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5-2023

## **Examining NDUFB1 Expression in Head and Neck Squamous Cell Carcinoma**

Addison Stevens

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# Examining NDUFAB1 Expression in Head and Neck Squamous Cell Carcinoma

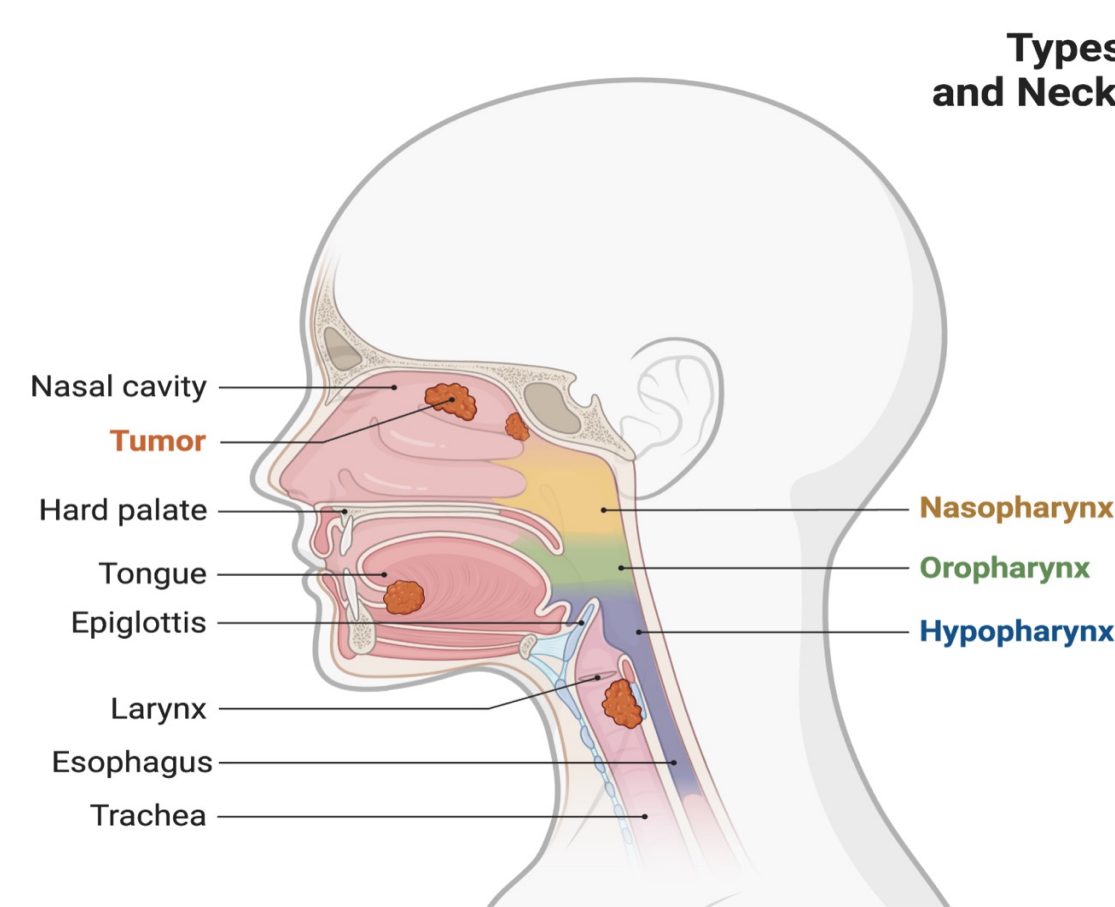
Addison Stevens<sup>1</sup>, Srijan Acharya<sup>2,3</sup>, Paramahansa Pramanik<sup>4</sup>, Elliot Carter<sup>3</sup>, Santanu Dasgupta<sup>2,3,5</sup>

<sup>1</sup>Department of Biomedical Sciences, <sup>2</sup>Mitchell Cancer Institute, <sup>3</sup>Department of Pathology, <sup>4</sup>Department of Mathematics and Statistics,

