Incomplete Caspase Signaling during Hibernation in the Golden-Mantled Ground Squirrel, *Spermophilus lateralis.*

by

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Hibernating golden-mantled ground squirrels, *Spermophilus lateralis,* experience numerous conditions during the winter that are known to be pro-apoptotic in other mammal systems (e.g. ischemia and reperfusion, acidosis, and increased reactive oxygen species). However, hibernators may invoke a protective phenotype to limit widespread cell damage and loss during the hibernation season. Could regulating apoptosis provide protection against the harmful conditions experienced during the hibernation season? To address this, the caspases, a class of cysteine-aspartate proteases crucial for executing apoptosis and inflammation were examined. Caspases participate in a complex and interwoven signaling network known as the caspase cascade. Using western blotting, I examined caspases 1-12 for evidence of apoptotic signaling during hibernation. Apoptotic executioner caspases 3 and 6 and inflammatory caspases 11 and 12 appeared activated during hibernation. Did these seemingly winter-activated caspases display increased activity? Enzymatic assays revealed no indications of dramatically increased caspase activity in hibernating squirrels. To better understand the implications of seeming caspase activation during hibernation, I used a systems-level approach to analyze numerous events downstream of caspase activation. Despite the pro-apoptotic conditions of hibernation and the apparent caspase activations, no evidence of increased downstream caspase activity or widespread apoptosis and inflammation was found during hibernation. Regulation of apoptosis during hibernation occurs at an unexpected locus that does not involve the prevention of caspase activation. Instead, partial activation of the caspase cascade does not result in predictable downstream processing, thus demonstrating regulation of apoptosis during hibernation involves inhibition of caspase activity after seeming activation. These data demonstrate the importance and utility of a systems-level approach in studying complex cellular signaling pathways like apoptosis during hibernation.