Name of Presenter:	Swati Jain
Degree of Presenter:	PhD
Co-Authors and degrees:	Robert Plenter BS; Trevor Nydam MD; Alkesh Jani MD
Division:	Renal Diseases and Hypertension
Presenter's Rank:	Junior Faculty
Research Category:	Basic Science
Title of Abstract:	Apoptosis during Cold Ischemia and Rewarming Involves a Caspase Independent Pathway
Please copy and paste your abstract here: (no more than 300 words):	Background: We have previously shown that hibernating ground squirrels (GS) tolerate cold ischemia (CI) for several days and their kidneys display no renal tubular epithelial cell (RTEC) apoptosis. We have also shown that mouse kidneys subjected to cold ischemia (CI) followed by transplantation (CI+Txp) demonstrate significant RTEC apoptosis that can be partially blocked with a pan caspase inhibitor, Q-VD-OPh. Since Q-VD-OPh only partially blocks apoptosis, we hypothesized that caspase independent apoptosis mediated by AIF and EndoG may be activated during CI+Txp in mice. Methods: GS and mouse RTECs were subjected to in vitro cold storage in saline solution followed by rewarming (CS/REW) in normal media. Donor mice kidneys were subjected to CI followed by kidney transplant or transplanted without CI. Results: Mouse kidney transplant: AIF translocation to the cytosol was detected only in mouse kidneys subjected to CI+Txp, whereas mouse kidneys that were not subjected to CI did not demonstrate cytosolic translocation of AIF. In contrast, EndoG cytosolic translocation was not detected.

In vitro: Mouse RTECs exposed to CS/REW had significantly increased apoptosis versus squirrel RTECs. Furthermore, mouse RTECs subjected to CS/REW had significantly increased AIF translocation from mitochondrial to cytosolic fraction versus squirrel RTECs. EndoG translocation could not be detected. Transfection of mouse RTECs with AIF (370-394) peptide blocks the AIF translocation to nucleus, therefore inhibits apoptosis in mouse RTECs subjected to CS/REW. Moreover, AIF (370-394) peptide and Q-VD-OPh acts synergistically to inhibit apoptosis in mouse RTECs subjected to CS/REW.
Conclusions:
Our data suggests that GS suppress caspase dependent and independent apoptosis. In contrast, in vitro CS/REW and CI+Txp in mice is characterized by both caspase dependent and caspase-independent apoptosis. The latter is mediated by AIF rather than EndoG. Complete blockade of RTEC apoptosis in clinical transplantation will therefore likely require inhibition of both caspase dependent and independent pathways.