Lipotoxicity and oxidative stress induced by palmitate in human muscle cells

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Introduction

FFA

↗ lipotocxicity and skeletal muscle dysfunction

→ Trigger by oxidative stress?

- Produced by mitochondria?
- Produced by NADPH oxidase family?

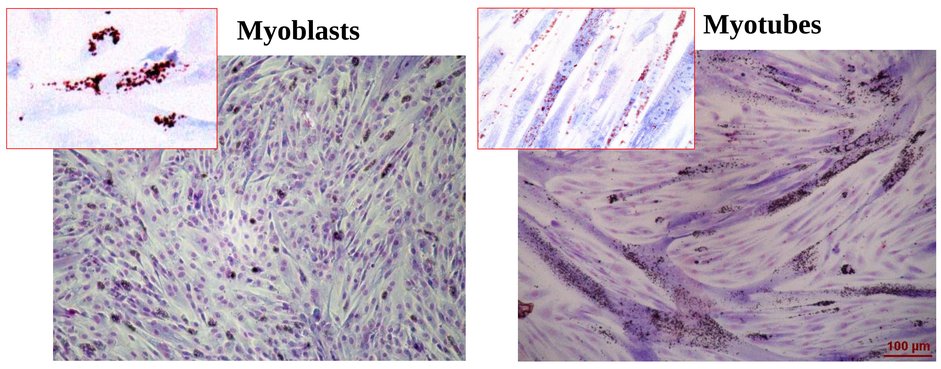
Methods

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).

