Novel Study of Myosin Heavy Chain 16 and Skull Shape Evolution

Abstract

Myosin Heavy Chain 16 (MYH16) is a super fast-twitch masticatory muscle protein expressed in most organisms. In 2004, Dr. Hansel Stedman's lab discovered a mutation in MYH16 in the human lineage, and hypothesized that it led to the increased brain size in humans relative to non-human primates. Due to the lack of modern bioinformatics tools at their disposal at the time, Dr. Stedman could not fully prove or disprove this hypothesis, leading to intense debate. In an attempt to provide more information on this subject, this project seeks for a relationship between the presence/absence of MYH16 and brain size relative to the organism's body. This study found that for organisms with mutated MYH16, there was a correlation between mutation and brain size, but for organisms with deleted MYH16, there was no such correlation. Though not a definitive answer, the results of this study provide an intriguing modification to Dr. Stedman's original theory.

Question & Hypothesis

Question: Is there a correlation between animals having mutated or no MYH16 and having a significantly changed skull shape or brain size from their most related unmutated counterparts?

Hypothesis: There is a correlation between MYH16 mutations and deletions and increased brain size since decreased force on the skull allows for the removal of skull crests, making room for a larger brain cap.

Background: MYH16

- Understood to be one of the strongest Myosins
- Located in masticatory muscles
- In 2004, Stedman Labs discovered a frameshift deletion in humans
- Stedman traced the mutation back to around 2.4 MYA
  - Roughly around the evolution of early humans
- Stedman hypothesized that MYH16's mutation in the human lineage led to increased brain-cap size
  - Less force on the skull means more room for the brain

Current Theories on Skull Shape Evolution

- Dean Falk proposed the "radiator theory"
  - Bipedalism increased blood flow to the brain
  - Supported by Dr. Gabrielle Russo with his note of the brain size change happening around the development of bipedalism
- Daniel Lieberman proposed the "soft-food theory"
  - Humans eating soft cooked foods allowed for jaw shape adjustments that made room for a larger brain cap

Materials

- MacVector (MacVector Inc.): Sequence analysis application
- Ensembl.com: Genome database from European Bioinformatics Institute
- BLAST: A software function where genome databases are searched for sequences matching the "blasted" sequence
- FGenesh+ (Softberry.com): Genome and protein analysis cloud computing for predicted proteins
- Phyre2 servers: Protein structure prediction
- PyMol modeling software: Molecular visualization software
- University of Illinois Urbana-Champaign Body and Brain Weights in the MSTE Data Archive
- Brain Facts and Figures University of Washington Data Archive

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Phase 1: Creating the Tree

1. Align all human Myosins (MacVector)
2. Trace back MYH16 with gene synteny and take one sample per major group (Ensembl)
3. Add new samples to the alignment (MacVector) and generate tree
4. Check that all samples are aligned in accepted evolutionary pattern

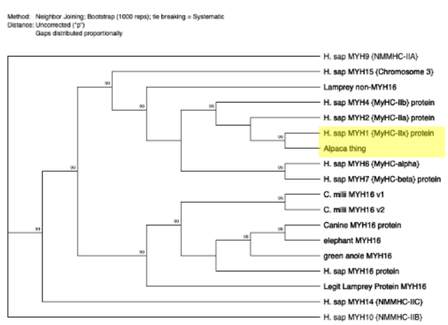
Phase 2: Finding Organisms for the Study

1. Use Ensembl and gene synteny to find proposed MYH16
   1. Where step 1 is impossible, use BLAST software to find possible MYH16
2. Check the MYH16 sample against the tree from phase 1
3. If the sample is MYH16, check for mutations with a Pustell Protein-DNA cross matrix and Pustell Protein Matrix (MacVector)
   1. For uncertain samples, model the protein on Phyre 2 servers
4. Record organisms with their MYH16 status

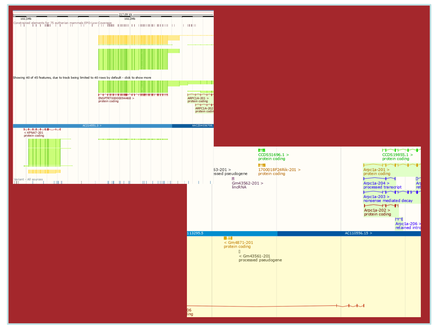
Phase 3: Statistical Analysis

1. For recorded organisms, find mass, brain mass, and skull pictures on college databases
2. Calculate Encephalization Quotient (EQ) for each organism
3. Compare EQ to MYH16 status
4. Look for correlations using Correlation Coefficient and T-Tests