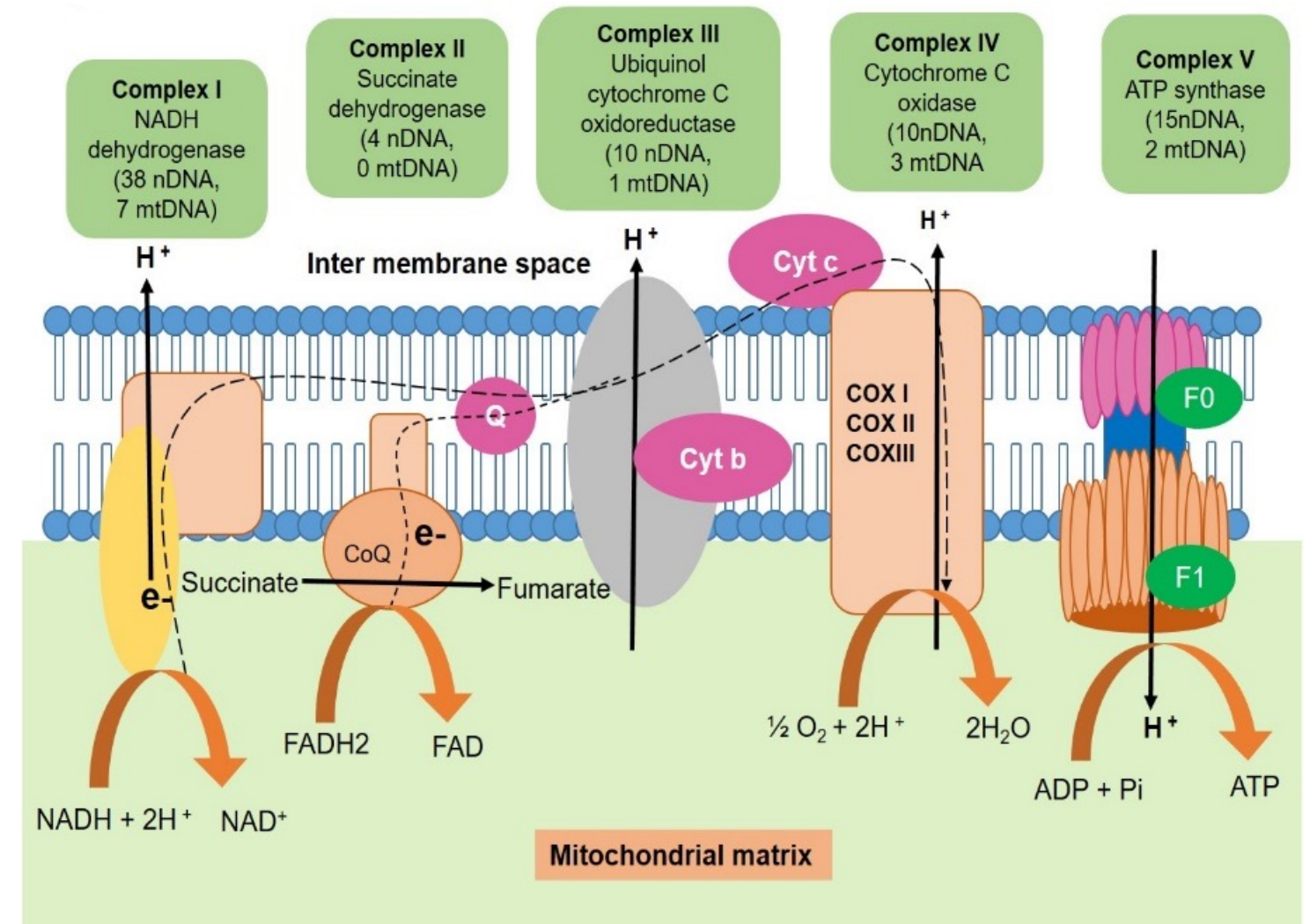
<sup>5</sup>Department of Biochemistry and Molecular Biology, University of South Alabama

## 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.<sup>1</sup>
- In 2023, there will be an estimated 67,000 new cases of HNSCC, along with 15,400 deaths, in the United States.<sup>1</sup>
- 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.

