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Abstract

Ischemic injury in skeletal muscle caused by hypoxic (low oxygen) conditions occurs in response to vascular and musculoskeletal traumas, diseases and following reconstructive surgeries. Thus, a thorough understanding of the effect of hypoxia on skeletal myoblasts is warranted to identify potential therapeutic targets. We have determined that treatment with cobalt chloride (to mimic hypoxic conditions) leads to decreased numbers of viable (attached) skeletal myoblast over time and an increase in the percentage of detached myoblasts. To determine the contribution of apoptosis (cell death) to this increase in detached myoblasts, we assessed PARP cleavage, a well-accepted marker for apoptosis. Consistent with cell death via the apoptotic process, PARP cleavage was detected following cobalt chloride treatment. We next assessed the effect of cobalt chloride on the expression of the pro-apoptotic Bcl2 family member PUMA. We determined that after three hours, cobalt chloride treated myoblasts possessed PUMA mRNA levels six times greater than untreated myoblasts. Further, this increase in PUMA mRNA resulted in a three-fold increase in PUMA protein. Future experiments will focus on determining the mechanism whereby cobalt chloride treatment results in increased PUMA levels.

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