HSF1 gene amplification and overexpression is associated with poor survival in head and neck squamous cell carcinoma patients.

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Abstract

Background: Head and Neck Squamous Cell Carcinoma (HNSCC) is an aggressive life- threatening disease associated with high mortality rates. HNSCC is genetically complex and difficult to treat. Collagen triple helix repeat containing 1 is a protein that in humans is encoded by the HSF1 gene.

Aim of the study: To analyse the expression of HSF1 gene in HNSCC and identify its role as a prognostic marker in HNSCC. To the best of our knowledge, this is the first study till date to provide strong evidence on the association of HSF1 gene expression with HNSCC.

Methodology: The HSF1 gene expression was analyzed using web tools (UALCAN) from the Cancer Genome Atlas (TCGA) data. Then, the genetic alterations of HSF1 were examined by the cBioPortal database. Moreover, the prognostic values of HSF1 in HNSCC patients were investigated *via* the Kaplan-Meier plotter.

Results: We observed that the mRNA expression level of HSF1 was increased in most cancers compared with normal tissues, especially in HNSCC. In addition, we also used Kaplan-Meier plotter to evaluate the prognostic value of HSF1 in HNSCC patients. It showed highly expressed HSF1 was significantly related with poor Overall Survival (OS) in HNSCC patients.

Conclusion: Our results indicate that the HSF1 amplification and over-expression could be considered as a prognostic marker for HNSCC.

Keywords: HNSCC, HSF1, Novel variants, Amplification, Expression.

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Introduction

Head and neck cancer is the 6th leading cancer among other cancers worldwide. Squamous cell carcinoma is the most common type of head and neck cancer. Around 90% of squamous cells are started as head and neck cancer and some other types of head and neck cancer arise from lymphomas, adenocarcinomas, and sarcomas cells. There are several primary risk factors associated with head and neck cancer, which include tobacco use, consumption of alcohol, Human Papillomavirus (HPV) infection, and Epstein-Barr Virus (EBV) infection. There are annually 650,000 people affected by head and neck cancer and around 300,000 patients die from this type of cancer [1]. Most males are highly affected by this cancer than females, the ratio is 4:1. In several regions the incidence in the male is 20 in 100,000, the regions include France, Europe, India, Hong Kong, Spain, Italy, and Brazil [2,3]. We can reduce the risk of head and neck cancer by avoiding using tobacco and alcohol because they are the primary factor. The HPV vaccination helps prevent HPV infections, which may prevent head and neck cancer [4,5].

Recent studies in molecular genetics revealed the majority of Head and Neck Squamous Cell Carcinomas (HNSCCs) have arisen within a contiguous field of preneoplastic cells [6-8]. Based on several studies conducted, it can be inferred that these alterations in several cellular molecules including DNA, RNA, and proteins play a significant role in tumor progression and the overall survival of the malignant cells [9,10]. Our recent studies also showed that DNA, RNA, and protein alteration are associated with several diseases [11-17]. Hence these markers can assist in early diagnosis and prediction of prognosis. The diagnosis of HNSCCs at an early stage can prevent extensive treatment and thus biomarkers can serve as a tool for diagnosis [18,19] .

Heat Shock Response Protein-1 (HSP1) was discovered in 1962 and it has been the master regulator of the heat shock response [20]. HSF1 mediated heat shock protein expression to protect the proteome and withstand these acute stresses. Moreover, HSF 1 plays an important role in various diseases, maybe none more important than cancer. HSF1 appears to have a pleiotropically function in cancer, including invasion, proliferation, and cell metabolism, by encouraging several aspects of malignancy [21]. Due to these and other functions of HSF1, patient outcomes in multiple forms of cancer were investigated as a biomarker. Expression of HSF1 alone was predictive of the outcomes of patients in many cancer types, but HSF1 activity markers were more predictive in other cases [22]. In addition to its role in tumor growth and progression, HSF1 is most closely associated with the regulation of the response to heat shock. The HSF1 gene is located on the

chromosome 8 long arm (8q24.3), with a gene of 23,135 bp and 13 exons. The resulting 2,193 bp mRNA and 529 amino acid proteins of HSF1 are expressed under physiological conditions.