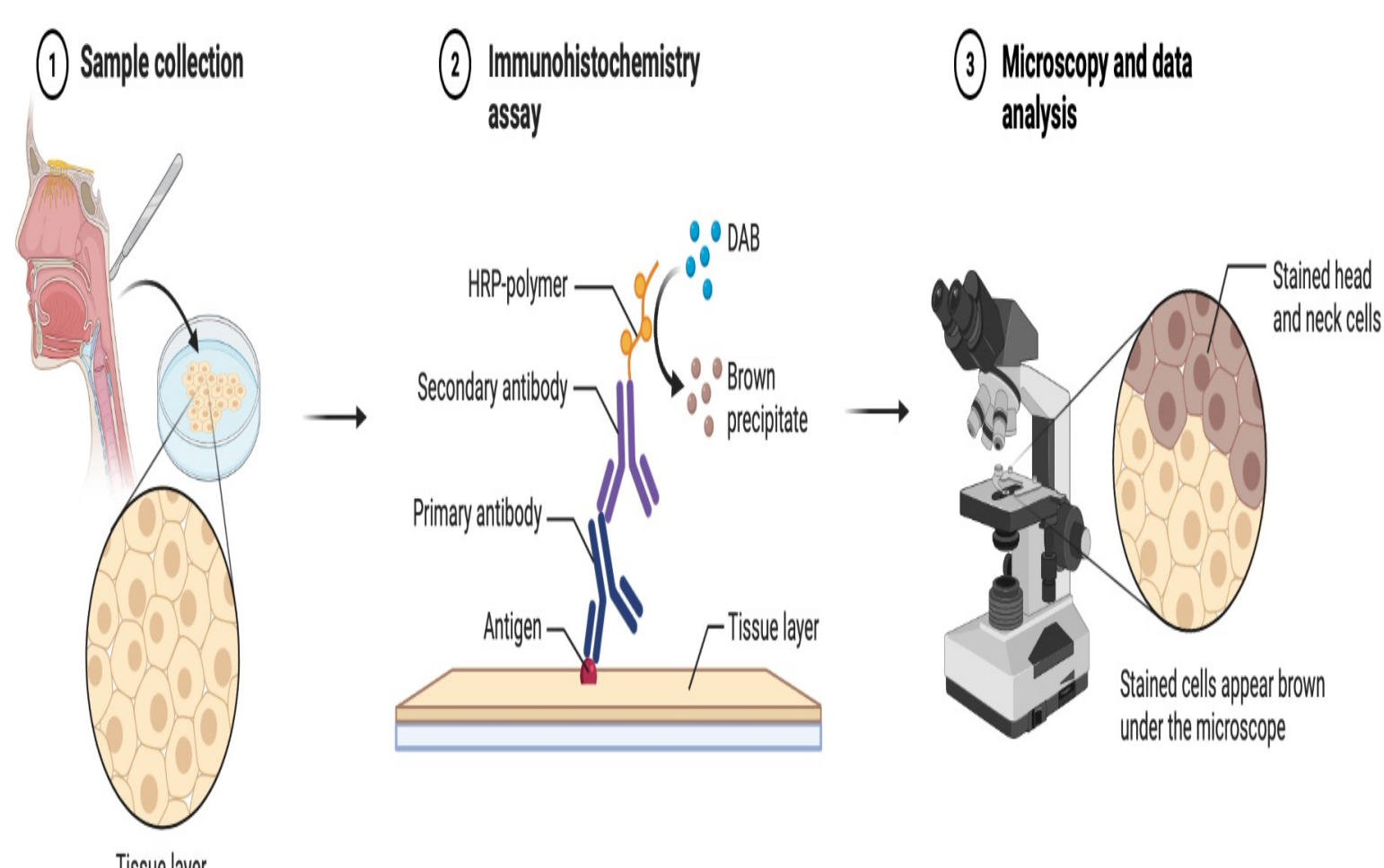
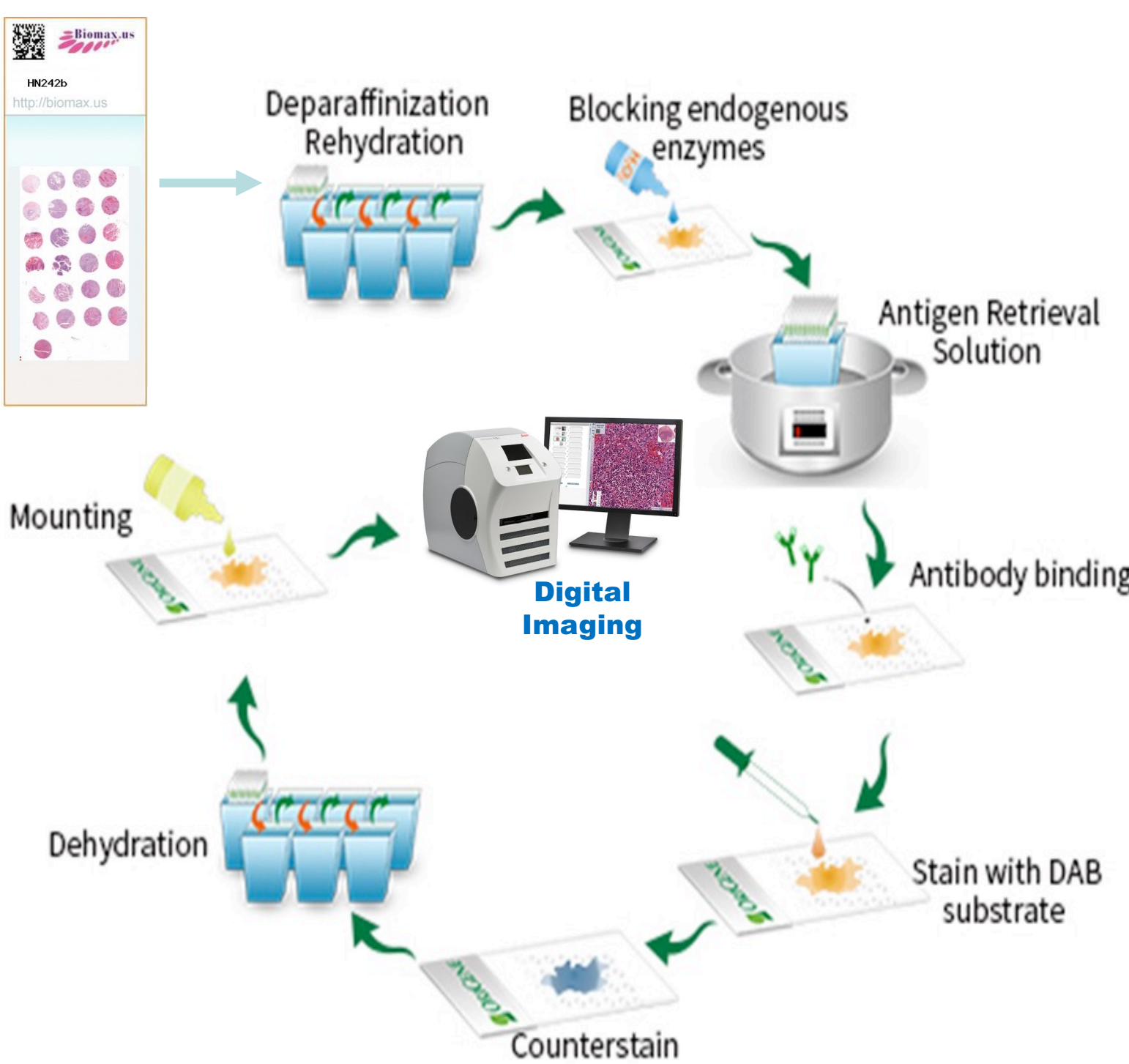
**Specific Aim 1:** Determine and compare NDUFAB1 protein expression in human head and neck cancer tissues to normal tissues by immunohistochemistry.

