Mechanism of B16-F10 melanoma cells death induced by *Bothrops leucurus* venom lectin: correlation with mitochondrial permeability transition

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Melanoma is a highly aggressive cancer with poor prognosis in metastatic stage. Mitochondrial permeability transition (MPT) is characterized by a nonspecific Ca²⁺-dependent pore opening sensitive to cyclosporin A that may lead to cell death either by necrosis or apoptosis. *Bothrops leucurus* venom lectin, BlL, is a Ca²⁺-dependent protein (30 kDa) composed by disulfide-linked dimers of 15 kDa with antibacterial activity. The aim of this study is to elucidate the BlL effect on cells and isolated mitochondria. The viability and proliferation of B16-F10 and GN-13 cell lines (murine melanoma and human fibroblast, respectively) treated with BlL (25-100 μg/mL) were evaluated and apoptotic cells was determined by flow cytometry. Rat liver mitochondria were isolated by differential centrifugation and oxygen consumption was measured by a Clark-type electrode in a standard reaction medium containing 200 μM EGTA and 300 μM ADP. ΔΨm was estimated by safranine O fluorescence. The results show that while BlL was cytotoxic against B16-F10 cells (IC⁵₀ value of 75μg/mL) no significant effects were observed in GN-13. B16-F10 cells death occur mainly by necrosis. After mitochondria incubation with BlL (10 μg/mL) the resting respiration increased by 57.1% while respiratory control and ADP/O ratio decreased by 40.5% and 8%, respectively. BlL significantly decreased ΔΨm and stimulated PTP opening. Overall, our data suggest a relationship between BlL-mediated cell death and mitochondrial membrane permeability transition.

Keywords: *Bothrops leucurus* lectin, melanoma, cytotoxicity, mitochondrial permeability transition

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