in the progression of HNSC.

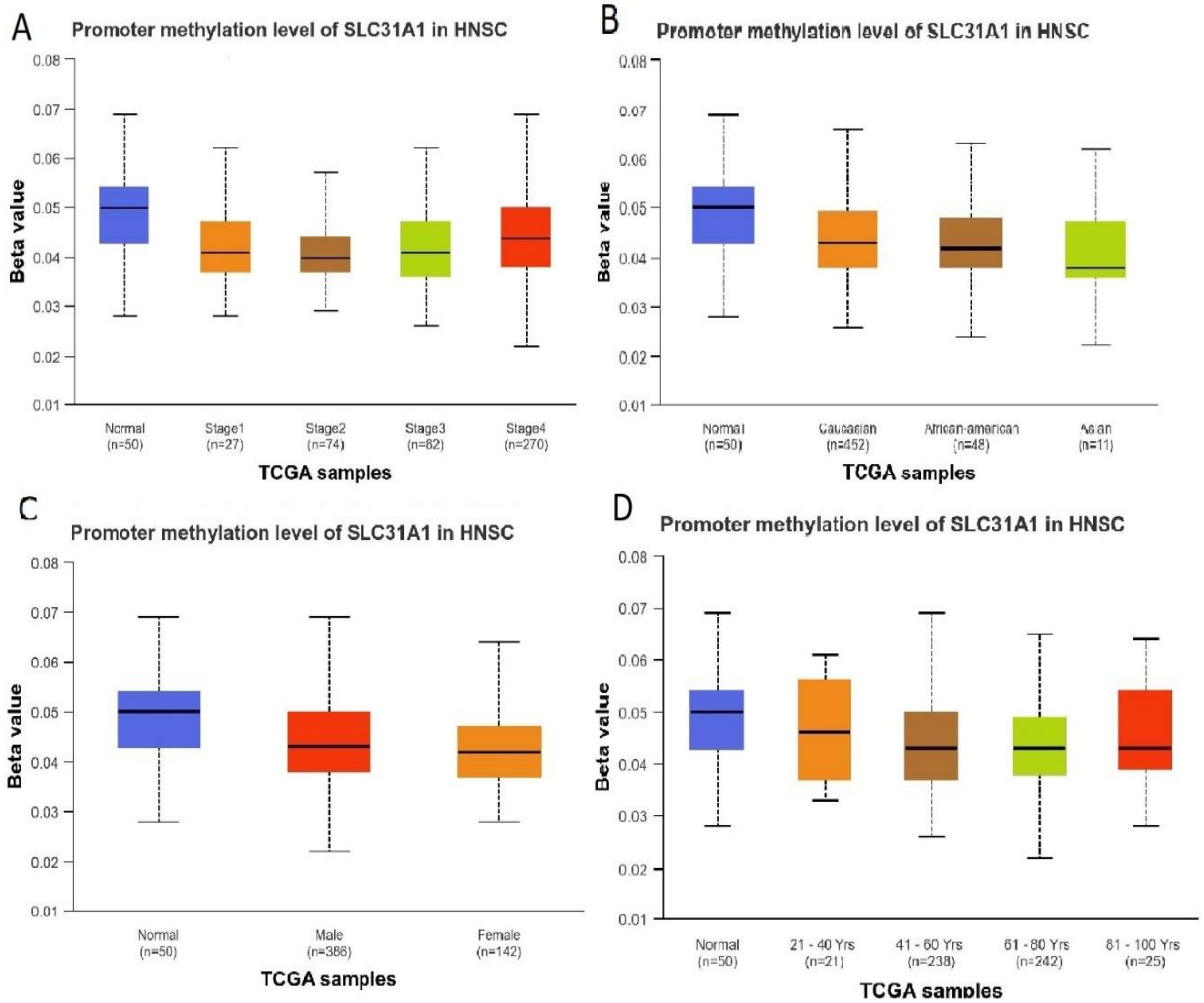

Figure 4: Promoter methylation pattern of SLC31A1 in normal control samples and HNSC samples stratified by different clinical variables.

Survival analysis of SLC31A1

Next, Km plotter was applied to explore prognostic significance of the SLC31A1 in HNSC patients. Results showed that the higher expression of SLC31A1 was associated with the poor prognosis of HNSC patients (Figure 5).

