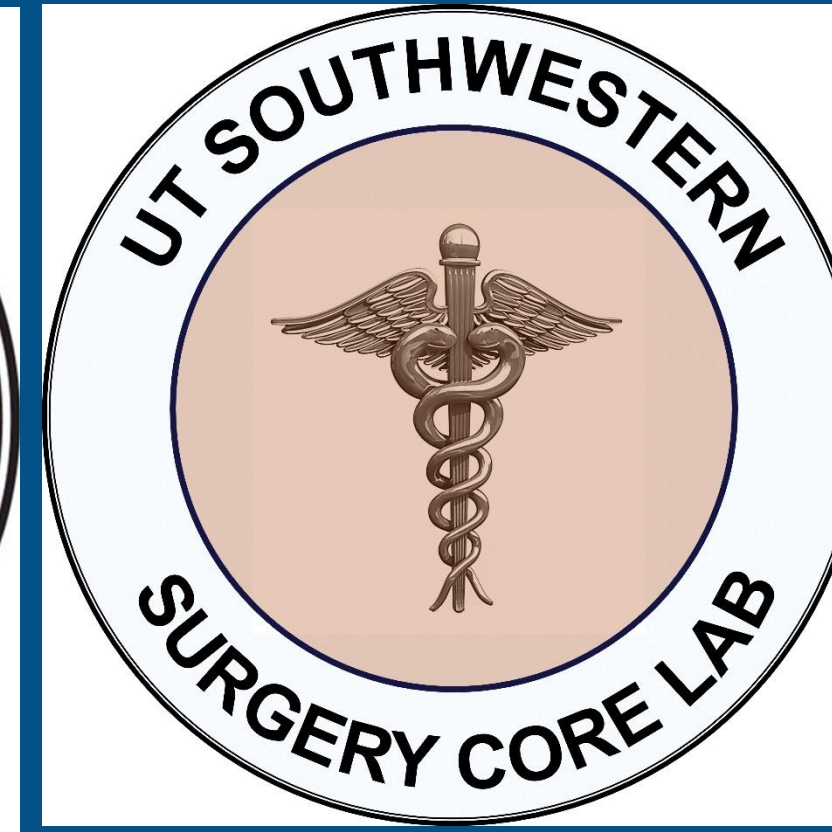


Myokine musclin expression is elevated in rats after burn

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BACKGROUND

Severe burns lead to a hypercatabolic state with significant endocrine disruption. Previous studies show detrimental effects of insulin resistance and hyperglycemia associated with muscle loss due to burns. In addition, studies show that epinephrine and insulin are increased after burns. The novel myokine musclin has been found to regulate glucose and is positively correlated with exercise. Musclin mRNA increases with insulin treatment, but decreases with epinephrine stimulation in myoblasts. Thus, we propose that musclin expression is controlled by a complex endocrine response after burns.

OBJECTIVE

1. Investigate systemic musclin levels in response to severe burn and musclin expression in myoblasts stimulated with burn serum
2. Investigate the expression of musclin in response to ER stress and the role of musclin in cell death

METHODS

In vivo:

Exp 1: Musclin levels in burn serum

Thirty-one Sprague-Dawley rats received 40% total body surface area (TBSA) burn. Rat serum was collected from 6 hours to 14 days after burn. Musclin levels were measured by ELISA.

