The Naked Mole-Rat Epigenome: Unraveling the Secret for Exceptional Longevity

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http://gilatzmon.wix.com/aging-and-longevity
Introduction

Aging
Aging is generally accompanied by age-associated chronic diseases such as Alzheimer’s disease\(^1\), type 2 diabetes\(^2\), cardiovascular disease (CVD)\(^3\), and many forms of cancer\(^4\)-\(^6\) affecting both the quality of life and lifespan.

Longevity
A term that can be defined as an individual’s ability to reach longer lifespan under ideal and proximal conditions\(^7\).

The Naked Mole-Rat
- Small rodent (~28 grams)
- Subterranean lifestyle
- Eusocial colonies
- Extraordinary longevity up to 28 years (8-10 times longer than any other rodent).
- Anti-aging mechanisms\(^8\)-\(^10\):
  - Cancer free good health in stressful environment
  - Unchanged physiological functions
  - Sustained reproductive capacity
  - Negligible senescence

Epigenetics
Epigenetic changes refer to gene expression alteration that arise from chromosomal changes without DNA sequence modification.

The mother’s diet during pregnancy affect the fur color of the litters\(^11\)

- DNA methylation: addition of a methyl group to a cytosine (CpG).

- Methylation changes at specific gene regions have been shown to be associated with age associated diseases\(^12\)-\(^16\).

Hypotheses
- Epigenetic changes associated with aging in NMRs could serve as markers for healthy lifespan.
- Epigenetic changes may represent one of the central mechanisms of gene regulation by which many age-related diseases are buffered.
- Older NMRs will exhibit differential epigenetic changes at sites distinct from younger NMRs.

The Naked Mole-Rat is an excellent candidate model to examine mechanisms leading to “successful” aging and longevity.
Methods

- NMRs Colony at UTHSCSA is the largest colony in the USA and contains the longest-lived of these animals, age reaching older than 32 years

![Image of NMRs]

- DNA extraction from blood samples

HELP-tag assay:
- Two lines from each DNA sample
  - Each is treated with a different restriction enzyme
  - HpaII - methylation sensitive
  - MspI - methylation insensitive
- Building two cDNA libraries
- Next generation sequencing (NGS)
- Sequence Data
- Methylation score (linear scaling of angle value) between 0 and 100 (% of unmethylation at that locus) and confidence score.

![Diagram of HELP-tag assay]

- Alignments and calculation of the methylation fraction

![Diagram of methylation calculation]
Preliminary Results

**Table 1. Genome size and CpG quantity**

<table>
<thead>
<tr>
<th></th>
<th>Genome length (bp)</th>
<th>CG found</th>
<th>% of CG</th>
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<tr>
<td>Human</td>
<td>3095694110</td>
<td>28104642</td>
<td>0.91</td>
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<tr>
<td>NMR</td>
<td>2015372038</td>
<td>13192873</td>
<td>0.65</td>
</tr>
</tbody>
</table>

**Figure 2.** Unsupervised clustering of HELPtag data from blood cells. The heat map shows more methylated loci in red, less methylated in yellow. Distinct patterns are obvious between 2YO to the 3YO groups.

**Figure 3.** Unsupervised clustering of HELPtag data (3400 loci - horizontal lines) from blood cells shows differences in methylation between 4 NMRs. The heat map shows more methylated loci in red, less methylated in yellow. Distinct patterns are obvious between 2YO (*set as reference) to the 7YO animal.

**Figure 4.** Animals' average and SE percentage of methylation (3400 loci) demonstrates increase methylation with age.
Conclusions

- Profiling the epigenome of NMRs, especially methylation patterns, is feasible.
- Subset of loci that characterizes these groups’ epigenetic aging features can provide insights into their exceptional healthy life.
- These epigenetic elements can be translated to human and will provide a candidate loci for manipulation either environmentally or by drug design.
- Similar to humans, NMRs demonstrate differential epigenetic profiling within ages. One year difference in NMR life can result with traceable epigenetic changes.

Future directions:

- Building the epigenomic aging profile for NMR
- EWAS
- Testing for candidate loci in human tissue/cell culture in different environments
- Translation of the results from in vitro to Human

Thank You

References