

Thapsigargin induces non-apoptotic programmed cell death in RBL-1 cells

Dr. Philip Steiner¹, Mr. Hubert Kerschbaum², Mrs. Ancuela Andosch^{1,2}, Mr. Korollus Melek³, Mrs. Susanna Zierler^{1,4}

¹Institute of Pharmacology, Johannes Kepler University, Linz, Austria, ²Department of Biosciences and Medical Biology, Paris Lodron University, Austria, ³Institute of Biochemistry and Molecular Medicine, University Bern, Switzerland, ⁴Walther Straub Institute of Pharmacology, Ludwig Maximilians University, Munich, Germany

Background and aims:

Thapsigargin (TG) is a potent sarco/endoplasmic reticulum (ER) Ca²⁺-ATPase (SERCA) inhibitor and, accordingly, elevates intracellular Ca²⁺ levels. Recent studies demonstrate that TG induces cell death in a variety of cancer cells. While it is assumed that TG triggers cell death by apoptosis, our cytological studies on rat basophilic leukemia cells (RBL-1) indicate a non-apoptotic mode of cell death.

Methods:

Since the ultrastructural hallmarks of TG-induced cell death are only sparsely described, we studied the morphological consequences of TG exposure in RBL-1 cells using 2D transmission electron microscopy (TEM) and 3D TEM tomography in correlation with laser scanning microscopy (LSM). To visualize and quantify the effects on different organelles, live-cell fluorescence markers such as ER-tracker, HOECHST, endocytosis-tracker and MitoTracker were used.

Results:

TG-exposed RBL-1 cells showed prominent ballooning of the perinuclear space, vacuolization, increased vesicle formation, mitochondrial enlargement and degradation as well as ER-swelling and anomalies. In particular, the TEM data failed to show apoptotic hallmarks such as nuclear fragmentation or the formation of apoptotic bodies in TG-exposed RBL-1 cells.

Conclusion:

Contrary to the prevailing theory that TG triggers apoptosis, our results suggest a non-apoptotic programmed cell death. Moreover, numerous non-apoptotic morphological hallmarks were found, which are reminiscent of autosis and paraptosis. Thus, while TG represents a potential tumor therapeutic agent, the underlying mechanisms of cell death are still ambiguous and might differ in distinct cell types.

Keywords:

apoptosis, autosis, paraptosis, thapsigargin, cancer