In vitro

Exp 2: Musclin expression in myoblasts in response to burn serum

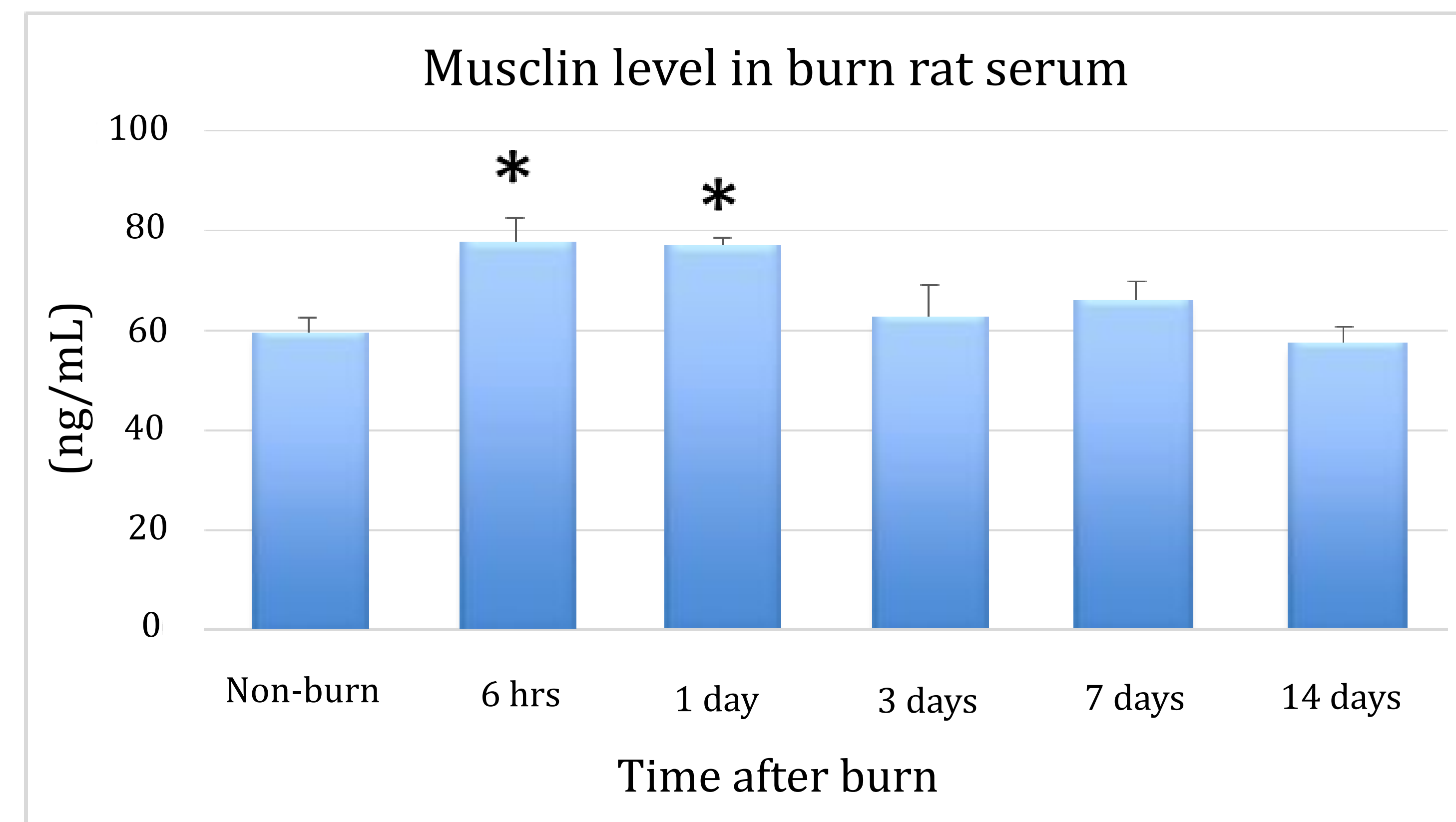
Mouse C2C12 myoblasts were stimulated with non-burn serum, 6-hour/72-hour/and 14-day post burn serum. Cell total RNA were extracted by RT qPCR analysis.

Exp 3: Musclin expression in myoblasts with ER stress inducer

Thapsigargin (TG), a non-competitive inhibitor of the sarco/endoplasmic reticulum Ca^{2+} ATPase, was added to C2C12 cells (100 μ M). Musclin expression was measured with western blot.

