Examining NDUFAB1 Expression in Head and Neck Squamous Cell Carcinoma

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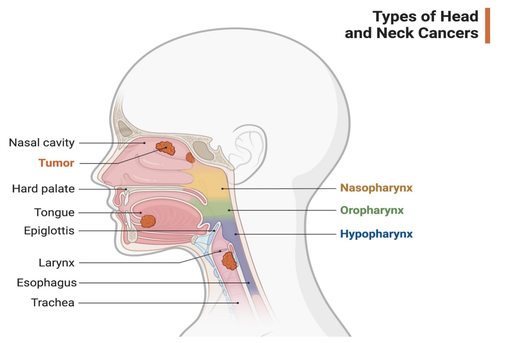
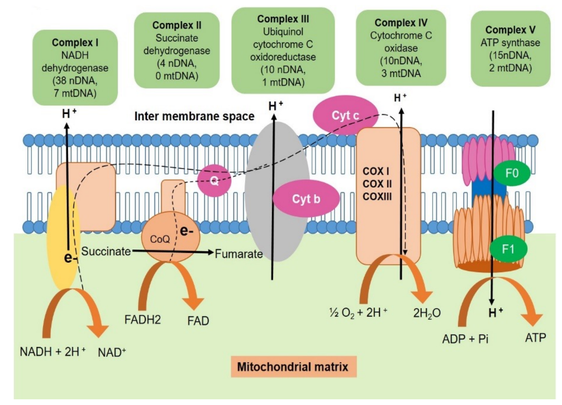
Introduction

- Head and neck squamous cell carcinoma (HNSCC) is approximately 4% of all cancers and 2% of all cancer associated mortality in the United States.[1]
- In 2023, there will be an estimated 67,000 new cases of HNSCC, along with 15,400 deaths, in the United States.[1]
- HNSCC locations include the oral cavity, oropharynx, nasopharynx, hypopharynx, and larynx.
- Major risk factors for HNSCC include tobacco use, alcohol use, and human papilloma virus (HPV).
- Epidermal growth factor receptor (EGFR) is currently the only approved molecular targeted therapy for HNSCC.
- Therefore, new therapeutics and biomarkers for HNSCC are warranted.



- Mitochondria are cytoplasmic organelles that are a major source of ATP through oxidative phosphorylation.
- Respiratory complex alterations reprogram metabolism and promote oncogenesis.
- Understanding respiratory complex alterations could provide insight for new therapeutic and biomarker development.
- NDUFAB1 is a nuclear-encoded protein of respiratory complex I (NADH dehydrogenase) in the electron transport chain.
- Previous studies have shown increased expression of NDUFAB1 in HNSCC compared to normal tissues.
- Previous studies also found abundant NDUFAB1 expression to be associated with clinical stage, tumor grade, HPV status, lymph node metastasis, and lower rates of survival.



Hypothesis and Specific Aims

Hypothesis: NDUFAB1 is abundantly expressed in HNSCC and predicts poor prognosis of these patients.

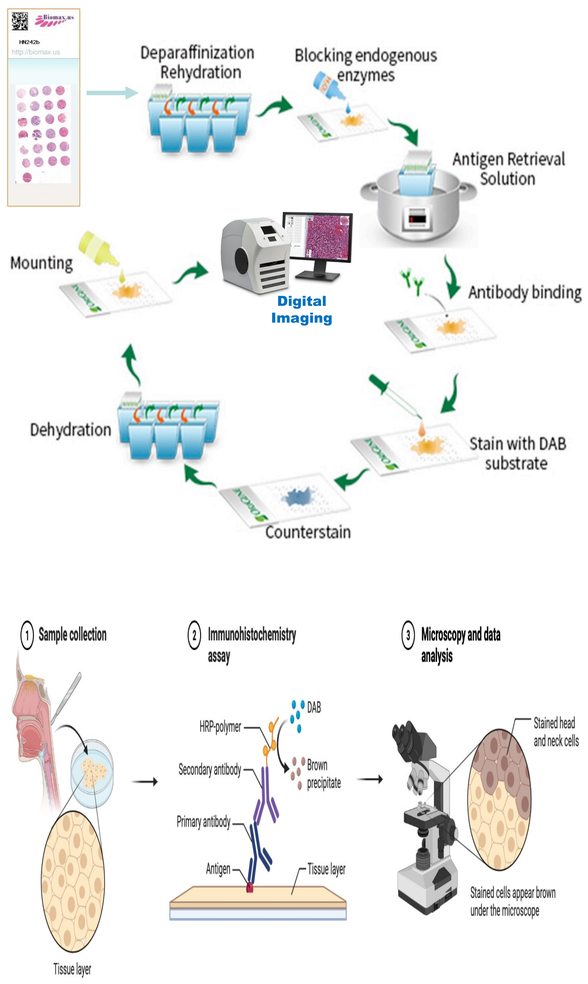
Specific Aim 1: Determine and compare NDUFAB1 protein expression in human head and neck cancer tissues to normal tissues by immunohistochemistry.