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

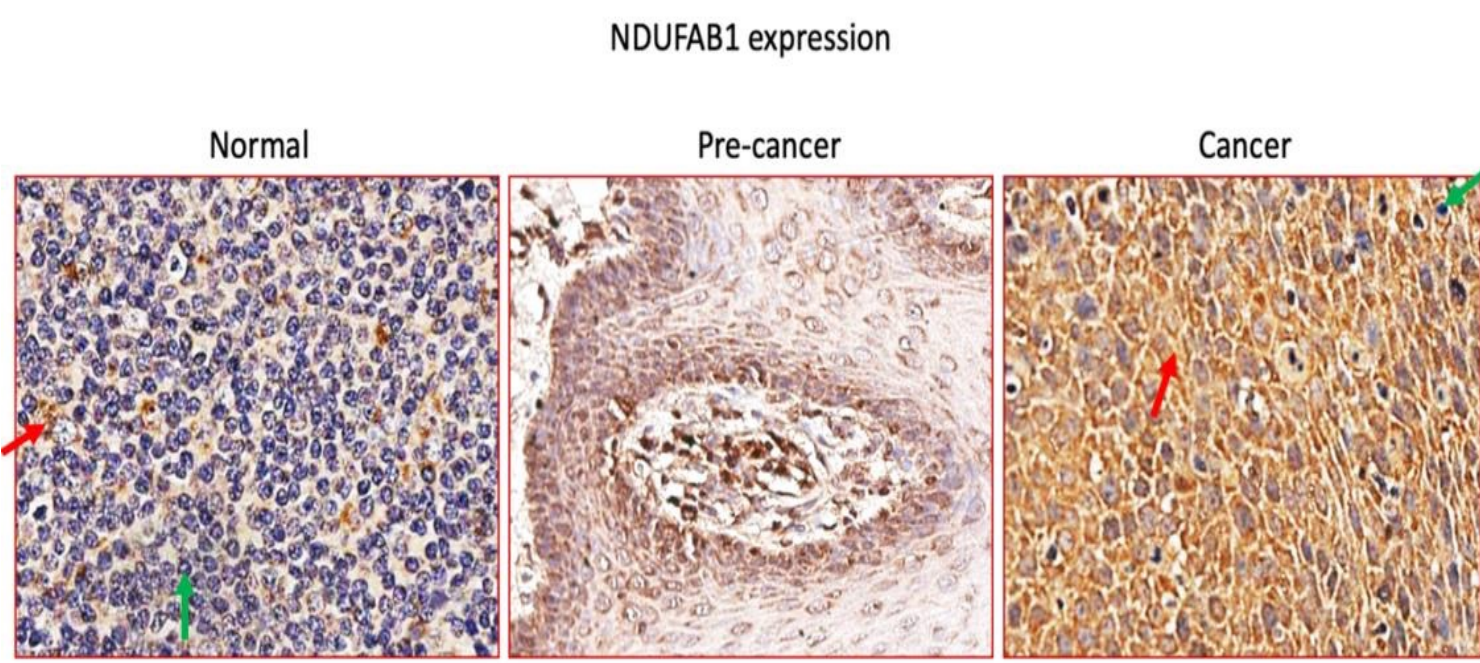
- This is accomplished by comparing data outcome from aim 1 with patients' clinical stage, histological grade, sex, age, and survival.

