Mitochondrial dysfunction and oxidative stress induced by palmitate in human muscle cell

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Introduction

- Lipotoxicity and skeletal muscle dysfunction
  - Trigger by oxidative stress?
    - Produced by mitochondria?
    - Antioxidant response set up?

Methods

FFA: Palmitate treatment (PA-300μM-24h) on human myoblasts and myotubes (after 4 days of diff.).

Cells were treated with PA complexed to BSA and compared to cells treated with BSA only (control condition).

Results

1 Significant accumulation of intramyocellular lipid droplets

Neutral triacylglycerols and lipid inclusions were labeled by Oil red O (red) staining and cells counterstained with Giemsa. Lipid Droplets were present in less myoblasts (20 ±8%) than myotubes (86 ±14%). Any lipid droplet was found in control cells. Bar graphs represent mean ± SEM.

No impact of PA on proliferation and differentiation levels

Total proliferative (a) and specific S Phase (b) myoblasts were count and related to the total number of cells. Area coverage of myoblasts (c) was determined on the total cell culture area, fusion index (d) were calculated as the ratio between myoblast nuclei and total nuclei; myoblast width (e) were measured to detect morphologic changes. Our analyses showed no significant difference with or without palemitate treatment. Bar graphs represented mean ± SEM.

2 Mitochondrial dysfunction in myoblasts and myotubes

(a) mitochondrial superoxide overproduction
(b) Mitochondrial activity
(c) Mitochondrial swelling

(a) Production of mitochondrial superoxide was detected using MitoSOX™ Red Mitochondrial Superoxide. (b) Vybrant® MTT Cell Assay was used to measured complex II activity. data suggest a decrease of mitochondrial activity. (c) Myoblast mitochondria treated with PA. (d) Control myoblast mitochondria. Inner membrane was disrupted by a swelling of mitochondrion and a increased of lamellar body. LB: Lamellar body DM: Dreal mitochondria INSMD: Inner membrane disruption.

PA treatment induces an important alteration of mitochondrial activity along with increased production of mitochondrial O₂⁻ and mitochondrial swelling.

3 Nrf2 expression & Antioxidant balance

Nrf2 transcription was observed after PA treatment along with increased antioxidant response in PA-treated myoblasts and myotubes.

Conclusion

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