α-Mangostin from Cratoxylum arborescens demonstrates apoptogenesis in MCF-7 with regulation of NF-κB and Hsp70 protein modulation in vitro, and tumor reduction in vivo

ABSTRACT

Cratoxylum arborescens is an equatorial plant belonging to the family Guttiferae. In the current study, α-Mangostin (AM) was isolated and its cell death mechanism was studied. HCS was undertaken to detect the nuclear condensation, mitochondrial membrane potential, cell permeability, and the release of cytochrome c. An investigation for reactive oxygen species formation was conducted using fluorescent analysis. To determine the mechanism of cell death, human apoptosis proteome profiler assay was conducted. In addition, using immunofluorescence and immunoblotting, the levels of Bcl-2-associated X protein (Bax) and B-cell lymphoma (Bcl)-2 proteins were also tested. Caspaces such as 3/7, 8, and 9 were assessed during treatment. Using HCS and Western blot, the contribution of nuclear factor kappa-B (NF-κB) was investigated. AM had showed a selective cytotoxicity toward the cancer cells with no toxicity toward the normal cells even at 30 μg/mL, thereby indicating that AM has the attributes to induce cell death in tumor cells. The treatment of MCF-7 cells with AM prompted apoptosis with cell death-transducing signals. This regulated the mitochondrial membrane potential by down-regulation of Bcl-2 and up-regulation of Bax, thereby causing the release of cytochrome c from the mitochondria into the cytosol. The liberation of cytochrome c activated caspace-9, which, in turn, activated the downstream executioner caspace-3/7 with the cleaved poly (ADP-ribose) polymerase protein, thereby leading to apoptotic alterations. Increase of caspace 8 had showed the involvement of an extrinsic pathway. This type of apoptosis was suggested to occur through both extrinsic and intrinsic pathways and prevention of translocation of NF-κB from the cytoplasm to the nucleus. Our results revealed AM prompt apoptosis of MCF-7 cells through NF-κB, Bax/Bcl-2 and heat shock protein 70 modulation with the contribution of caspaces. Moreover, ingestion of AM at (30 and 60 mg/kg) significantly reduced tumor size in an animal model of breast cancer. Our results suggest that AM is a potentially useful agent for the treatment of breast cancer.

Keyword: α-Mangostin; Apoptosis; Mitochondria; Protein array; Caspace 3/7; NF-κB