HSF1 regulates the expression of Heat Shock Proteins (HSPs) including HSP27, HSP70, and HSP90. HSF1 regulates cellular metabolism, including glycolysis and lipid metabolism. It also serves as a regulator of different signaling pathways, such as HuR-HIF-1, Nuclear Factor-kappaB (NF-κB), PI3K-AKT-mTOR, Mitogen-Activated Protein Kinase (MAPK) pathways, slug, and Protein Kinase C (PKC). HSF1 also plays a crucial role in the regulation of microRNAs (miRNAs) and long non-coding RNAs (lncRNAs). Role of HSF1 in cancer by anti-apoptosis, regulating cell proliferation, Epithelial-Mesenchymal Transition (EMT), metastasis, migration, invasion, and maybe a potential therapeutic target for human cancers [23]. HSF1 is not frequently mutated in cancers, but 15% of breast cancer reported increased mRNA expression. Besides, 25% of breast cancer has HSF1 mRNA upregulated which suggests that, apart from gene amplification, other mechanisms that increase the expression of HSF1 [24]. The expression of HSF1 and its association with cancer susceptibility and progression is a controversial topic. Hence more research in the area of HSF1 gene expression and analysis of the association between this HSF1 gene expression and tumor susceptibility progression of cancer should be promoted. This present study aimed to analyze the HSF1 gene expression in HNSCC which is an aggressive malignancy with high morbidity and mortality rates.

Materials and Methods

Gene expression analysis

The present study initially analyzed the HSF1 expression in HNSCC (n=520) and normal tissues (n=44) using data from the TCGA dataset. We used the UAlCAN database to analyze the HSF1 mRNA expression in primary HNSCC and normal tissues.

Survival analysis by Kaplan-Meier Plotter

In the present study, the prognostic values of HSF1 at mRNA level in HNSCC was analyzed using Kaplan-Meier Plotter is an online database containing gene expression profiles and survival information of cancer patients.

Results and Discussion

In many cancers, HSF1 is either overactive or exaggerated, but it loses activity in neurodegenerative diseases that result in neuronal death due to the unfolding protein response. In several fields of tumor biology, HSF1 has now been recognized as having a clear link to patient outcomes. Although the heat shock response is linked with cancer for decades, HSF1 has altered its role in prostate cancer and was first mentioned in 2000 **[25]**. HSF1 is shown to have altered expression or function in various cancers subsequently, such as breast cancer, colorectal cancer, gynecologic cancer, hepatocellular carcinoma, lymphomas, melanoma, oral cancer, pancreatic and other cancers [26].

In the present study, the HSF1 expression in HNSCC was first determined using the UALCAN database. We found that HSF1 was highly expressed in various types of cancer including HNSCC (Figure 1; p<0.05).

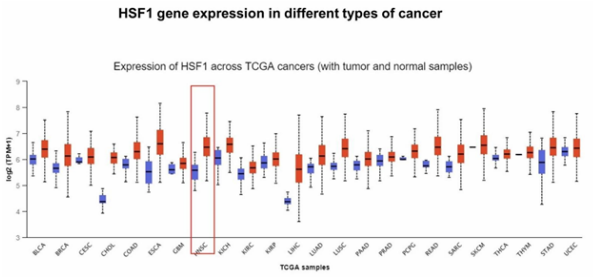

Figure 1. *HSF1 mRNA expression pattern in normal and tumor tissues (UALCAN database; p<0.05). The X-axis shows the types of tissues HSF1 were expressed and Y-axis exhibits log2 fold change values of HSF1 expression. Red boxes represent tumor tissues; blue boxes represent normal tissues. HSF1 mRNA is highly expressed in different types of cancer including HNSCC.*

Also, the UALCAN database was used to evaluate the exact HSF1 mRNA expression in HNSCC and normal tissues. We found that the mRNA level of HSF1 was significantly up-regulated in HNSCC compared to normal tissues (Figure 2; p<0.05).

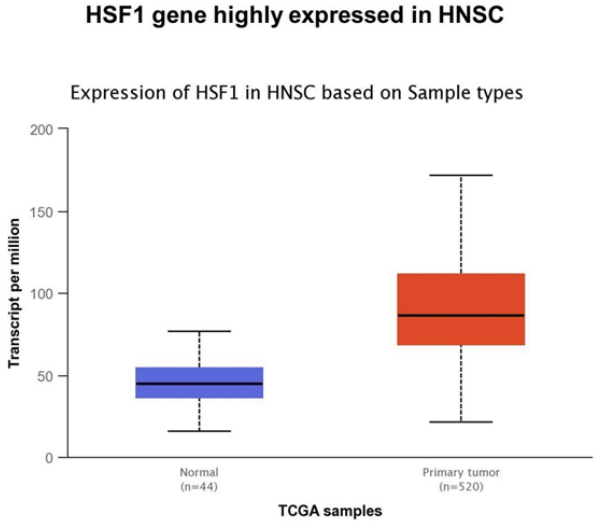

Figure 2. *Expression of HSF1 in head and neck squamous cell carcinoma based on sample types (P<0.05). The X-axis shows a comparison between normal and primary tumours and the Y-axis shows transcript (HSF1) per million.*