Specific Aim 2: Examine the clinico-pathological correlation of NDUFAB1 expression in predicting HNSCC outcome.

- This is accomplished by comparing data outcome from aim 1 with patients' clinical stage, histological grade, sex, age, and survival.

Methods

- Immunohistochemistry was performed on 10 normal tissues and 50 head and neck squamous cell carcinoma tissues.
- Tissues were formalin-fixed and paraffin-embedded.
- The digital microscope analyzes the whole tissue section from each subject.
- A three point intensity scale (+1, +2, +3), combined with a percent positivity of cells (0-100%) was used to determine the H-score of each tissue.
- H-score (0-300), which determines NDUFAB1 expression, was used as an index to compare outcome.



Results

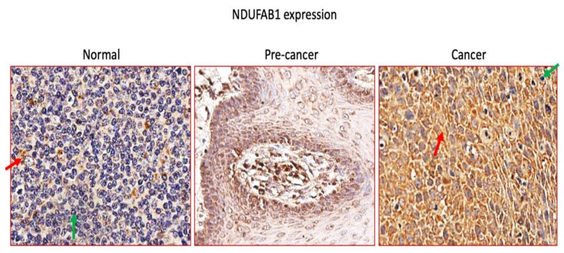


Figure 1. Differential expression pattern of NDUFAB1 in normal, precancerous, and cancerous oral tissues. NDUFAB1 appears brown after immunohistochemistry due to DAB staining (red arrows). Nuclei appear blue under digital microscope due to hematoxylin counterstaining (green arrows).

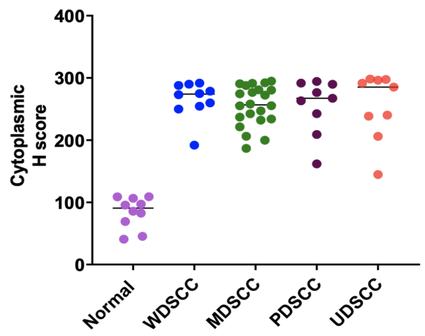


Figure 2. NDUFAB1 expression is significantly increased in the different progressive tumor grades of HNSCC patients compared to normal controls. Abbreviations: WDSCC = well differentiated squamous cell carcinoma, MDSCC = moderately differentiated squamous cell carcinoma, PDSCC = poorly differentiated squamous cell carcinoma, UDSCC = undifferentiated squamous cell carcinoma. The H-scores of each tissue were analyzed and compared after digital imaging.

Tissue Comparison	P-value
Normal vs Grade 1 (WDSCC)	<0.0001
Normal vs Grade 2 (MDSCC)	<0.0001
Normal vs Grade 3 (PDSCC)	<0.0001
Normal vs Grade 4 (UDSCC)	<0.0001
Grade 1 vs Grade 2 (WDSCC vs MDSCC)	0.4158
Grade 1 vs Grade 3 (WDSCC vs PDSCC)	0.5653
Grade 1 vs Grade 4 (WDSCC vs UDSCC)	0.6209
Grade 2 vs Grade 3 (MDSCC vs PDSCC)	0.992
Grade 2 vs Grade 4 (MDSCC vs UDSCC)	0.9955
Grade 3 vs Grade 4 (PDSCC vs UDSCC)	0.9921

Table 1. The increased expression of NDUFAB1 in tumor tissues is tumor grade independent. Abbreviations: WDSCC = well differentiated squamous cell carcinoma, MDSCC = moderately differentiated squamous cell carcinoma, PDSCC = poorly differentiated squamous cell carcinoma, UDSCC = undifferentiated squamous cell carcinoma. A student's t-test compared the H-scores of normal tissues to progressive tumor grades and the H-scores of progressive tumor grades to each other.

