

Original Article

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A Brief Analysis PIK3CA Expression in Head and Neck Squamous Cell Carcinoma

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Abstract:

Background: Head and neck squamous cell carcinoma (HNSCC) are the 6th common carcinoma worldwide. PIK3CA gene often dysregulated in many cancers including HNSCC. Here, we aimed to observe PIK3CA expression in HNSCC using publicly free common dataset like TCGA and using bioinformatics tool GEPIA2.

Material and methods: Bioinformatics tools often used to predict overall and survival analysis. Here, we used the TCGA dataset to examine PIK3CA expression. We use GEPIA2 to investigate PIK3CA gene expression in HNSCC patients to view overall and disease-free survival.

Results: We observed that the most mutated gene is TP53 with 266 (number of cases gene mutated) out of 378 (number of simple somatic mutation) in contrast to PIK3CA gene 63 (number of cases gene mutated) out of 378 (number of simple somatic mutation). Overexpression of PIK3CA related to lower risk of death with treatment and lower risk of disease recurrence.

Conclusion: To understanding of the role played by the PIK3CA gene to establish a biomarker in head and neck squamous cell carcinoma (HNSCC).

Key words: HNSCC, PIK3CA, GEPIA2, TCGA.

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Introduction:

Head and Neck squamous cell carcinoma (HNSCC) is a heterogeneous cancer, arising from squamous cells of the mucous membrane of various anatomical regions including aero digestive tract, oral cavity, paranasal regions, nasopharynx, oropharynx, hypopharynx, and larynx[1].

The organs involved in speech, voice, smell, taste, and hearing—all essential structures for survival—can have their structure and function destroyed by HNSCC[2]. According to GLOBOCAN 2020, there are approximately 19.3 million new cancer cases and 10.0 million cancer deaths worldwide[3]. Head and neck cancer ranks sixth among all cancer types in the World with an incidence rate of 5.47% and a mortality rate of 5.13%[4]. Males are 60% more likely than females to get a tobacco-related cancer (NICRH and BSMMU cancer registry data). Additionally, this registry shows that more than 60% of cancer cases occur in people between the ages of 30 and 65 [5]. According to another study, 20% or more of all malignancies in Bangladesh are HNSCC [6].

PIK3CA (Phosphatidylinositol 3-Kinase) located at chromosome 3 (cytogenetic band 3q26.32), which is composed of 85 kDa regulatory subunit and 110 kDa catalytic subunit. PIK3CA often mutated in many cancer including HNSCC that may be loss of 3p and gain of 8q at their position [7, 8]. In most cases mutated PIK3CA act as oncogene and associated with HPV associated HNSCC [9], resulting alteration of cell cycle E2F1 gene[10].

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In HNSCC, there is 63% mutation occur as reported previously [11]including 79 missense and 1 inframe mutation and involve cell proliferation pathways [12, 13].In addition to HNSCC, PIK3CA mutations also involve in PI3K/AKT/mTOR signaling pathways and are linked to colorectal and breast cancers. [14].PI3K/AKT/mTOR signaling pathways are significant, even though tumor homeostasis may follow any of the ten hallmarks of cancer. Therefore, using TCGA and GEPIA2 datasets, we investigated PIK3CA expression (part of PI3K/AKT/mTOR signaling pathways) in HNSCC in this study.

Methods and materials:

First, we used the freely accessible public database GDC Data Portal at the TCGA [15] program to try and find the genes that were highly expressed in HNSCC.Second, from GDC data portal, we selected exploration section and selected total 524 HNSCC cases [Other and unspecified part of tongue (130), larynx (115), other and ill-defined sites in lip, oral cavity (71), floor of mouth (56), tonsil (46), other and unspecified parts of mouth (43), base of tongue (24), gum (11), oropharynx (10), hypopharynx (9), palate (5), lip (3) and other and unspecified major salivary gland (1)].Since our aim in the

literature review was to look at PIK3CA in HNSCC, we looked at PIK3CA expression using the TCGA dataset. Third, we usedbioinformatics tool GEPIA2[16] to investigate PIK3CA gene expression to explore overall survival and disease-free survival.

Results:

1. Genes overexpressed in HNSCC

Using the GDC data portal, we investigated the top 10 genes out of 713 genes that are often overexpressed or mutated in HNSCC (**Table 1**). Among the top 10 overexpressed/mutated we selected PIK3CA which stands top 8th position. There are 524 HNSCC cases overall in the GDC data portal, but only 507 of those cases have all of the accessible data.There were 713 gene alterations, 6630 mutations, and primarily squamous cell type neoplasms among the 507 patients. In the instance of PIK3CA, there were 86 cases in which the gene was altered, 507 cases were examined for simple somatic mutations, and 35 cases in the cohort had a unique simple somatic mutation.This indicates that overexpression in the form of 35 distinct simple somatic mutations in the instance of PIK3CA occur among the 507 cohort of HNSCC samples.