## Methods

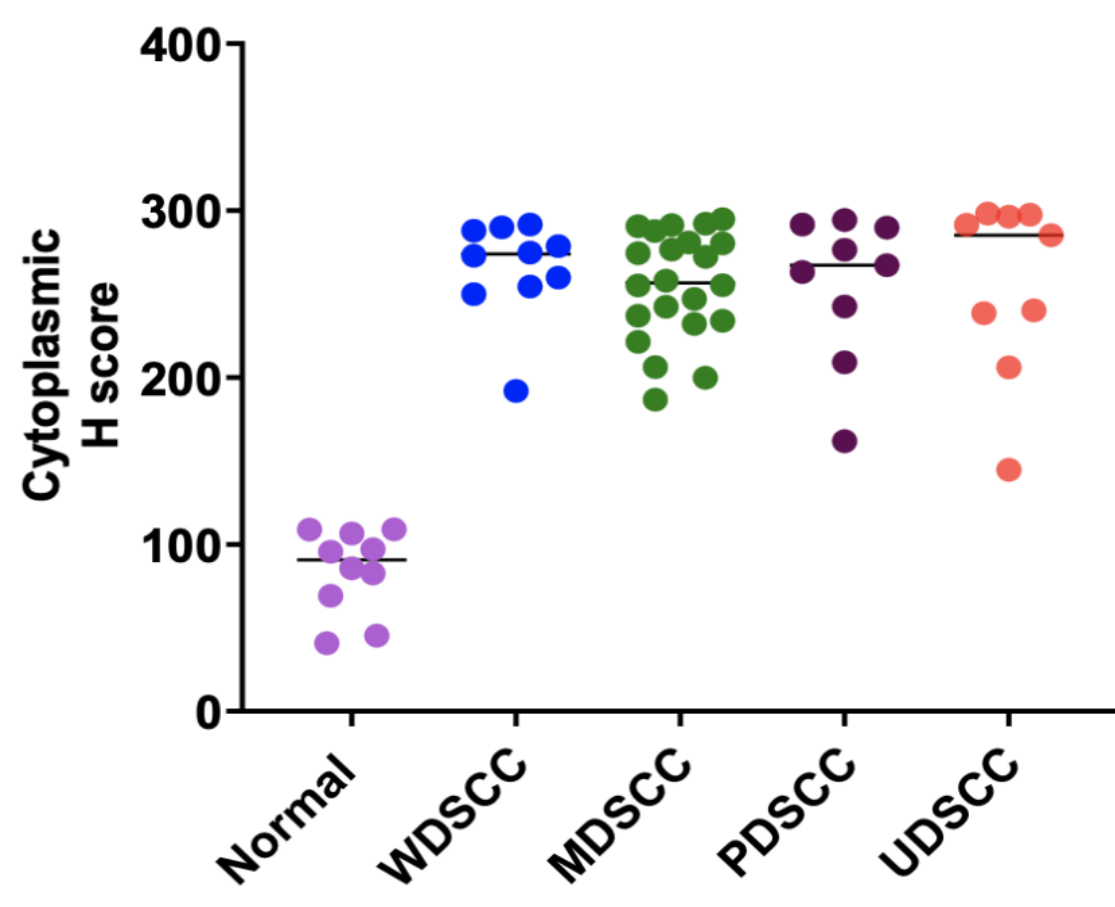
- Immunohistochemistry was performed on 10 normal tissues and 50 head and neck squamous cell carcinoma tissues.
- Tissues were formalin-fixed and paraffin-embedded.
- The digital microscope analyzes the whole tissue section from each subject.
- A three point intensity scale (+1, +2, +3), combined with 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



