**PROAPOPTOTIC PROTEINS BAX AND BAK IN A MYOCARDIUM DAMAGE MODEL MEDIATED THROUGH DIGITALIS INTOXICATION**

**M.C. Ramirez-Ortega1**, I. Cuevas-Escobar1, J.F. Carrillo-Hernandez2,

M.L. Ibarra-Lara1, G. Pastelin-Hernandez1

1Instituto Nacional de Cardiologia "Ignacio Chavez", Mexico D. F., 2Instituto Nacional de Cancerologia, Mexico D. F., Mexico

Bak and Bax proteins form oligomeric complexes that disrupt the mitochondrial membrane permeability. To analyze if these proteins participate in apoptosis induced by ouabain in guinea pig hearts this study was performed.

Methods: We utilized 32 guinea pigs. One untreated control group (n=12), and four groups (n=5/group) treated at different time-intervals (4, 12, 24 and 48 h) with ouabain (327 nmoles/kg, intraperitoneally). Mitochondria-enriched and soluble fractions were prepared by differential centrifugation from the homogenized fresh heart. Proteins from both fractions were separated by electrophoresis followed by Western blotting. Changes in expression and oligomerization status of Bak and Bax were analyzed at mentioned time-intervals.

Results: In the mitochondria rich fraction an increase in Bax and Bak proteins expression was observed at 12-24-h post-treatment. The greatest monomeric expression of Bak occurred at 12 h. Bax-monomer was expressed at 24-h at levels double than control. In soluble fraction, Bax was detected at similar levels than controls at 12 and 48 h. Dimeric forms for Bax and Bak were detected in the mitochondria. Dimeric-Bax was overexpressed at double levels of controls at 12 h. Dimeric–Bak was expressed similarly in control and treated groups at 4 and 12 hours, with levels decreasing to half at 24 and 48 h.

Conclusions: During apoptosis induction by ouabain, Bak and Bax undergo changes in its expression levels, in mitochondrial and soluble fractions. Presence of dimeric-Bax in the mitochondrial fraction mostly at 12 h suggests that activation of this proapoptotic protein occurs at this time interval.