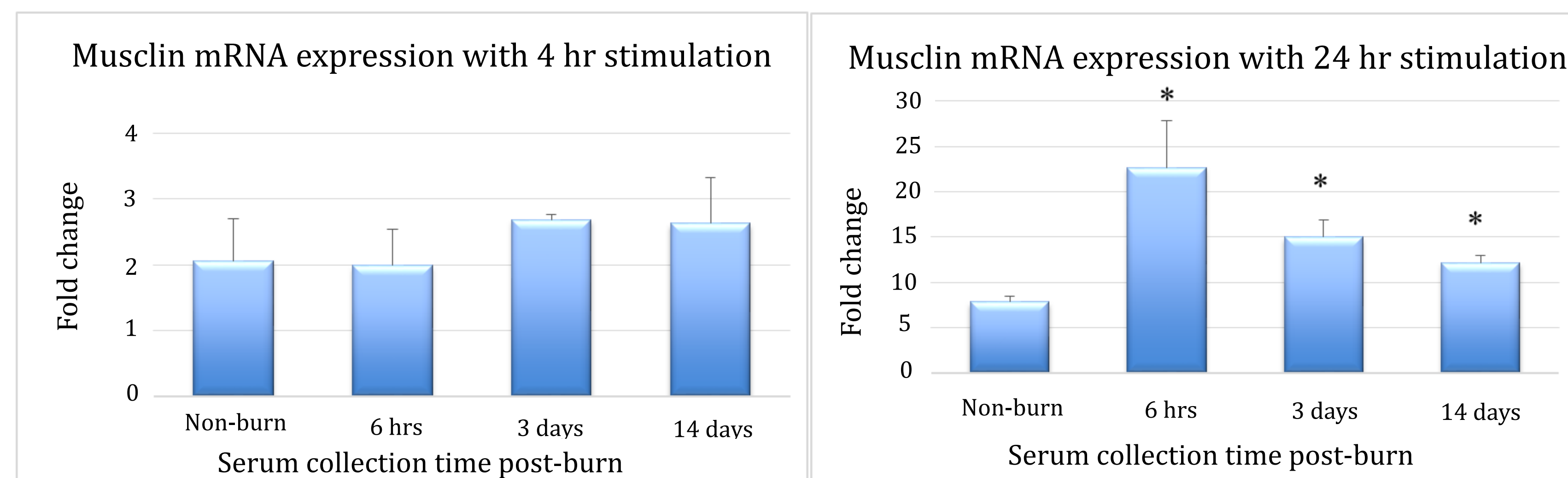
Exp 4: Effect of musclin on burn serum stimulated cell death
C2C12 cells with 10% normal rat serum, 10% 6 hour-post-burn serum and musclin peptides (P1:31-75; P2:80-130) (Phoenix Pharm). Caspase 3 substrate fluorescent intensity (indicating cell death) was measured with Invitrogen commercial kit.

