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Data Article

Effect of mitochondrial fission inhibition on C2C12 differentiation

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ABSTRACT

The differentiation of skeletal muscle is commonly examined in cell culture using the C2C12 line of mouse skeletal myoblasts. This process shares many similarities with that which occurs during embryonic development, such as the transient activation of caspases. Here, we examined the effect of inhibiting mitochondrial fission, using mdivi-1, on the ability of C2C12 cells to terminally differentiate. This was performed using immunofluorescent identification of cell morphology and myosin expression, as well as immunoblotting for markers of muscle differentiation. Furthermore, the effect of mdivi-1 administration on activation of caspase-2 and -3 was assessed using spectrofluorometric measurement of specific enzyme activity.

Specification Table

Subject area	*Biology*
More specific subject area	*Skeletal muscle differentiation, caspases, mitochondrial fission*
Type of data	*Graphs and Figures*
How data was acquired	*Fluorescent microscopy, spectrofluorometry, immunoblotting*
Data format	*Analyzed*

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Experimental factors	*C2C12 cells were differentiated with the inhibitor of mitochondrial fission, mdivi-1*
Experimental features	*Cells were differentiated while being exposed to different concentrations of mdivi-1 and collected at various time points during the differentiation process. They were then prepared for spectrofluorometric measurement of caspase activity or were assessed for the markers of muscle differentiation, myosin and myogenin, using immunoblotting. Separate cells were immunostained for myosin and imaged using fluorescent microscopy to evaluate changes to cell morphology.*
Data source location	*University of Waterloo, Waterloo, Ontario, Canada*
Data accessibility	*All data are provided with this article*

Value of the data

- The data demonstrate that myogenic differentiation is prevented by administering a chemical inhibitor of mitochondrial fission.
- Although mitochondrial fission has complex roles regarding the regulation of apoptotic signaling, its inhibition led to significant elevations in caspase activity in this context.
- These data provide evidence that proper mitochondrial fission is important for the dramatic changes that accompany cellular adaptations, and support the execution of further studies in this regard.

1. Data

Here, we present data regarding the effect of mitochondrial fission inhibition on skeletal muscle differentiation. Although similar experiments have been published by others [1,2], the data presented here support and add to the conclusions made by these researchers. Of note, the relatively higher

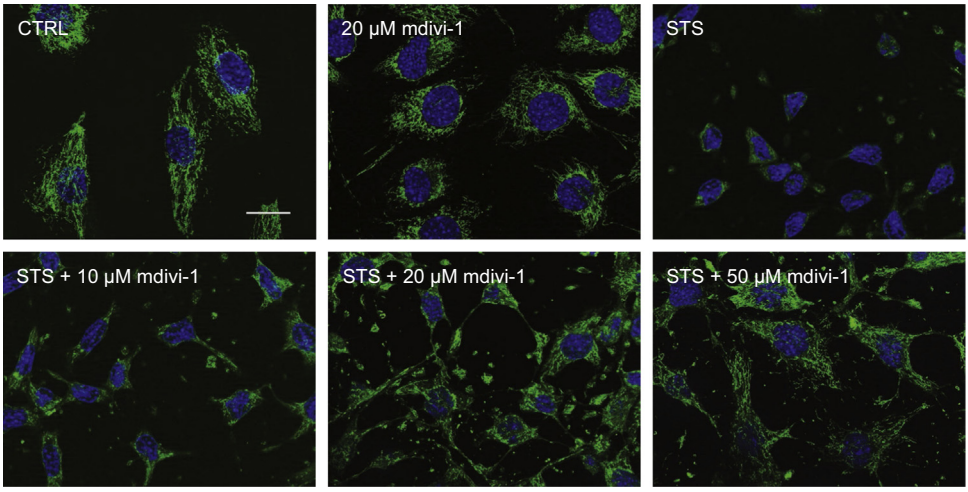

Fig. 1. Determination of working mdivi-1 concentrations. Increasing concentrations of mdivi-1 progressively inhibited apoptosis-associated changes in cell morphology induced by 2 h of 2 μM STS. Mitochondria were visualized using MitoTracker (green) and nuclei with DAPI (blue). Scale bar represents 20 μm.