SPACEFLIGHT AND THE MOUSE EYE: RESULTS FROM EXPERIMENTS ON SHUTTLE MISSIONS STS-133 AND STS-135

Susana B. Zanello, Division of Space Life Sciences, USRA
Corey A. Theriot, Department of Preventive Medicine and Community Health, UTMB
Claudia Prospero Ponce, Department of Pathology and Genomic Medicine, TMH
Patricia Chevez-Barrios, Department of Pathology and Genomic Medicine, TMH
Problem

Vision alterations associated with globe flattening, choroidal folds and papilledema, shown in some crew members returning from long duration missions (Mader, et al. Ophthalmology 2011)

Hypothesis

Ocular neuroanatomical changes observed in the VIIP syndrome are accompanied by retinal changes at the molecular and cellular level that may affect retinal health and physiology

Previous work

Scarce evidence from animal flight experiments, showing cell loss in retina of rat neonates aboard STS-72 (Tombran-Tink & Barnstable, 2005) and cell disruption in retina of rats aboard Cosmos 782 and 936 (Philpott, et al. 1978, 1980)
Experimental design

• Tissue sharing

• STS-133  Balb/cJ mice, 10-12 week old (albino)
  Conditions: vivarium (VIV), AEM, flight (FLT)
  Duration of flight: ~13 days
  Tissue collections: R+1, R+5, R+7

• STS-135  C57BL/6 mice, 9-11 weeks old
  Conditions: AEM, flight
  Duration of flight: ~13 days
  Tissue collections: R+1

Objective

Investigate evidence of ocular (retinal) changes associated with spaceflight:
✓ histological markers of cellular death and damage
✓ molecular markers of oxidative stress
✓ gene expression markers of stress
STS-133 Histology

Corneal epithelium

AEM R+7-Basal edema and acanthosis

FLT R+1 - Caspase 3 positive

Retina

FLT R+1 Caspase-3 positive
**STS-133 Histology**

**GFAP**: glial fibrillary acidic protein (glial activation)

**β-amyloid**: neuronal injury

*Post-laminar β-amyloid + region*
STS-133 Oxidative stress DNA damage (8OHdG)
STS-133 Gene Expression

Oxidative stress

Cellular stress

Cellular death
STS-133 Gene Expression

- **Oxidative stress**
- **Cellular stress**
- **Cellular death**

Graphs showing normalized expression values over days post return, highlighting the impact of stress responses.
STS-135 Histology

#16 AEM

#52 FLT

....% more caspase-3 + RGC in FLT vs AEM (n=3)
STS-135 Histology

AEM

FLT

8OHdG
STS-135 Microarray

- Microarray processing and analysis performed at the UTMB Genomics Core Laboratory (n=3)
- Ingenuity systems iReport generated
- Affymetrix mouse expression array: 40,000 genes
- Differentially expressed genes: 139
## STS-135 Pathway analysis

<table>
<thead>
<tr>
<th>PATHWAYS</th>
<th>PROCESSES</th>
<th>DISEASES</th>
</tr>
</thead>
<tbody>
<tr>
<td>ER stress</td>
<td>RNA processing (mRNA splicing)</td>
<td>Cancer</td>
</tr>
<tr>
<td>Pyrimidine metabolism</td>
<td>Cell death of sensory neurons and RGC</td>
<td>Neurodegeneration of nerves and nervous tissue</td>
</tr>
<tr>
<td>Cytokine production and signaling (IL-1, IL-6, IL17)</td>
<td>Apoptosis of microglia and neuronal cells</td>
<td>Degeneration of optic nerve</td>
</tr>
<tr>
<td>Sphingosine-1-P signaling</td>
<td>Stabilization and assembly of desmosomes</td>
<td>Reactivation of herpes virus</td>
</tr>
<tr>
<td>Axonal guidance and actin cytoskeleton</td>
<td>Axon branching</td>
<td></td>
</tr>
<tr>
<td>Molecular mechanisms of cancer</td>
<td>ER stress response</td>
<td></td>
</tr>
</tbody>
</table>
STS-133/135: Melanopsin

Light input + non-visual photoreceptor(s) → Molecular clock (SCN) → Output: circadian effectors, gene expression, physiological rhythms, behavior

Reppert & Weaver, 2002, Nature 418, 935-941
STS-133/135: Melanopsin
Summary

These preliminary results suggest that:

• Oxidative stress and neuronal loss occur in the retina of mice exposed to spaceflight
• Damage is preferentially localized in RGC
• Oxidative and cellular stress is reversible upon return to Earth
• Damage is also evidenced by glial activation and neuronal/axonal injury
• ER stress and neuronal/glial cell death pathways are implicated in neuronal cell loss
• Susceptibility to cellular stress may affect the response and resistance to the effects of spaceflight in the retina and thus, the susceptibility to further damage (degeneration)
• Melanopsin expression and/or survival of ipRGC may be compromised under the stress of spaceflight conditions
Acknowledgements

- Rich Boyle
- ARC/KSC tissue sharing
- HRP