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Mitochondria-Targeted Antioxidants in Aging related functional changes in the heart and aorta: MitoTEMPO improves aged-cardiovascular performance

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Aging is accumulation of numerous modifications at different levels of the cardiovascular system, resulting in adverse remodelling of the heart and blood vessels.

Age-associated cardiac changes

- Impaired contractility
  - Decreased reserve
  - Norepinephrine dysregulation

- Abnormal rhythmicity
  - Increase in arrhythmia
  - Atrial fibrillation

- Vascular changes
  - Dilation of large arteries
  - Intimal media thickening
  - Increased stiffness
  - Endothelial dysfunction

- Vascular-ventricular mismatching
  - Decreased LV elastance
  - Diminished cardiac reserve

- Diastolic dysfunction
  - Decreased early diastolic filling
  - Increased late diastolic filling
  - Impaired ability of LV to relax

- LV hypertrophy
  - Increased wall thickness
  - Cardiomyocyte hypertrophy
  - Heart failure
Aging and Oxidative Stress

• In 1972 Denham Darman suggested that free radicals cause damage of mitochondria a key determinant of aging processes.

• Unbalanced ROS leading to cellular dysfunction

• Mitochondria the main source of ROS production

*Therefore maintain mitochondrial function is good strategy to protect heart during aging.*
Mitochondrial Targeted Antioxidants

• Decreasing mitochondrial ROS prevents myocardial dysfunction

• Low molecular weight antioxidants (α-Tocopherol, N-acetylcycteine) decreased mitochondrial damage *in vitro*, but their effects *in vivo* is limited

• Antioxidants can be targeted to mitochondria by several methods
  ➢ Hydrophobicity and positive charge
  ➢ Binding with high affinity to intramitochondrial structure
  ➢ Metabolic conversions by specific mitochondrial enzymes

MitoTEMPO conjugated with lipophylic cation TPMP and accumulates 100-500 fold in mitochondria
**General findings of experimental animals (6- vs. 24- month)**

<table>
<thead>
<tr>
<th>Groups</th>
<th>Body Weight (g)</th>
<th>Heart Weight/Body Weight</th>
<th>Systolic pressure (mmHg)</th>
<th>Diastolic pressure (mmHg)</th>
<th>TAS (mM Trolox)</th>
<th>TOS (µM H₂O₂)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adult-rats</td>
<td>330±11</td>
<td>0.43±0.02</td>
<td>119±2</td>
<td>75±0.6</td>
<td>1.05±0.11</td>
<td>0.25±0.06</td>
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<tr>
<td>(n=30)</td>
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<tr>
<td>Aged-rats</td>
<td>380±8.3*</td>
<td>0.52±0.02*</td>
<td>143±4*</td>
<td>83±1.1*</td>
<td>0.37±0.09*</td>
<td>0.77±0.22*</td>
</tr>
<tr>
<td>(n=35)</td>
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</table>

**ECG intervals (s)**

- **P-R**
  - Adult: [Graph]
  - Aged: [Graph]

- **R-R**
  - Adult: [Graph] *
  - Aged: [Graph] *

- **QT**
  - Adult: [Graph] *
  - Aged: [Graph] *

**Heart Rate (beat/min)**

- Adult: [Graph] *
- Aged: [Graph] *
Mitotempo treatment reduces ROS level in senescent cardiomyocytes

mitoTempo: 1 µM- 1-Hour
Mitotempo restores cytosolic Na\(^{+}\), Ca\(^{2+}\) and Zn\(^{2+}\) levels in senescent cardiomyocytes

![Graphs showing the effects of Mitotempo on Na\(^{+}\), Ca\(^{2+}\) and Zn\(^{2+}\) levels in young, aged, and A+MitoT cardiomyocytes.](image)

- **Amplitude**
  - Young: 0.20 ± 0.05
  - Aged: 0.25 ± 0.06
  - A+MitoT: 0.30 ± 0.04

- **Basal**
  - Young: 0.05 ± 0.01
  - Aged: 0.10 ± 0.03
  - A+MitoT: 0.08 ± 0.02

- **Basal [Na\(^{+}\)]\(_i\)**
  - Young: 0.60 ± 0.10
  - Aged: 0.80 ± 0.15
  - A+MitoT: 0.70 ± 0.12

- **[Zn\(^{2+}\)]\(_i\)**
  - Young: 1.2 ± 0.3
  - Aged: 2.0 ± 0.5
  - A+MitoT: 1.5 ± 0.4

**CARDIAC ACTION POTENTIAL**

- **Inward Current**
- **Outward Current**
- **Rest**
- **Rising Phase**
- **Plateau**
- **Repolarization**
- **Ca\(^{2+}\)**
- **K\(^{+}\)**
- **Na\(^{+}\)**
Mitotempo improves Left Ventricular function in senescent cardiomyocytes

Langendorff perfusion

- LVEDP (%)
- Time to Peak (ms)
- Time to half relaxation (ms)
Mitotempo improves contraction-relaxation function of aortic rings in senescent cardiomyocytes

\[ \text{Phe} (\mu M) \]
\[ 0.1 \ 0.5 \ 1 \ 5 \ 10 \ 50 \ 100 \]
\[ \text{Contraction} (\%) \]
\[ -7.0 \ -6.5 \ -6.0 \ -5.5 \ -5.0 \ -4.5 \ -4.0 \]
\[ \text{Log}[\text{Phe}] \]
\[ \text{Adult} \quad \text{Aged} \quad \text{Adult+MitoT} \quad \text{Aged+MitoT} \]

\[ \text{Ach} (\mu M) \]
\[ 0.1 \ 0.5 \ 1 \ 5 \ 10 \ 50 \ 100 \]
\[ \text{Relaxation} (\%) \]
\[ -7.0 \ -6.5 \ -6.0 \ -5.5 \ -5.0 \ -4.5 \ -4.0 \]
\[ \text{Log}[\text{Ach}] \]
TURAN’s Lab