Induction of Apoptosis from Exposure to Aristolochic acid

Micheli Sossai Spadeto and Victor Ventura de Souza*
Department of Biology, Federal University of Espirito Santo–Center for Natural, Exact and Health Sciences, Brazil

Abstract
The genus Aristolochia gathers plants used in folk medicine, which present in their composition the Aristolochic acids I and II. Among the major toxicological events caused by Aristolochic acids is programmed cell death. Aristolochic acid induced apoptosis occurs in target organs such as the liver, kidney, pancreas, testis, ovary, intestine and lung. Although it is beneficial for the cure of various diseases Aristolochias or Aristolochic acids I and II can also cause necrosis. The objective of this work was to compile information about the effects of Aristolochic acids on the induction of apoptosis. The present work makes an alert when consuming this herb, and derivatives that possess Aristolochic acids.

Keywords: Aristolochia • Cell death • Gene synthesis • Caspase 3

Introduction
The Aristolochic acids (AAs) I and II are present in all plants of the family Aristolochiaceae, commonly used to aid in weight loss and the cure of rheumatism, arthritis, arthrosis, reduction bacterial infection and of malarial fever. [1] On the other hand, the AAs are considered apoptosis inducers mainly in renal cells, the target organ of AAs [2]. Apoptosis is a physiological event that occurs naturally in all organisms. However, when it is grossly induced, it causes irreversible damage. The cells that are entering into apoptosis have a characteristic morphology with condensation of the genetic material followed by fragmentation of the nucleus [3,4] (Figure 1).

Case summary
The process of renal lesion occurs in 4 stages: [5] The first step is the loss of cell adhesion, releasing the cells from the cytoskeleton; The vimentin, an important component of the cytoskeleton, is no longer expressed; Subsequently, cell membrane disruption occurs and, finally, Migration of cells. These four events cause oxidative stress inducing apoptosis.

Apoptosis induced by AAs
AAs and their derivatives are extremely toxic to LLC–PK1 cells, inducing injuries, which stimulate the synthesis of Caspase 3 and 7, leading to apoptosis, regardless of dosage. AAs strongly interfere with gene expression, stimulating the expression of genes related to apoptosis, cell cycle regulation, nuclear organization and mitochondrial transport. Consequently, apoptosis inducing proteins such as BAX (BLC–2–associated protein), BCL2L2, BNIP3, CASP3, CCL2, NAIP (protein inhibitor), PRKCA (protein kinase C), TNFRSF1B, TNFRSF21 and TNFSF6 are expressed. AAs can also induce apoptosis from mitochondrial stress, causing loss of membrane permeability, thus releasing cytochrome C. Apoptosis was verified by different pathways: activation of ERK 1/2, synthesis and activation of caspase 3, increased synthesis of Bax protein (pro–apoptotic protein) and decline in Bcl–2 (anti–apoptotic synthesis), Apoptotic control. On the other hand it was observed that the synthesis of the Bcl–2 protein was reduced drastically while the Bax protein had its synthesis attenuated. This decrease in protein synthesis can be attributed to the presence of AAs. The AAI induces apoptosis via ERK 1/2 in addition to the activation of the p38 protein, thus concluding that AAI–induced apoptosis is correlated with loss of glutathione (GSH).

Calcium and activation of apoptosis
Mitochondria were isolated from HK–2 renal cells in order to ascertain the potential of AAI in mitochondrial membrane permeability. The AAI at the 50 μm dosage along with 20 μm Ca²⁺ for 24 h resulted in mitochondrial swelling, Ca²⁺ extraplation, membrane depolarization and cytochrome C outflow, decreased ATP production and high activity of caspase 3, pro–apoptotic protein. Other authors using the same methodology and cell line, but with 100μm of AAs induced stress and increased concentration of intracellular Ca²⁺ and changes in mitochondria. Thus, they concluded that the toxicological and cytotoxic effect of AAs is potentiated when the concentration of Ca²⁺ increases. Corroborating with this information, an experiment with HK–2 renal cells showed that Ca²⁺ potentiates the effect of AAI on mitochondria. In addition, it has been shown that AAs also cause oxidative stress in the endoplasmic reticulum releasing calcium into the intracellular environment, so calcium accumulation leads to mitochondrial stress and consequently apoptosis.

Conclusion
AAs are potent inducers of apoptosis, leading to cell death on a large scale. We do not recommend the use of any herb of the family Aristolochiaceae and no product containing the AAs.

References
2. Pozdzik AA, Salmon IJ and Debelle FD. *Aristolochic acid induces...
proximal tubule apoptosis and epithelial to mesenchymal transformation. 


How to cite this article: Micheli Sossai Spadeto and Victor Ventura de Souza. "Induction of Apoptosis from Exposure to Aristolochic acid." J Cytol Histol 12 (2021): 577.