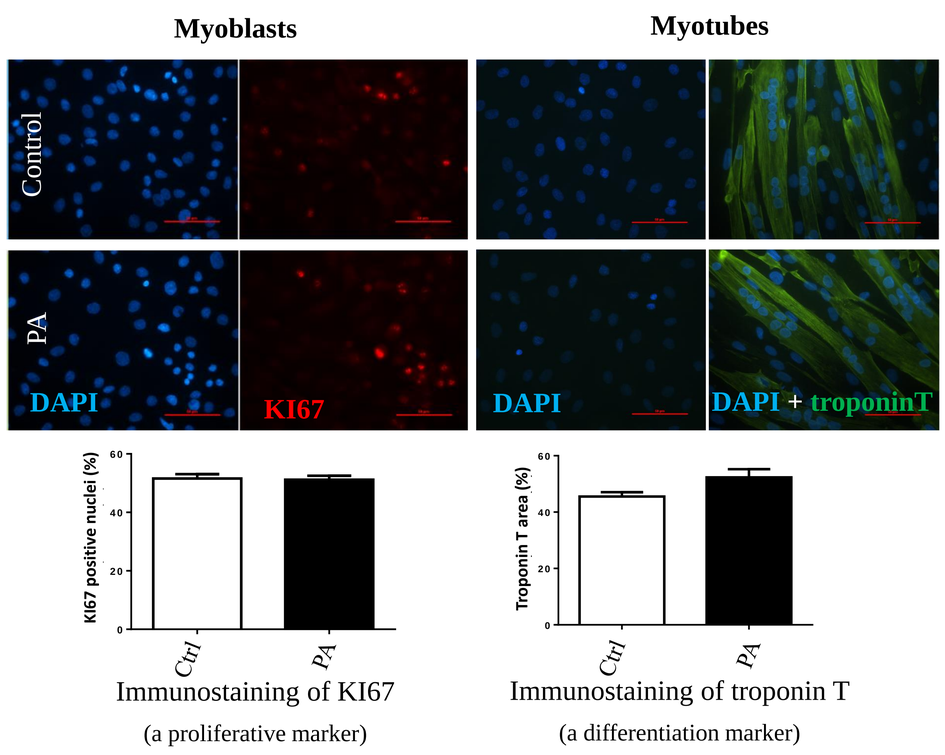
Results

1 Significant accumulation of intramyocellular lipid droplets



Giemsa and Oil red O stainings after PA treatment

No impact of PA on proliferation and differentiation

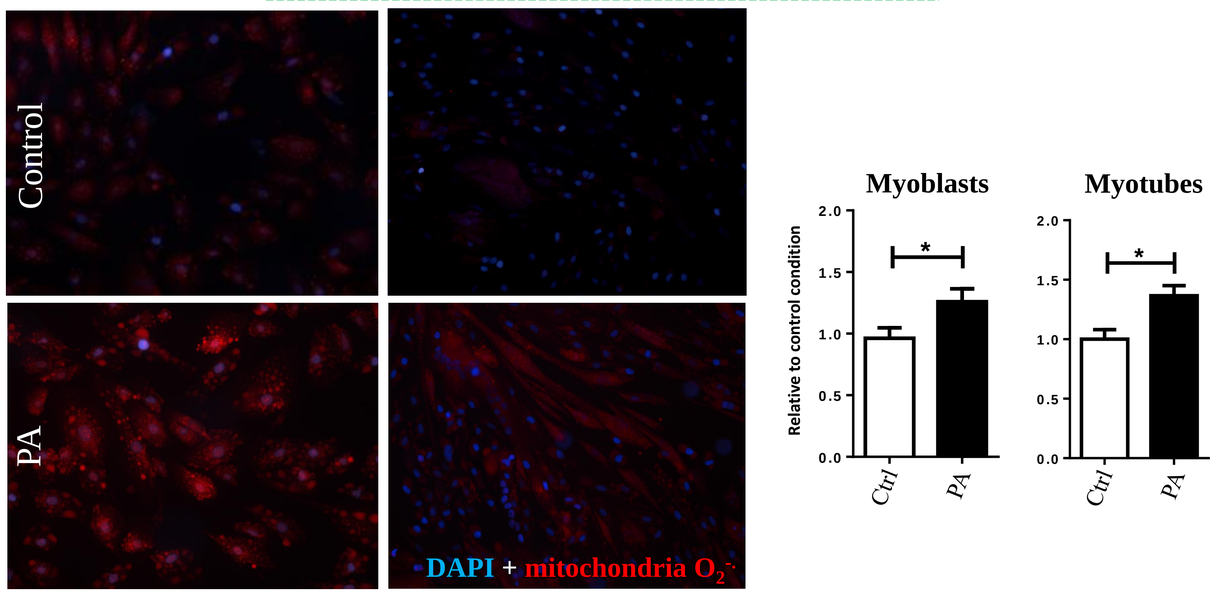


Immunostaining of KI67 (a proliferative marker)

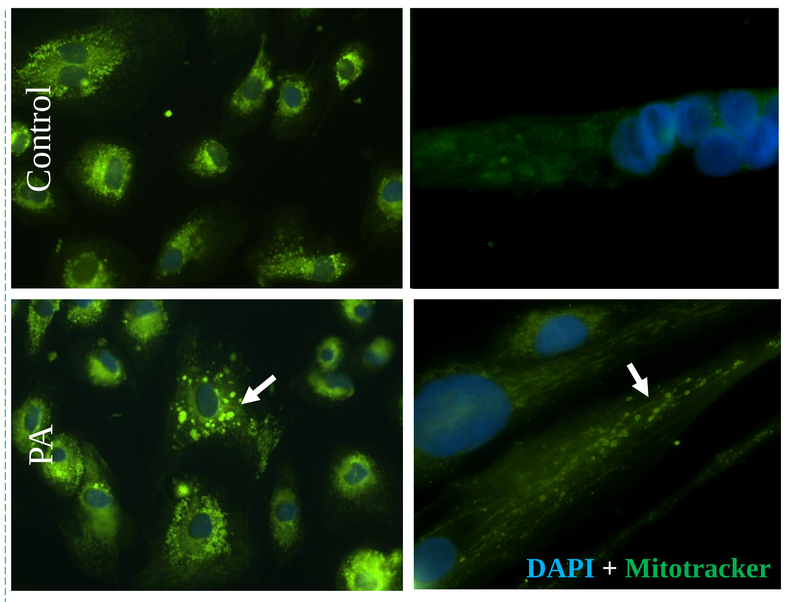
Immunostaining of troponin T (a differentiation marker)

