# Characterizing the Diversity of Heat Shock Proteins in *Nematostella vectensis* and *Saccharomyces cerevisiae*

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## Introduction

Heat shock proteins (HSPs) are a highly conserved family of proteins critical to both environmental stress response and cellular function under normal conditions. More specifically HSPs are chaperone proteins that work to refold other misfolded proteins. Data suggests that HSPs have neuroprotective effects against protein aggregation diseases such as Alzheimer's.

Previous research indicates that HSP70 has evolved multiple isoforms or variants in organisms such as the sea anemone *Nematostella vectensis* and *Saccharomyces cerevisiae* budding yeast. While the presence of these isoforms is established, the function and evolutionary history is not fully understood.

***Saccharomyces cerevisiae* under DIC microscope**
https://www.youtube.com/watch?v=PTGYLYru2Sw

**Adult *Nematostella vectensis* polyp.**
https://acumen.cas.lehigh.edu/content/neurogenesis-sea-anemones

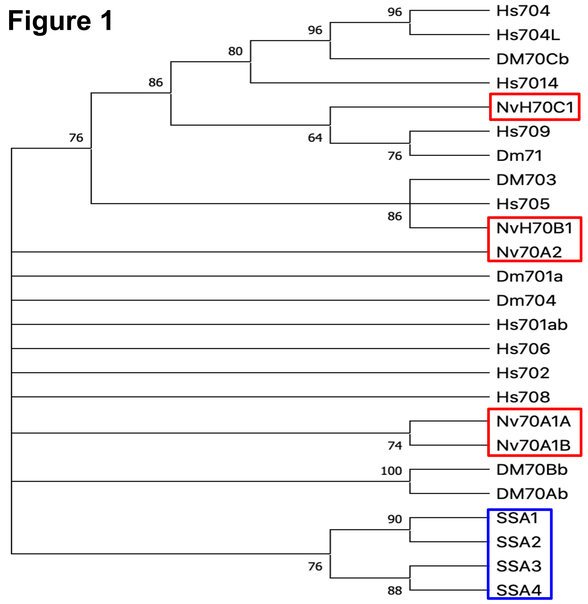
## Method

An alignment was provided containing the amino acid sequence for HSP70 isoforms in various model organisms. Using this alignment, the following was carried out:

1) Genetic analysis using the MEGAX software, denoting specific amino acid differences in each isoform.
2) Isolation of denoted changes into novel alignments.
3) Construction of initial phylogenetic trees through MEGAx.
4) Bootstrapping of generated trees through the RaXML software.

## Results

**Figure 1**



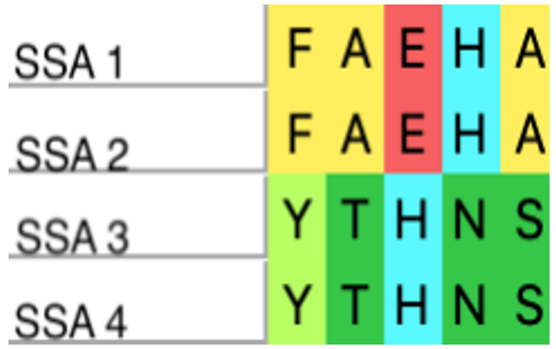

The Phylogenetic tree showcases the relationship between mimic sites in HSP70 isoforms of four different model organisms.

Nv = *Nematostella vectensis*
SSA = *Saccharomyces cerevisiae*
Hs = *Homo sapiens*
DM= *Drosophila melanogaster*

From further site analysis it was found that 40% of heavily characterized mimic sites evolved to S/T/Y in at least one homolog. It was also found that 88% of moderately characterized mimic sites evolved to S/T/Y in at least one homolog.

Another notable trend was found with isoforms HS709, NvH70C1, and Dm71. These isoforms evolved the same amino acid independent of their specific organism at multiple sites. Sites that exemplify this trend include # 71, 81, 172, 505, 861, 177, 169, and 696.

**Figure 2**




Site #s 90, 189 249, 250, and 317.

The sites shown in figure two illustrate a trend specific to *Saccharomyces cerevisiae*. Here SSA 1 and 2 see evolution independent of SSA 3 and 4.

## Objectives

The purpose of this research is to contribute towards a better understanding of the HSP70 isoforms. This project is guided by two broad goals:

1) To illustrate the phylogenetic relatedness of these isoforms within their specific organism and between other common model organisms.
2) To understand the functional diversity of HSP70 isoforms specifically in *Nematostella vectensis* and *Saccharomyces cerevisiae* budding yeast.

Focus will be placed on evolution of D and E amino acids to the phosphorylatable amino acids S,T, and Y. This type of evolution can alter both the structure and function of the isoforms.

## Data Collection

Data was collected mainly on sites including the following amino acids:

Serine (S), Tyrosine (T), Threonine (Y), Aspartic Acid (D), and Glutamic Acid (E).

Sites were considered heavily characterized by mimics if 12 or more isoforms conserved D/E at one site. If 5 or more isoforms conserved D/E, the site was considered moderately characterized by mimics.

All sites that were either moderately or heavily characterized were included into a new alignment and a phylogenetic tree was constructed.

Other sites were also isolated into novel alignments for analysis. These included sites that evolved from D/E to S/T/Y and those that evolved from D/E to a non-regulatable amino acid.

## Conclusions

The following conclusions can be drawn from analysis of the HSP70 alignment:

- S/T/Y sites do evolve, nonexclusively, from D/E sites. From the data it's seen that between 40-88% of S/T/Y sites likely evolve from D/E sites.
- Despite being present in different organisms, the mimic sites of HS709 and Dm71 are closely related. These isoforms have also evolved similarly to NvH70C1 at multiple sites.
- SSA 1+2 are likely to have evolved separately from SSA 3+4 based on both the phylogenetic tree and amino acid sites seen in the figures.

In terms of functional diversity, this data creates a baseline for future molecular experiments. More specifically the trends from this data can be used to choose specific amino acid sites for mutation.