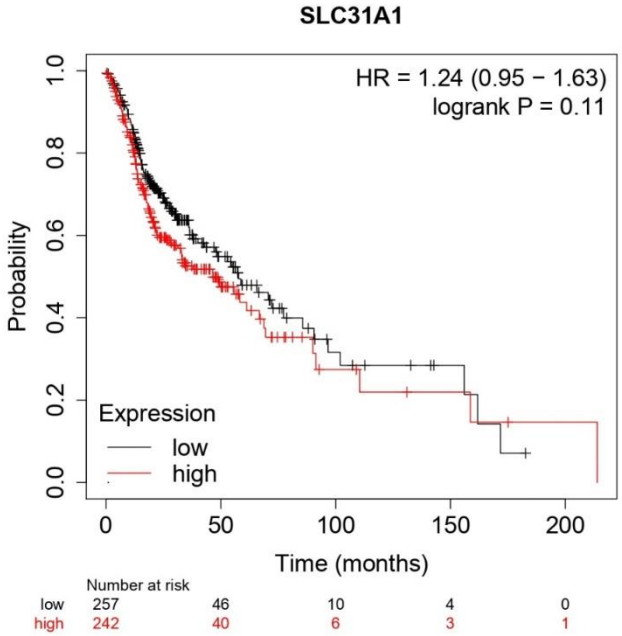

Figure 5: Survival curve of SLC31A1 in HNSC patients.

Discussion

Our investigation into the role of SLC31A1 in Head and Neck Squamous Cell Carcinoma (HNSC) has yielded substantial findings, offering a nuanced understanding of its expression patterns, promoter methylation, and prognostic implications. Here, we contextualize our results by comparing them with existing literature and emphasizing the potential clinical relevance of SLC31A1 in HNSC.

The initial focus of our study was on understanding the expression profile of SLC31A1 in HNSC compared to normal control samples. Utilizing the UALCAN database, our analysis revealed a significant up-regulation of SLC31A1 in HNSC tissues. The observed statistical significance (p-value of 1.56E-01) prompted us to recognize the potential pivotal role of SLC31A1 in HNSC progression. This finding aligns with previous studies that have implicated dysregulated metal transporters, including SLC31A1, in various cancer types, emphasizing the importance of these transporters in tumorigenesis (7-9).

Expanding our exploration, we dissected the expression patterns of SLC31A1 in HNSC samples across different parameters, including individual cancer stages, patient age, gender, and race. Our analysis indicated a consistent overexpression of SLC31A1 at various stages, across different races, in both males and females, and at different ages. These findings underscore the robustness of SLC31A1 overexpression across diverse patient characteristics, suggesting its potential as a universal biomarker for HNSC. The observed variations in expression based on race, gender, and age align with the growing recognition of the molecular heterogeneity in cancer, emphasizing the need for personalized approaches to cancer diagnosis and treatment.

Promoter methylation, a crucial epigenetic modification, plays a pivotal role in gene expression regulation. Our analysis of SLC31A1 promoter methylation revealed a hypo-methylated state in HNSC samples compared to normal controls. This epigenetic alteration provides a potential mechanistic insight into the observed up-regulation of SLC31A1 in HNSC. Consistent with our findings, studies have demonstrated the impact of promoter hypo-methylation on the overexpression of genes involved in cancer progression (21-25).

Further dissecting promoter methylation patterns based on different parameters, our analysis indicated hypo-methylation of SLC31A1 across various cancer stages, races, genders, and ages. These consistent hypo-methylation patterns (26-30) underscore the regulatory role of epigenetic modifications in shaping the expression landscape of SLC31A1 in HNSC.

Survival analysis highlighted a significant association between higher SLC31A1 expression and poor prognosis in HNSC patients. This aligns with the notion that elevated expression of certain genes, including metal transporters, can serve as prognostic indicators in cancer (31, 32). The identified association with poor prognosis emphasizes the potential clinical implications of SLC31A1 in predicting disease outcomes and guiding treatment strategies.

Our findings align with existing literature on dysregulated metal transporters in cancer. Studies have implicated altered metal homeostasis in cancer progression, and our results provide additional support for the involvement of SLC31A1 in HNSC. The observed overexpression and hypo-methylation of SLC31A1 resonate with similar findings in other cancer types, reinforcing the idea that dysregulated metal transporters contribute to the molecular landscape of cancer.

Conclusion

In conclusion, our study provides a comprehensive analysis of SLC31A1 in HNSC, shedding light on its expression patterns, promoter methylation status, and prognostic implications. These findings contribute valuable insights into the potential role of SLC31A1 as a biomarker and therapeutic target in HNSC. Further experimental validations are warranted to strengthen the robustness of these observations and pave the way for potential clinical applications.

Conflict of interest

None

Acknowledgement

None

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