2 Alteration of mitochondria in myoblasts and myotubes

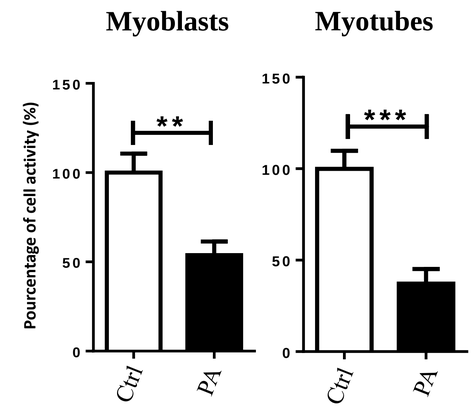
a) Superoxide production by mitochondria



b) Mitochondrial swelling



c) MTT assay



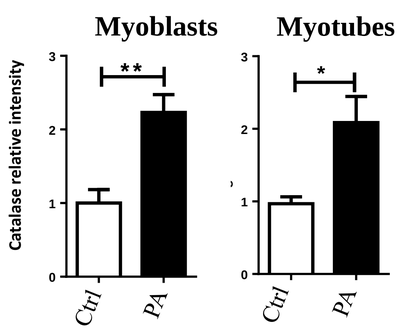
(a) Increase of superoxide production highlighted by MitoSOX™ Red Mitochondrial Superoxide Indicato (b) swelling of mitochondria (c) Decrease of mitochondria activity highlighted by a Vybrant® MTT Cell Assay

=> PA induced an important alteration of mitochondria activity along with increased mitochondrial $O_2^{\cdot-}$ and mitochondrial swelling.

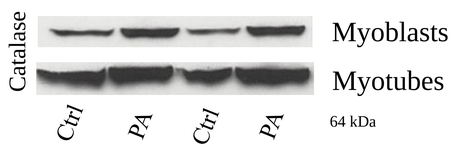
3 NOX expressions & Antioxidant balance

	MYOBLASTES		MYOTUBES	
	BSA	PA	BSA	PA
NOX2 (N=5)	1,00 ± 0,00	0,92 ± 0,11	1,00 ± 0,00	0,62 ± 0,23
NOX4 (N=5)	1,00 ± 0,00	1,46 ± 0,31	1,00 ± 0,00	1,32 ± 0,29
P47^{PHOX} (N=3)	1,00 ± 0,00	1,23 ± 0,12	1,00 ± 0,00	1,16 ± 0,15
P22^{PHOX} (N=2)	1,00 ± 0,00	0,88 ± 0,07	1,00 ± 0,00	1,20 ± 0,18

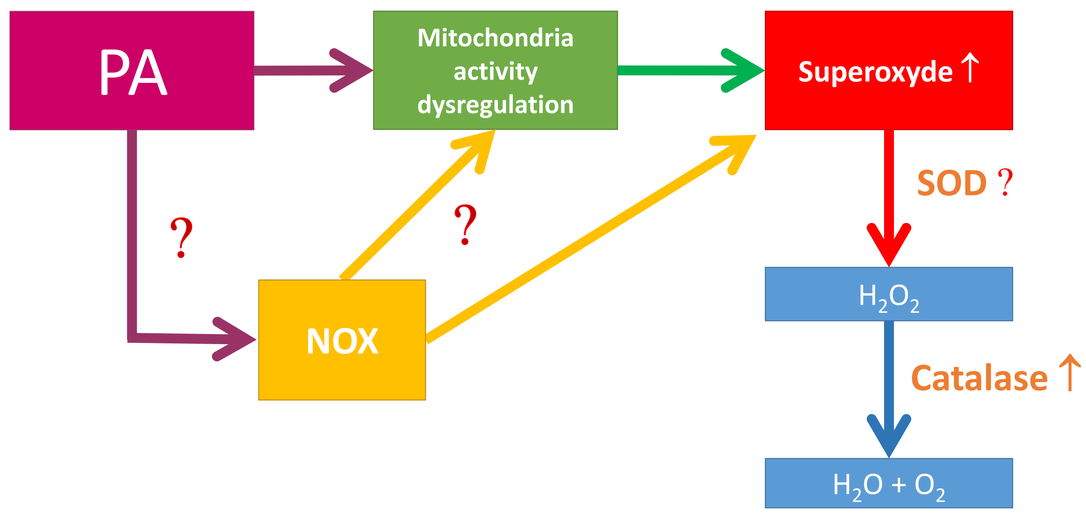
No change of *NOX* mRNA expression after PA treatment in myoblasts and myotubes was observed.



Catalase protein expression was increased by PA in myoblasts and myotubes.



Conclusion



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