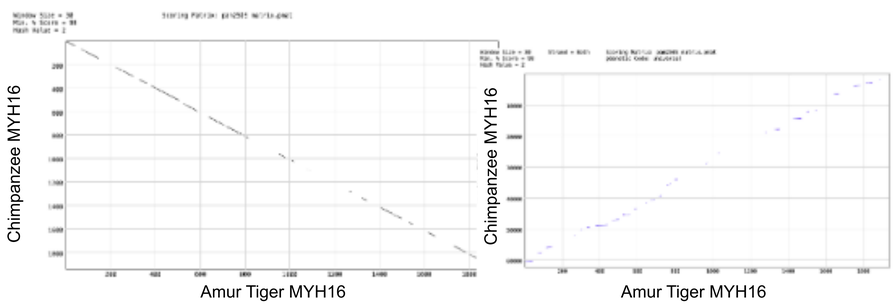
1- Final Phylogenetic Tree



2 - Genome Maps

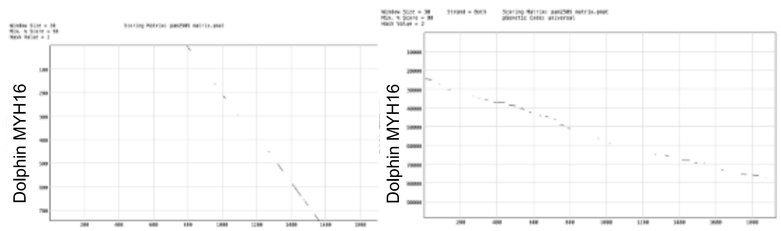


2- Unmutated Example Matrices



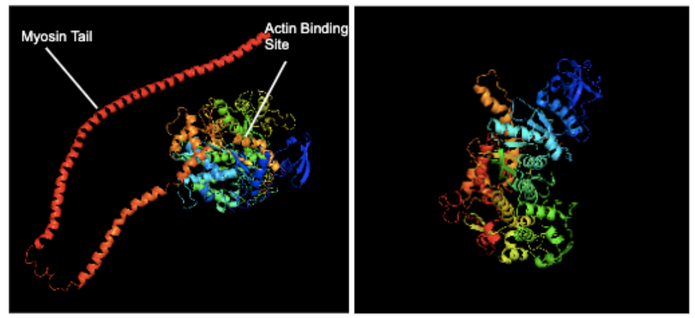
Note the clear linear relationships that connect with the axes at the corners of the matrices

2- Mutated Example Matrices



Note that the sequences lack alignment by the shortened lines that connect with the axes too soon

2- MYH16 Models Using PHYRE 2 and PyMol



Chimpanzee (unmutated) Human (mutated)

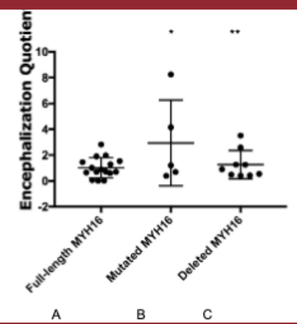
3 - Average EQ and Correlation Coefficient

MYH16 Status (n)	Unmutated MYH16 (16)	Mutated or Deleted MYH16	Deleted MYH16 (9)	Mutated MYH16 (5)
Average EQ	1.02	2.34	1.27	2.94
Correlation Coefficient Between EQ and MYH16 Status	N/A	0.12	0.19	0.46

$$EQ = brain{-}weight \,/\, (0.12 \times body{-}weight^{((2/3))})$$

3- Comparison of MYH16 Status and EQ

The T-Test shows a statistically sound increase in EQ for organisms with mutated MYH16, but not for organisms with deleted MYH16



*p<.05 Vs. A

**p = NS vs. B and vs. A

Conclusions

There was a direct correlation between frameshift deletions in MYH16 and increased EQ, but there was no similar correlation for deletions (removals) of MYH16 and EQ. This would imply that mutated MYH16 somehow plays a role in skull development more profound than the role of the removal of MYH16 when compared to those with normal MYH16.

New Hypothesis: mutated MYH16 binds to actin, but due to its lack of a tail, it exerts no pull, and only takes up binding space. It may even destroy muscle tissue, allowing for less exertion on the skull and the removal of skull crests, as proposed by Stedman in 2004.

Why It Matters

1. Novel procedure provides easy detection of mutations.
2. New hypothesis may help bring an answer to the long debated question: how did the human skull shape evolve?
3. Further annotation of MYH16 in the database for other organisms may provide ease of understanding for further studies and eliminate the need for many drastic measures taken here.

Future Directions

1. Test the new method of mutation detection for accuracy against conventional methods.
2. Stain and examine MYH16 and mutated MYH16 in muscle fibers for size and calcium uptake.