Table-I: List of top 10 genes commonly mutated in HNSCC

Gene Symbol	Gene name	Affected cases in cohort Number of cases where gene mutated/cases tested for simple somatic mutation	Number of Mutation (unique simple somatic mutation in the gene in cohort)
TP53	Tumor protein p53	357/507 (70.41%)	267
FAT1	FAT atypical cadherin 1	116/507 (22.88%)	138
MUC16	Mucin 16, cell surface associated	106/507 (20.91%)	156
CDKN2A	Cyclin dependent kinase inhibitor 2A	102/507 (20.12%)	55
CSMD3	CUB and Sushi multiple domains 3	101/507 (19.92%)	148
NOTCH1	Notch receptor 1	94/507 (18.54%)	98
LRP1B	LDL receptor related protein 1B	95/507 (18.74%)	124
PIK3CA	Phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha	86/507 (16.96%)	35
KMT2D	Lysine methyltransferase 2D	71/507 (14.00%)	79
NSD1	Nuclear receptor binding SET domain protein 1	56/507 (11.05%)	72

2. PIK3CA overexpressed in HNSCC

We use GEPIA2 (Gene Expression Profiling Interactive Analysis) to explore PIK3A expression in their dataset. In GEPIA2 dataset overexpression of PIK3CA found in tumor (number of tumor tissues= 519) and number of normal tissues is 44; Expression value $\log_2(\text{TPM}+1)$. $\log_2(\text{TPM}+1)$ in GEPIA2 represents gene expression level calculation, it is a logarithmic transformation of Transcripts Per Million (TPM). [TPM is transcript per kilobasemillion means how much RNA present in any sample, divided by RSEM (RNA sequence by expectation-maximization)]. PIK3CA is overexpressed in HNSCC tumors (red box) relative to normal tissue (grey box), with the number of instances shown by a dot as shown in **Figure 1**. Here, $\log_2\text{FC}$ (how one sample is different from another) cutoff value was 1 and p-value cutoff 0.01. This comprehensive analysis highlights TPM's diagnostic and prognostic significance in cancer, making it a valuable biomarker for disease detection and patient outcome prediction.

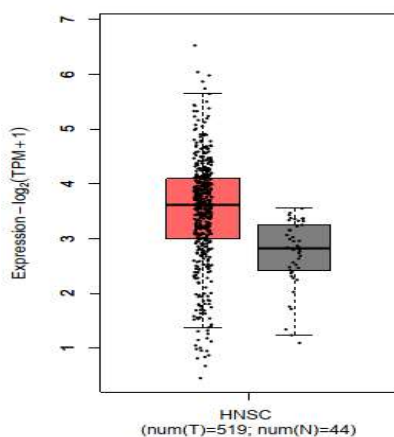


Figure 1: Overexpression of PIK3CA observed in HNSCC.

PIK3CA over expressed in HNSCC tumor (red) in comparison to normal tissue (grey) and dot represents number of incidence.

3. Analysis of Overall Survival and Disease-free survival

From GEPIA2, we categorized total 518 HNSCC patients in to two group; low PIK3CA expression (259) and high PIK3CA group (259), where median cutoff value was 50% each (as default). In case of overall survival hazard ratio

(HR) 0.94; nearer to 95% confidence interval, log rank p value was 0.65. That means lower risk of death with treatment (HR 0.94 indicates 6% lower risk of death), although no significant difference observed in survival (log rank p value was 0.65). On the other hand, disease free survival hazard ratio is 0.83; log rank p value was 0.26. That means Hazard ratio of 0.83 indicates lower risk of disease recurrence and Log rank p value of 0.26 suggests no significant difference.

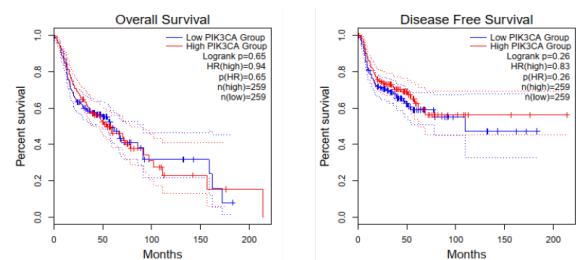


Figure 2: Comparison of overall and disease-free survival among high and low PIK3CA expressed groups. Between patients with high and low expression of PIK3CA mutations in HNSCC, overall survival was 94% and disease-free survival was 83%.

Discussion

Head and Neck squamous cell carcinoma (HNSCC) is a heterogeneous devastating disease [17]. The five years' overall survival of HNSCC is less than 50% [18] which warrants more importance for early diagnosis and monitoring of treatment outcome for this deadly disease. Therefore, the main purpose of our study was to establish a diagnostic biomarker in HNSCC.

Our goal in this study was to examine PIK3CA expression in the TCGA dataset and evaluate overall and disease-free survival in GEPIA2 dataset. PIK3CA overexpressed in the TCGA dataset cohort (**Table 1**). PIK3CA overexpression observed in tumor tissue versus normal tissue from GEPIA2 datasets (**Figure 1**). PIK3CA overexpression influenced the overall and disease-free survival of HNSCC patients in GEPIA2 datasets resulting lower risk of death with treatment and lower risk of disease recurrence, although no significant difference observed in OS and DFS (**Figure 2**). Using bioinformatics tool like GEPIA2, we will be able to suggest that overexpression of PIK3CA

resulting decreased risk of disease recurrence and lowered chance of mortality hence may be used as a biomarker in patients with HNSCC. Further research warranted to establish our suggestion.

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Conflicts of Interest Disclosure

The authors do not have any conflicts of interest to declare in relation to this work.

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