HSF1 has various functions in cancer to promote tumorigenesis and tumor progression. HSF1 supports malignancy by the cancer-specific transcriptional program. HSF1 plays a role in

the invasive and migratory capability of cancer cells, and although in metastasis [27]. HSF1 plays various roles in tumor biology from repairing DNA to angiogenesis to metabolism [28,29].

These functions support the HSF1 gene to malignancy of cancer cells. HSF1 inhibitors are developed in preclinical *in vivo* studies, in the future they may be treated successfully in humans.

Recent studies reported that high expression of HSF1 is associated with cancer patient's prognosis. In the present study, high HSF1 expression was found related to poor survival rates in HNSCC patients (p=0.057, Figure 3).

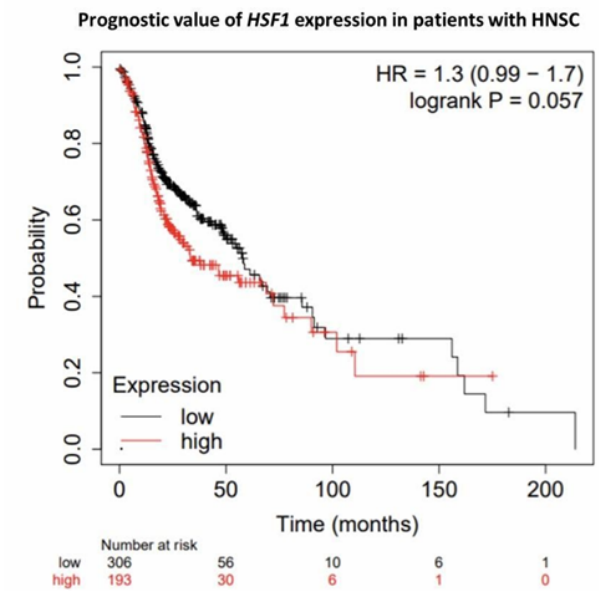

Figure 3. *Kaplan-Meier curves indicated that HNSCC patients had poorer overall survival with high expression of HSF1 mRNA (p=0.057) . Redline shows the cases with highly expressed HSF1 and the black line is indicated for the cases with lowly expressed HSF1. The Y-axis is the survival probability, and the X-axis represents time (months).*

High expression of the HSF1 gene can be associated with poor overall survival in HNSCC patients [30].

Apart from HNSCC, gall bladder carcinoma, breast cancer also showed decreased overall survival rate associated with increased expression of the HSF1 gene [31].

HSF1 is a strong predictor of a patient with cancer, especially breast and colon cancer, so it could be a possible biomarker in further studies of HSF1.

The present study results were also following previous literature in the case of increased HSF1 expression and decreased overall survival rate of HNSCC patients.

Mutations in HSF1 gene associated with different types of cancer, we also found HSF1 amplified in 11% of HNSCC patients, suggesting dysregulation of HSF1 associated with HNSCC development (Figure 4).

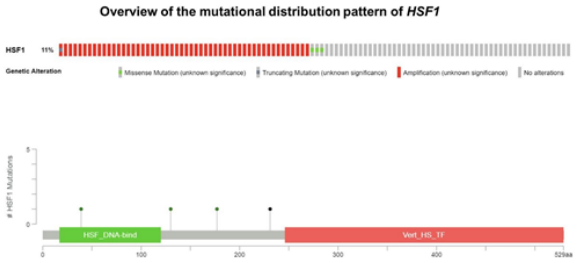

Figure 4. *Overview of the mutational distribution pattern of HSF1. The upper panel represents the mutational distribution pattern of HSF1 in HNSCC patients. The lower panel represents mutations in HSF1.*

Conclusion

Thus the present study provides information on HSF1 gene alterations that are associated with tumorigenesis in head and neck squamous cell carcinoma. In conclusion, this data demonstrates that HSF1 plays an important role in the oncogenesis of HNSCC.

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Authors Contribution

All the authors contributed equally in concept, designing, carrying out the research and analysis of the study.

Conflict of Interest

The authors declare no conflict of interest.

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