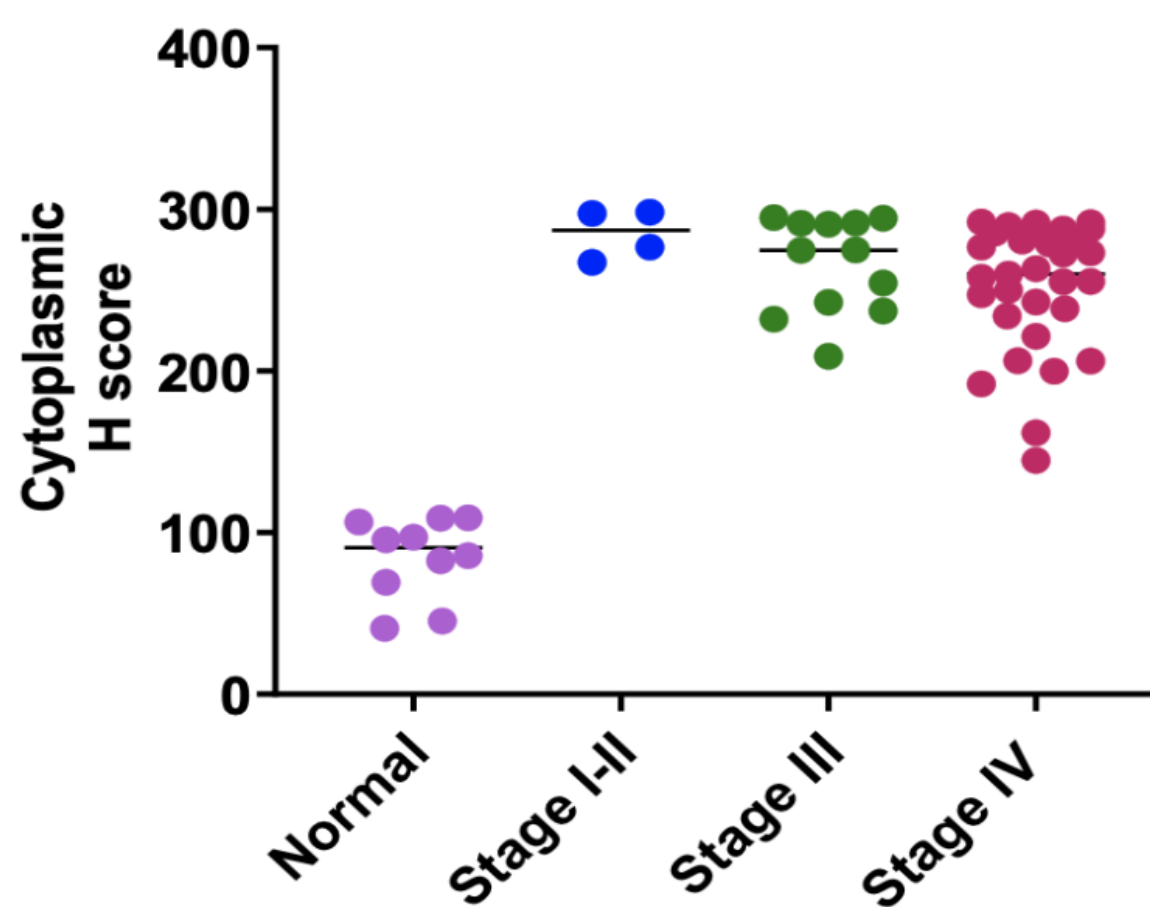
**Figure 1. Differential expression pattern of NDUFAB1 in normal, precancerous, and cancerous oral tissues.** NDUFAB1 appears brown after immunohistochemistry due to DAB staining (red arrows). Nuclei appear blue under digital microscope due to hematoxylin counterstaining (green arrows).