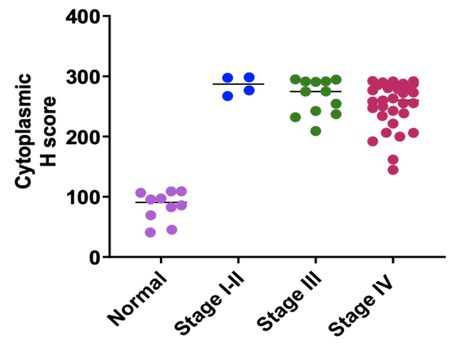


Figure 3. NDUFAB1 expression is increased in all the stages of HNSCC patients. H-scores of each tissue were analyzed and compared after digital imaging.

Results (cont.)

Tissue Comparison	P-value
Normal vs Stage 1-2	<0.0001
Normal vs Stage 3	<0.0001
Normal vs Stage 4	<0.0001
Stage 1-2 vs Stage 3	0.2386
Stage 1-2 vs Stage 4	0.114
Stage 3 vs Stage 4	0.291

Table 2. The significantly increased expression of NDUFAB1 in cancerous tissues does not correlate with the different stages. A student's t-test compared the H-scores of normal tissues to progressive clinical stages and the H-scores of progressive clinical stages to each other.

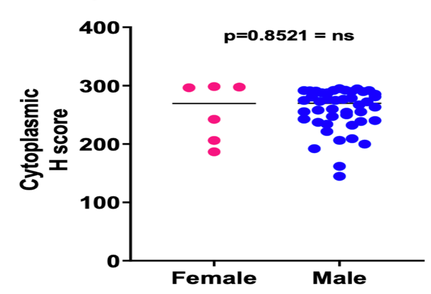


Figure 4. NDUFAB1 expression is independent of patient sex. A t-test compared the H-scores of cancerous tissues between male and female patients.

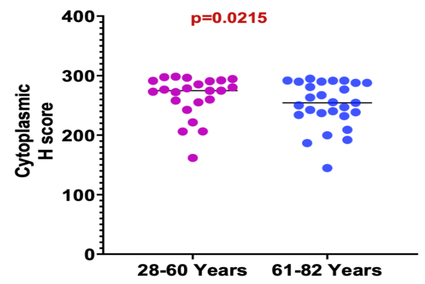


Figure 5. NDUFAB1 expression pattern is age-dependent. H-score of cancerous tissues from patients in two different age groups were compared using a t-test.

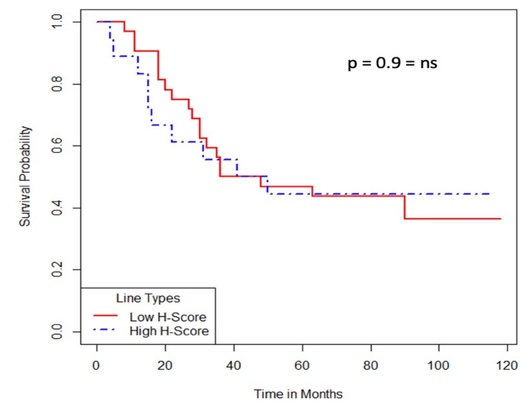


Figure 6. NDUFAB1 expression is not correlated with patient survival. H-score (NDUFAB1 expression) of the cancerous tissues was compared to the survival data collected from each of the patients.

Conclusions

- NDUFAB1 is overexpressed in HNSCC compared to normal tissues.
- NDUFAB1 is more expressed in all clinical stages and tumor grades and is associated with patient age.
- Abundant NDUFAB1 expression predicts poor prognosis of patients with HNSCC.
- NDUFAB1 bears potential for targeted therapeutic development in HNSCC.

References:
1. Cancer.Net. *Head and neck cancer - statistics*. (2023). https://www.cancer.net/cancer-types/head-and-neck-cancer/statistics.