Muscle mitochondrial function and contemporary anti-retroviral therapy

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Connection between mitochondria and NRTIs in skeletal muscle

Depletion of muscle mitochondrial DNA in AIDS patients with zidovudine-induced myopathy

Enrica Arnaudo, Marinos Dalakas, Sara Shanske, Carlos T. Moraes, Salvatore DiMauro, Eric A. Schon

Mitochondrial aging is accelerated by anti-retroviral therapy through the clonal expansion of mtDNA mutations

Brendan A I Payne, Ian J Wilson, Charlotte A Hateley, Rita Horvath, Mauro Santibanez-Koref, David C Samuels, D Ashley Price, Patrick F Chinnery
Mitochondrial dysfunction in skeletal muscle

Mitochondrial dysfunction

Myofiber defects

Physiological decline

Payne et al., 2011
Aims

• To elucidate whether contemporary NRTIs cause mitochondrial dysfunction.
• To objectively quantify mitochondrial dysfunction in skeletal muscle in the contemporary setting.
Methods

Cohort – 37 PLWH:

• 13 NRTI naïve;
• 10 exposed to contemporary NRTIs only;
• 14 currently on contemporary NRTIs but previous exposure to older NRTIs

1. HIV related clinical characteristics assessed
2. Multiplex immunofluorescence for mitochondrial respiratory chain complexes I and IV on frozen 10µm tibialis anterior sections
3. Molecular analysis of mtDNA defects within single fibers

<table>
<thead>
<tr>
<th>Contemporary NRTIs</th>
<th>Historical NRTIs</th>
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<tr>
<td>Tenofovir (TDF)</td>
<td>Azidothymidine (AZT)</td>
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<tr>
<td>Abacavir (ABC)</td>
<td>Zalcitabine (ddC)</td>
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<td>Lamivudine (3TC)</td>
<td>Didanosine (ddI)</td>
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<tr>
<td>Emtricitabine (FTC)</td>
<td>Stavudine (d4T)</td>
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Multiplex immunofluorescence

Laminin – myofiber boundary marker
Porin – mitochondrial mass
NDUFB8 – complex I
MTCO1 – complex IV
NRTI treated PLWH have higher CI and CIV deficiency
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Complex I deficiency and clinical factors

Univariate analysis

Multivariate model showed that historical and contemporary NRTI exposure, but not age, were both predictive of CI deficiency
mtDNA analysis in single fibers

Multiplex qPCR assay for:
- \textit{MT-ND1} (minor arc)
- \textit{MT-ND4} (major arc)

- Deletions quantified
- Copy number quantified

Adapted from Taylor & Turnbull, 2005
OXPHOS deficient fibers contain mtDNA deletions
Higher frequency of major arc deletions in historical NRTI group
Conclusions

• As expected, PLWH with previous exposure to older NRTIs had a higher level of complex I and IV deficiency than ART naïve PLWH
  • Using an objective and quantitative method for assessing mitochondrial dysfunction

• Surprisingly, patients exposed only to contemporary NRTIs had a higher proportion of complex I deficient fibers than ART naïve PLWH.
  • Suggesting contemporary ART may not be as mitochondrially clean as thought

• Complex I defects predominate
  • Potential therapeutic interest?

• Oxidative phosphorylation deficient fibers predominantly contain mtDNA deletions.
Future work

• Further investigation of molecular mechanisms behind mitochondrial defects in contemporary NRTIs.

• Multivariate analysis of mitochondrial dysfunction and physical function/frailty
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