**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.

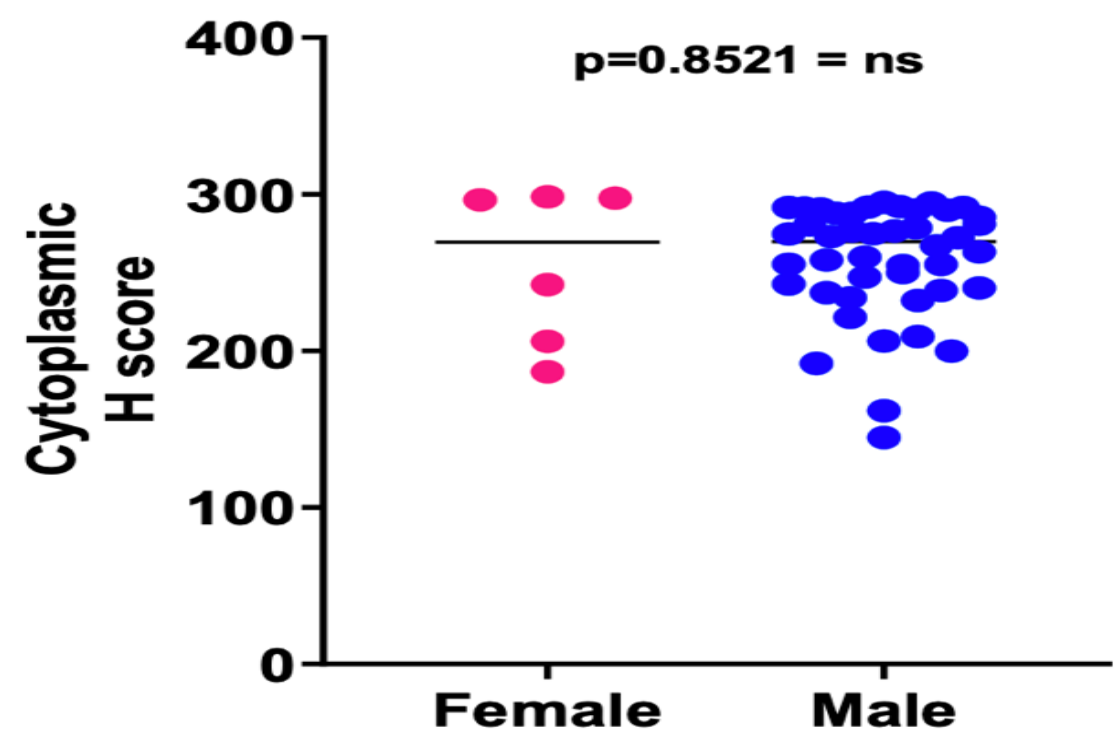


**Figure 3. NDUFAB1 expression is increased in all the stages of HNSCC patients.** H-scores of each tissue were analyzed and compared after digital imaging.

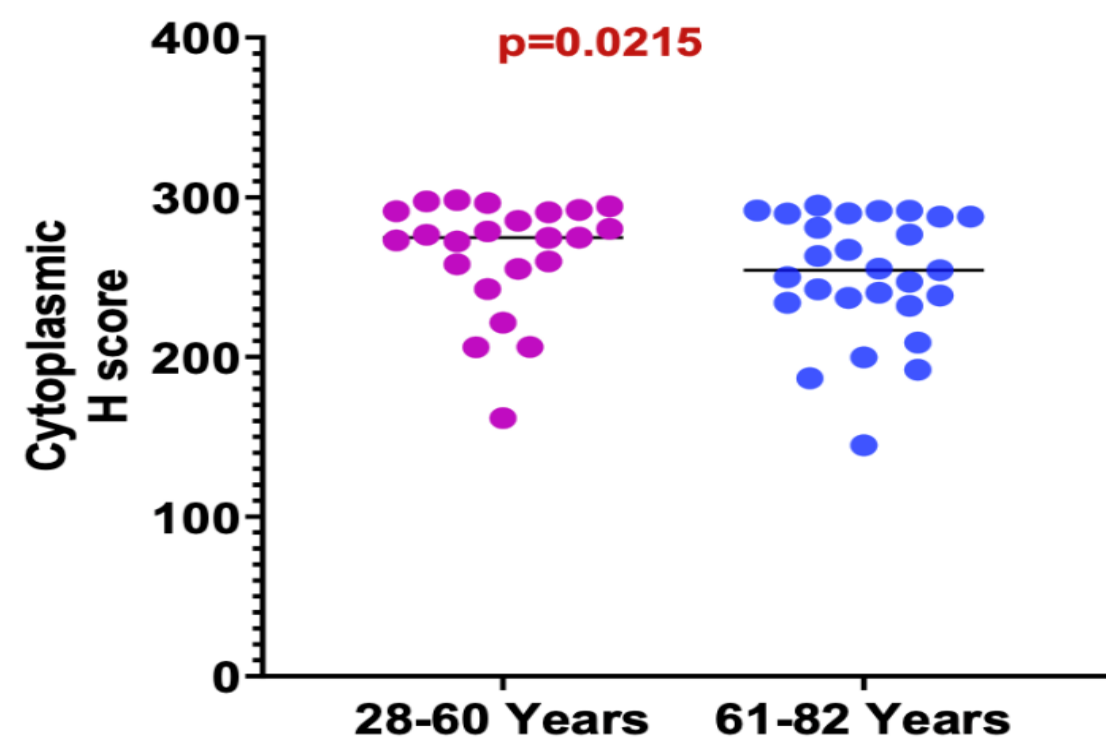
## Results (cont.)