RESULTS #1: MUSCLIN LEVELS IN RAT SERUM AFTER BURN



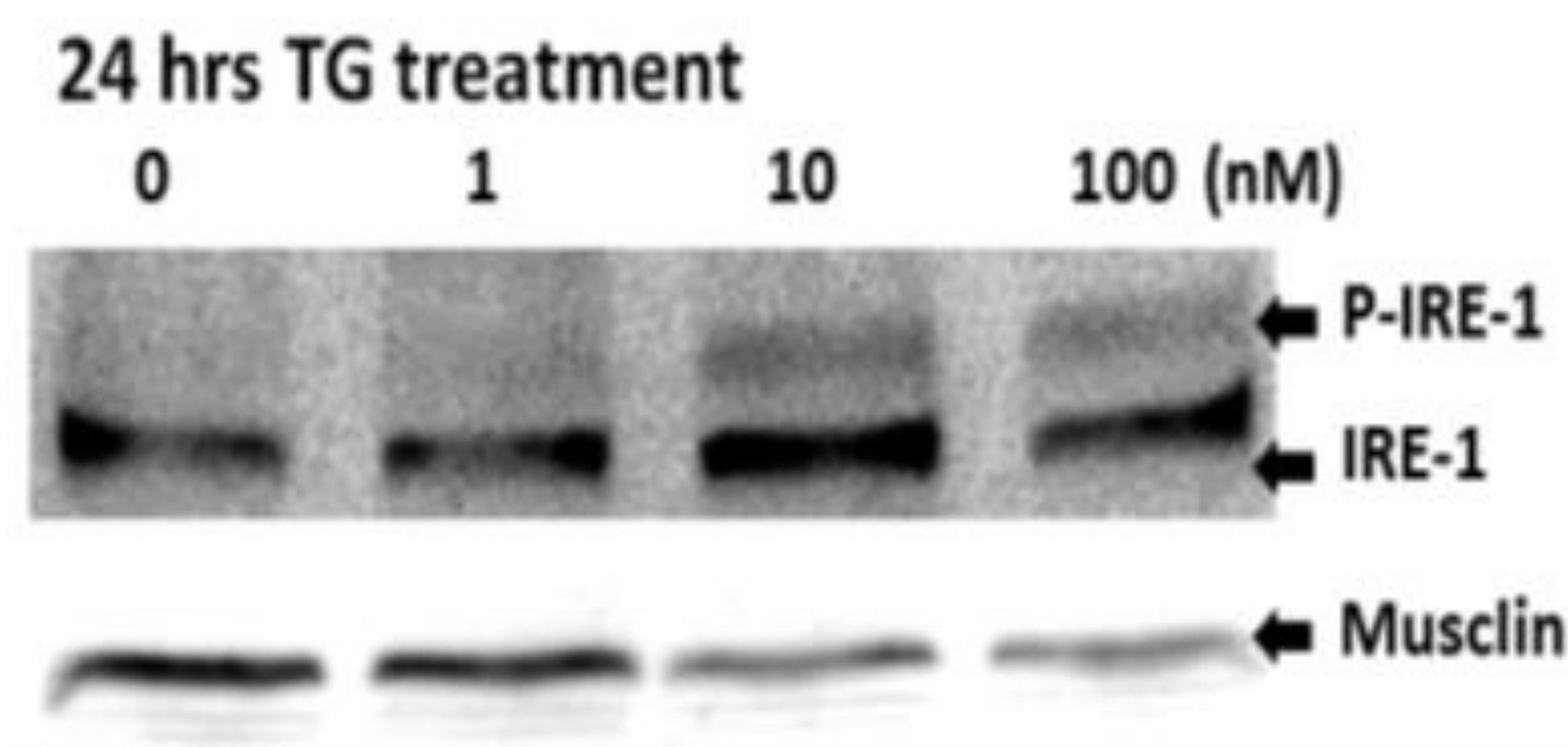
* p<0.05, between non-burn and burn

RESULTS #2: MUSCLIN mRNA EXPRESSION IN C2C12 CELLS STIMULATED WITH RAT BURN SERUM

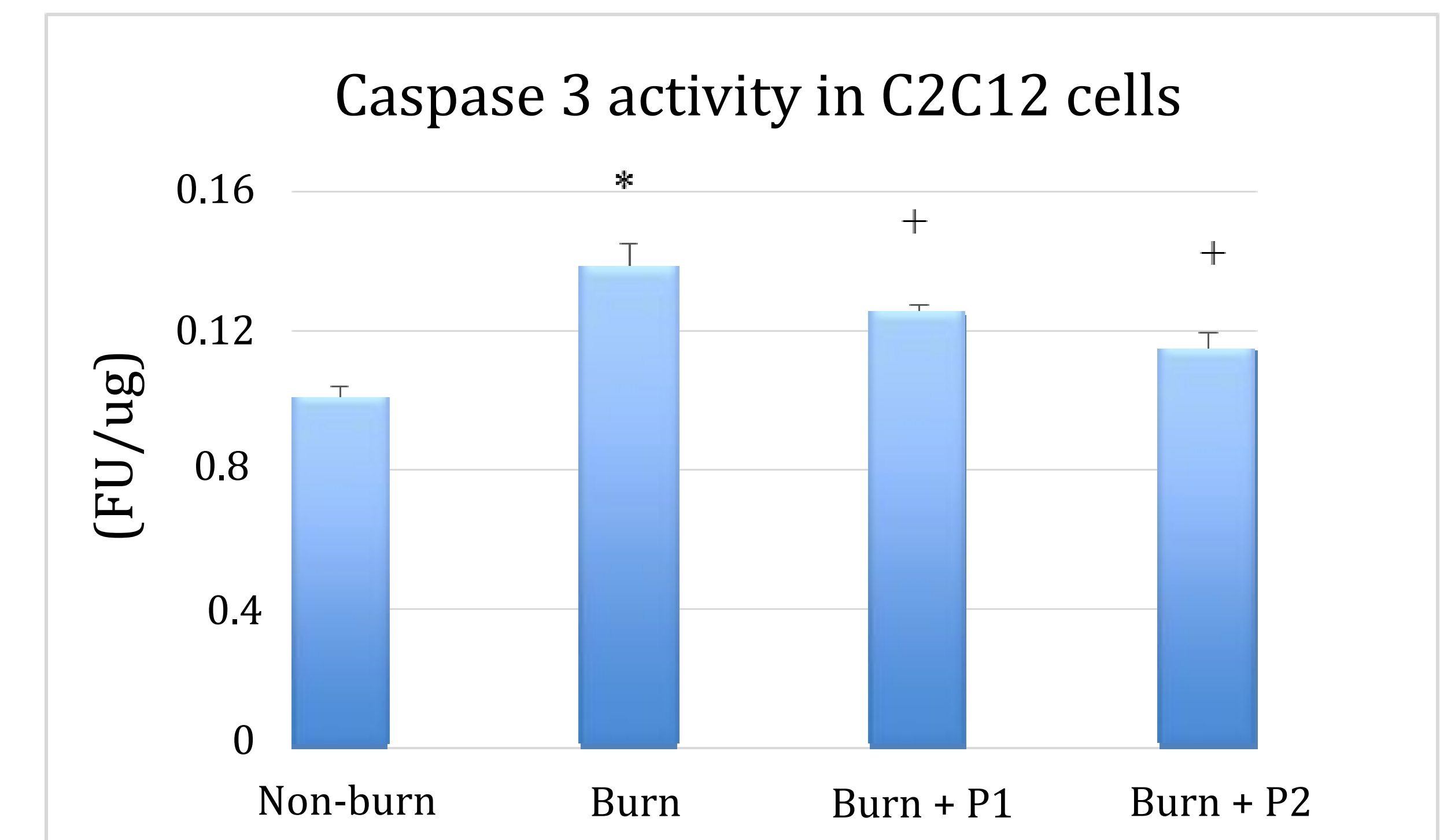


* p<0.05, between non-burn control and post-burn serum at each of the three time points

RESULTS #3: ER STRESS INDUCER INHIBITS MUSCLIN EXPRESSION



RESULTS #4: MUSCLIN PEPTIDES DECREASE CELL DEATH IN BURN SERUM STIMULATED MYOBLAST



* p<0.05, between non-burn and burn
† p<0.05, between burn and burn + peptides

CONCLUSION

1. Musclin levels increase in rat serum after burn systemically. *In vitro* data confirmed muscle cells expressed musclin in response to burn serum stimulation.
2. Musclin is negatively associated with ER stress; exogenous musclin alleviates cell death *in vitro*, suggesting a protective role of musclin.

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