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

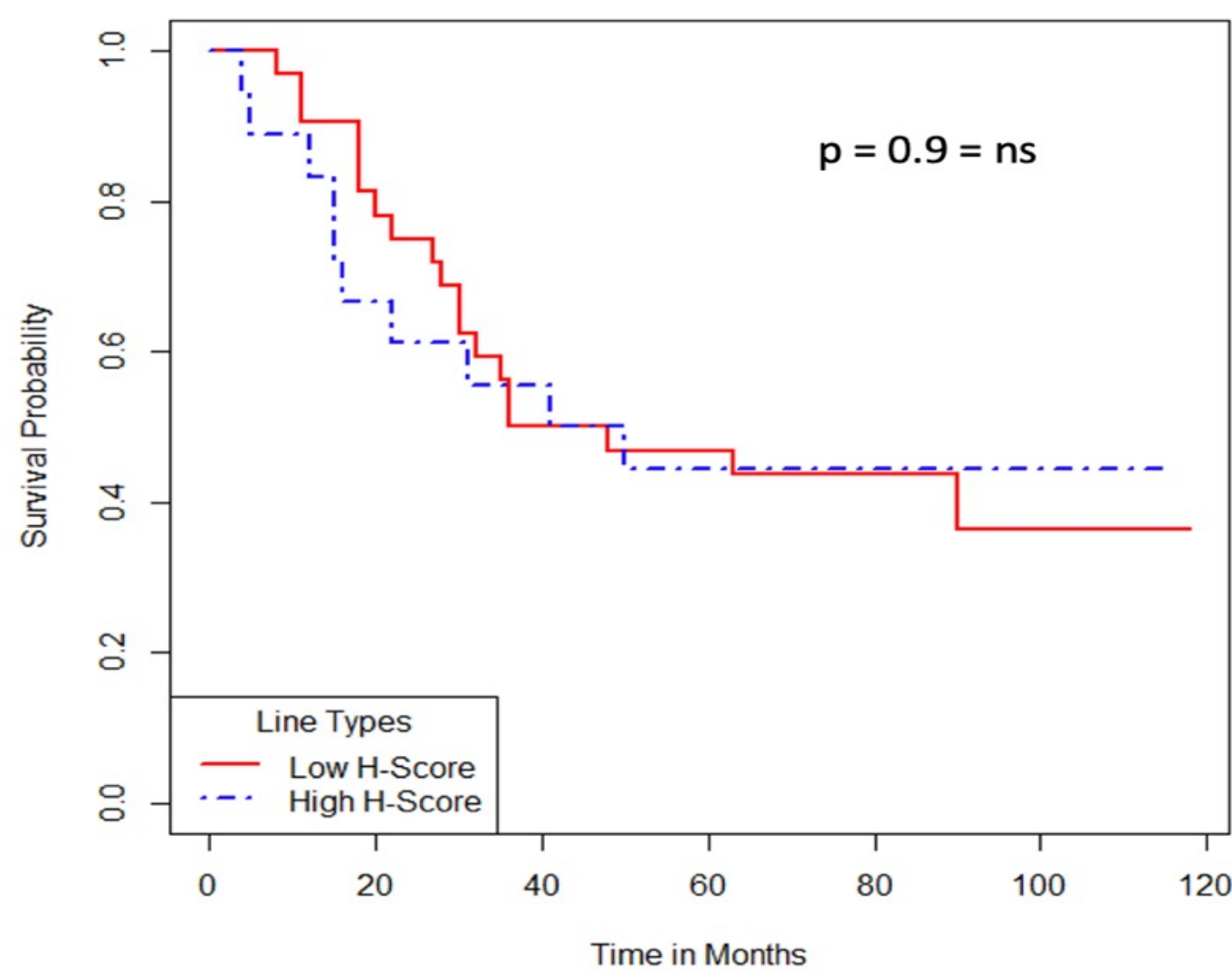
**Table 2. The significantly increased expression of NDUFAB1 in cancerous tissues does not correlate with the different stages.** A student's t-test compared the H-scores of normal tissues to progressive clinical stages and the H-scores of progressive clinical stages to each other.



**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>.